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Textbook of

Dental & Oral Histology

with Embryology

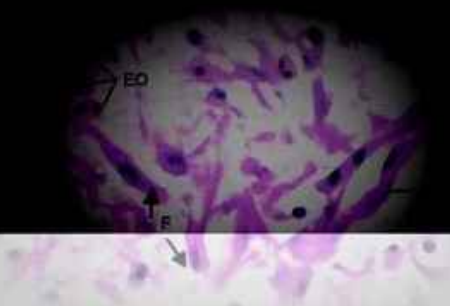
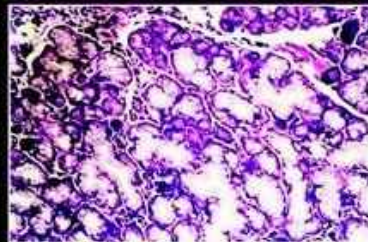
& MCQs

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Mithilesh Chandra
Girish Chandra
Nidhee Chandra**

**2nd
Edition**

Forewords
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JAYPEE



Textbook of
Dental and Oral Histology with Embryology
and Multiple Choice Questions

Textbook of Dental and Oral Histology with Embryology and Multiple Choice Questions

Second Edition

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It is a great pleasure for me to write foreword for *Textbook of Dental and Oral Histology with Embryology and Multiple Choice Questions*. This subject is very important and is the foundation for all dental clinical subjects and research. This book fills the gap in the teaching and research of Dental and Oral Histology with Embryology.

This book is enriched with plenty of very good colored photomicrographs. In addition, labeled figures have also been extensively given which make the subject more appealing and crystal clear. The language of this book is very simple and easy to understand. This book covers all the topics prescribed by the Dental Council of India and Indian and Foreign Universities.

In the end an exhaustive list of all multiple choice questions is given which covers each and every topic of the subject. It makes this book invaluable for students preparing for various competitive examinations.

I very strongly recommend this book to each and every dental student.

Anil Kohli

Foreword

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Hony. Dental Surgeon to the President of India,

President, Indian Association of Public Health Dentistry

I feel pleasure in writing foreword for *Textbook of Dental and Oral Histology with Embryology and Multiple Choice Questions* written by Dr Satish Chandra. This is an important subject and is the foundation for all dental clinical subjects.

The book written in a simple language covers topics of the syllabi laid down by the Dental Council of India and is enriched with color photographs and labeled figures which is an added attraction. Inclusion of exhaustive list of multiple choice questions makes it more useful for dental students.

RK Bali

Foreword

It is with great pleasure I am writing the foreword for the second edition of *Textbook of Dental and Oral Histology with Embryology and MCQs*. The first edition was very well received by the students and teachers. All the shortcomings of the first edition have been removed in the second edition. This book is an excellent contribution to dental literature. Authors have updated and included relevant topics in this edition. The multicolored and very well labeled figures make the subject very easy and clear for the students to assimilate.

The second edition is more comprehensive and will definitely help the students as it includes new topics and figures. The authors have to be congratulated for further improving on their innovative excellent format. This book can be regarded as a textbook-cum-atlas and I am glad that it is provided at an affordable cost.

The special feature of the book is the multiple choice questions, which are very useful especially for the students preparing for various *competitive examinations*.

I have observed that the book has completely covered the syllabi proposed by the Dental Council of India and followed by all the Universities. Overall this is an excellent book. The book is very strongly recommended to all BDS and MDS students, teachers, researchers and the practitioners.

I wish the authors and this book all success.



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We acknowledge with thanks the efforts of Dr AK Jain, Assistant Director, Institute of Pathology, New Delhi, for providing the electronmicrographs for our book. Our thanks are also due to Dr Mohit Chandra, for his help and Shri Lalbabu, technician for preparation of excellent sections.

The undertaking of this book would have been impossible without the sacrifices made by our spouses and children. They have been a constant source of inspiration to us. To them, a special gratitude is offered for their patience and support.

We bow in gratitude to the Almighty God for His blessings.

Finally we acknowledge our sincere thanks to M/s Jaypee Brothers Medical Publishers (P) Ltd., New Delhi and Shri Jitendar P Vij (Chairman and Managing Director) and Mr Tarun Duneja (Director Publishing) for their acceptance and endeavor to bring out this text in an excellent book form.

Preface to the Second Edition

It is with immense pleasure and satisfaction that we present the second edition of book for use by the students, researchers and practitioners in dental sciences. Thorough knowledge of basic sciences is very essential for a more rationale foundation of clinical procedures.

The book has been designed in such a way that the students, teachers, research scholars and practitioners will get full and clear mental picture of the subject. To achieve this high quality colored and labeled photomicrographs have been given along with elaborate well-labeled schematic diagrams.

Almost all the figures have been improved and have been made multicolored to make them more clear and easy to understand. A new chapter, 'Introduction to oral and dental tissues' have been added to facilitate easy and clear understanding of the subject matter by the beginners. The chapter on development and growth has been enlarged to include the development of palate, maxilla, mandible and tongue in detail.

The importance of molecular biological aspects including gene therapy and tissue engineering which regulate the structure, functions, healing and rebuilding the oral and dental tissues have been included. These will be very useful for future managements of oral and dental diseases. The subject matter has been made very clear, by easy and lucid language and fully-labeled colored figures. At a glance all figures make subject matter very clear.

There is an increasing trend for giving MCQs in the regular university examination question papers of BDS course. In some of the universities it has been made compulsory to include a part in the question paper on MCQs. For the benefit of the students of such universities and for the students appearing for various competitive examinations more than 450 multiple choice questions with answers on every topic have been included. Plenty of MCQs on recent advances have been added. For the benefit of the clinicians, clinical considerations have been added at the end of the chapters.

It is hoped that this book will be extremely useful for undergraduate and postgraduate students, teachers, researchers and clinicians. Our aim has been to provide a high quality book-cum-atlas on the subject.

Your suggestions for further improvements are most welcome.

Authors

Preface to the First Edition

It is with immense pleasure that we present this *Textbook of Dental and Oral Histology with Embryology and Multiple Choice Questions* for use by the students, researchers and practitioners in dental sciences. Dental and oral histology with embryology is very important basic science and lays down the foundation for all clinical subjects and paraclinical subjects.

Thorough knowledge of basic sciences is very essential for a more rationale foundation of clinical procedures. A better understanding of basic sciences can make a difference between an inventor-cum-researcher and one with outdated knowledge, between an excellent clinician and another clinician who treats the patients only like a technician, between one who leads and another one who follows. For research on any clinical problem detailed knowledge of basic sciences is a must.

Our aim has been to provide a high quality book on the subject. Need of such a book was being felt for a long time specially by the students, teachers and researchers.

This book has been designed in such a way that the students, teachers, research scholars and practitioners will get full and clear mental picture of the subject. To achieve this high quality color-labeled photomicrographs have been given along with elaborate well-labelled schematic diagrams. Special stains have also been used for best results. The tooth histology has been shown by using high quality photomicrographs of ground sections as well as decalcified sections. Some of the photomicrographs included in the book are extremely rare.

There is an increasing trend for giving MCQs in the regular university examination question papers of BDS course. In some of the universities it has been made compulsory to include a part of the questions papers as MCQs. For the benefit of the students of such universities and for the students appearing for various competitive examinations more than 500 multiple choice questions with answers on every topic have been included in the book. For the benefit of the clinicians, clinical considerations have been added at the end of the chapters.

The subject matter has been made very clear, by easy and lucid language and very well-labeled colored figures. At a glance all figures make subject matter very clear.

It is hoped that this book will be extremely useful for undergraduate & postgraduate students, teachers, researchers and clinicians.

Authors

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Introduction to Oral and Dental Tissues

- The tooth
 - Enamel
 - Dentin
 - Pulp
 - Cementum
- Periodontal ligament (PDL)
- Oral mucous membrane
- Bone and alveolus
- Salivary glands
- Eruption of teeth
- Shedding of deciduous teeth
- Temporomandibular joint

In this chapter a brief introduction of the basics of the subject and the important chapters is given to facilitate the students to easily and clearly understand the subject matter.

THE TOOTH

Approximately twenty percent of the surface area of the oral cavity are constituted by teeth. Maxillary teeth constitute about 11 percent and mandibular teeth constitute about 9 percent surface area. The tooth has two parts, crown and root, divided by a cervical line. Usually crown is visible in oral cavity and root remain inside the gums and jaw bone and is not visible. The part of the tooth visible in oral cavity is the clinical crown usually called as crown. In healthy conditions in young adults anatomical crown i.e. up to only cervical line is visible in oral cavity and anatomical crown (crown up to cervical line) is equal to the clinical crown (visible part of crown in oral cavity).

Anatomical crown is covered by enamel (hardest and most mineralized tissue of the body). The anatomical root is covered by the cementum. The anatomical crown and root is divided by a thin line called cervical line. In all further descriptions, unless specified otherwise the crown means anatomical crown and the root means anatomical root.

The supporting tissues of the tooth surrounding the roots constitute periodontium which consist of cementum, periodontal ligament and the alveolar bone. The tooth is suspended in the socket of alveolar bone by the fibers of periodontal ligament. These fibers act as shock absorber. The inner most part of the tooth in the crown and root is hollow and is called pulp cavity which is filled with most vital part of tooth called pulp. Pulp in crown and root is covered by the dentin. The dentin of the crown is covered by enamel and of the root is covered by cementum (Fig. 1.1).

In childhood the face and jaws are smaller hence fewer teeth of smaller size are present. These smaller teeth are called deciduous or primary teeth. With the growth, the jaw bones grow in size but the teeth once fully formed do not grow in size. Therefore to have the larger sized teeth the deciduous teeth which are of smaller size have to be shed and permanent teeth of larger size erupt in oral cavity replacing the smaller deciduous teeth. Permanent teeth are larger in size and more in number to bear the increased work load of the adulthood.

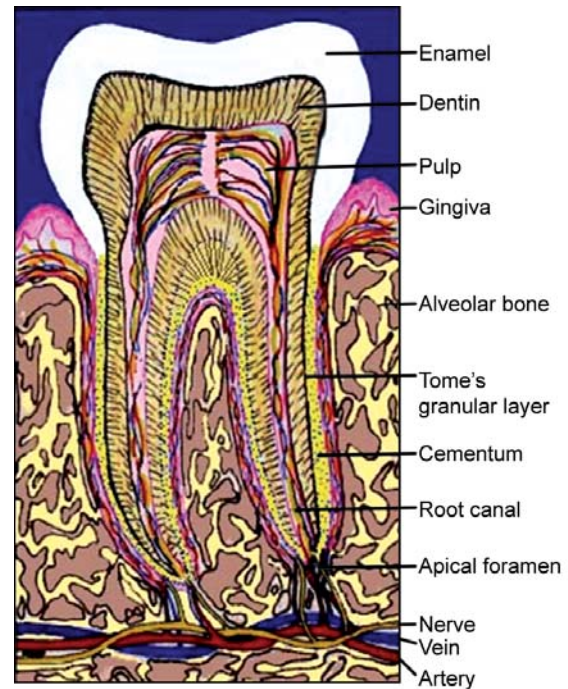


Figure 1.1: Mesiodistal longitudinal section of the mandibular molar tooth and supporting structures. In the pulp chamber, pulp is shown at two different levels. In the right half is shown the central region of the pulp showing thicker blood vessels and in the left half is seen the peripheral pulp showing thinner blood capillaries

The name of the cells ending with blast do generate the tissues, like ameloblast (generate enamel) osteoblast (generate bone) odontoblast (generate dentin) and cementoblast (generate cementum). Likewise the words ending with clast do resorb the tissue like osteoclast and odontoclast etc. Odontoblast and odontoclast are also known as dentinoblast and dentinoclast respectively.

Enamel

The enamel is brittle in nature and may fracture under masticatory load. Enamel is the only ectodermal derivative in the tooth. Enamel consist of 96 percent of inorganic material in the form of apatite crystals. Enamel is formed by the cells called ameloblasts which disappear once enamel is completely formed, hence when enamel is destroyed it cannot be regenerated. Enamel is insensitive, nonvital, selectively permeable and ionic exchange can occur between the enamel and the environment of the oral cavity especially saliva.

To prevent dental caries fluoride is topically applied to the surface of the enamel. By this hydroxyapatite of the enamel is converted to fluorapatite which is more resistant to the dissolution in acid and thus prevent dental caries.

Dentin

The bulk of the tooth is formed by dentin. Dentin is slightly resilient yellowish white, sensitive and avascular tissue which is less calcified than enamel. Dentin surrounds the pulp in the pulp chamber. Dentin consists of dentinal tubules. These tubules contain the extension of the odontoblasts made up of cytoplasm, and are called odontoblastic processes. Present around the tubules is the calcified matrix called intertubular matrix. The walls of the tubules are more calcified than the intertubular matrix.

Enamel must be supported by the resilient dentin to withstand the masticatory forces. Dentin supports the enamel to prevent fracture of enamel under masticatory forces. The specialized cells which form and maintain the dentin are called odontoblasts. Their bodies remain in the pulp and are aligned along the inner border of dentin, and there they form the peripheral boundary of the pulp. The dentin is capable of repair as odontoblasts deposit rapidly more dentin as and when required and also slowly and regularly throughout the life. The dentinoenamel junction is scalloped to create a mechanical retention. The dentin and pulp act as one unit. Dentin protects the pulp and the pulp nourishes the dentin.

Pulp

The central hollow part of the tooth contains the soft and most vital and vascular part of the tooth which is called pulp. Pulp is lost in dried ground sections, leaving an empty pulp chamber. Developmentally and functionally dentin and pulp are alike and may be considered simultaneously. All the functions of the pulp are related to the dentin. Functions of the pulp are as follows.

- A. Formative (produces dentin)
- B. Nutritive (nourishes dentin)
- C. Protective (protect dentin from damage by providing sensitivity to dentin)
- D. Reparative (pulp produces dentin for repair as and when required). The pulp is connected through periapical foramen with periodontal ligament and alveolar bone.

Cementum

Cementum is a mineralized connective tissue. It is bone like in structure covering the root of the teeth. Like dentin

cementum is also continuously formed to compensate for occlusal wear and to keep the tooth in occlusion. Cementum is also avascular and non-innervated. Cementum is formed by cementoblasts. Cementum is firmly interlocked with the dentin of the root. Cementum contains fifty percent inorganic material like apatite crystals and rest fifty percent is organic matrix mainly collagen.

The cementum is of two types acellular (primary) and cellular (secondary). Acellular cementum is covering the cervical portion of the root. The cellular cementum is covering the apical portion of the root. It is called cellular because in it cementoblasts become entrapped in the lacunae of their own matrix, like osteocytes get entrapped in bone. The cementoblasts which get entrapped in the cementum are called cementocytes. Periodontal fibers on one hand are anchored into the cementum of the tooth and on the other hand into the bundle bone of alveolar bone. In this way the tooth remain suspended in the bony socket.

PERIODONTAL LIGAMENT (PDL)

The PDL is a very specialized connective tissue. Its width is about 0.2 mm. It is made up of fibers which on one hand are embedded in the cementum and on the other hand are embedded in the alveolar bone. In this way they connect tooth to the bone. PDL fibers are made up of collagen and act as shock absorber. The PDL also has sensory function. When teeth of opposing arch as soon as touch each other, through proprioceptive fibers in PDL the sensation is perceived. When the opposing teeth strike with heavy force the sensation of pain is perceived by PDL.

ORAL MUCOUS MEMBRANE

The oral cavity is lined by a specialized mucosa which is well adapted to perform its functions.

It consists of two layers, an epithelium which is superficial and connective tissue (lamina propria) which is deeper. Functions of the oral mucosa are (a) lining (b) protecting and (c) taste. Histologically oral mucosa is of following three types (a) masticatory mucosa (b) lining mucosa and (c) specialized mucosa.

The masticatory mucosa covers the gingiva and hard palate. It is tightly attached to the underlying bone by the lamina propria. Its covering epithelium is keratinized

so as to bear the forces of food bolus during mastication without damage. The lining mucosa is flexible and nonkeratinized. The lamina propria is loosely bound to the underlying structures. The dorsal surface of the tongue is covered by specialized mucosa which contains papillae and taste buds.

For their eruption the teeth perforate the oral mucosa. The mucosa immediately surrounding the erupted tooth is called gingiva. Gingiva is absent before eruption and disappears after loss of tooth.

BONE AND ALVEOLUS

The teeth are attached to the alveolar processes of the jaw bone by the PDL. When the teeth are lost the alveolar process are also gradually lost. Alveolar processes of the jaws form and support the sockets of the teeth. The orthodontic treatment is made possible by this property of the bone, to form under tension and resorb under pressure. This property is not present in cementum.

SALIVARY GLANDS

There are three pairs of major salivary glands, the parotid, submandibular and sublingual. The minor salivary glands are numerous and are scattered throughout the oral cavity except in gingiva and anterior part of hard palate. The basic histological structure of major salivary glands are similar. The salivary gland is like a bunch of grapes.

Saliva is a complex fluid. Normally exposed portion of each tooth is continuously bathed with saliva.

ERUPTION OF TEETH

The jaws of infants are smaller and can accommodate only smaller and lesser number of teeth which are only sufficient for soft and limited diet, required and taken in infancy. Adults take harder food and more in quantity, for them stronger, larger and more number of teeth are required. Teeth do not grow hence the deciduous (primary) teeth of infancy shed and in their place permanent teeth erupt in growing jaws. In adulthood jaws become grown up to full size and stronger. Hence to meet the requirements human being have two dentitions.

SHEDDING OF DECIDUOUS TEETH

Shedding of deciduous teeth is the physiological process by which deciduous teeth are removed to create space for the permanent teeth. For shedding the roots of the deciduous teeth are resorbed. The successional tooth develop lingually and erupt in an occlusal and vestibular direction. The developing tooth occupy a position directly apical to the shedding deciduous tooth and exerts pressure on the root. This pressure causes resorption of root resulting in exfoliation of the deciduous tooth.

TEMPOROMANDIBULAR JOINT (TMJ)

TMJ is formed by the articulation of lower jaw with the cranium and upper facial skeleton. TMJ is a synovial, bicondylar diarthrodial joint. It shows both side to side sliding and hinge movements. During mastication the masticatory muscles move the mandible in opening and closing direction and side to side direction. TMJ functions in speech, mastication and deglutition.

General Embryology and Growth & Development of Oromaxillofacial Structures

- Introduction—The cell
- Growth and development
- Fertilization and cleavage
- Formation of germ layers
- Origin of facial tissues
- Branchial arches: Formation and derivatives
- Development of facial prominences
- Development of palate
 - Development of primary palate
 - Development of secondary palate
- The process of fusion of nasal septum and palatine processes
- Development of tongue
 - Muscles of tongue
- Growth of face and jaws
 - Development of maxilla and mandible
 - Common developmental features between both the jaws
 - Growth of cranium
- Clinical considerations
 - Facial clefts
 - Fissural (inclusion, developmental) cysts
 - Developmental anomalies of tongue
 - Syndrome: Treacher Collin's syndrome
 - Hemifacial microsomia

INTRODUCTION—THE CELL

The human body is composed of cells, intercellular substance and fluid in which the tissues are bathed. The cell is the smallest living unit of the body capable of independent existence. The cell is composed of nucleus and cytoplasm. The nucleus contains the fundamental structures of life, which are deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). Various cell organelles in cytoplasm perform other essential functions (Figure 2.1). Cells vary in size, shape, structure and function. They carry out the vital processes of absorption, assimilation, respiration, conduction, growth, reproduction and excretion.

The intercellular substance is the product of these cells. It surrounds the cells and provides nutrition to them, takes up waste products and provides shape to the body. It may be as soft as loose connective tissue and may be as hard as bone, cartilage or cementum of teeth.

The third component, fluid, contains blood and lymph, and circulates in vessels throughout the body. It also includes the tissue fluid which surrounds each cells of the body. The life of all the cells is limited. White blood cells (leukocyte) have a life span of only a few hours to a few days. Red blood cells have a life span of one hundred and twenty days. The surface-covering cells are replaced frequently. The important structures of a typical cell are as follows.

Cell Nucleus

Barring a few exceptions like mature red blood cells, a nucleus is present in all the cells of the body. Depending on the shape of the cell, usually it is round to ovoid. The number of nuclei in a cell may vary. The nucleus contains chromosomes. Deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) are produced in the nucleus. DNA contains genetic information in the cell and the RNA is responsible for carrying information from the DNA to sites of actual protein synthesis, which are located in the cell cytoplasm. Nuclear membrane that surrounds the nucleus is similar to plasma membrane, that is, it consists of two phospholipid layers. The nucleus is responsible for the metabolism, growth and reproduction of the cell (Fig. 2.1).

Cell Cytoplasm

Cytoplasm contains structures essential for creation and absorption of cell products. The cytosol present in cytoplasm contains the organelles and solutes. The cytosol produces energy from raw materials and also excretes waste products. All these functions are also carried out by the endoplasmic reticulum (ER). These are cavities in the cytoplasm bound by a parallel membrane. They contain newly acquired and synthesized proteins. In the same cell, ER, is of two types: (a) smooth surfaced and (b) granular surfaced. Ribosomes are present on the surface

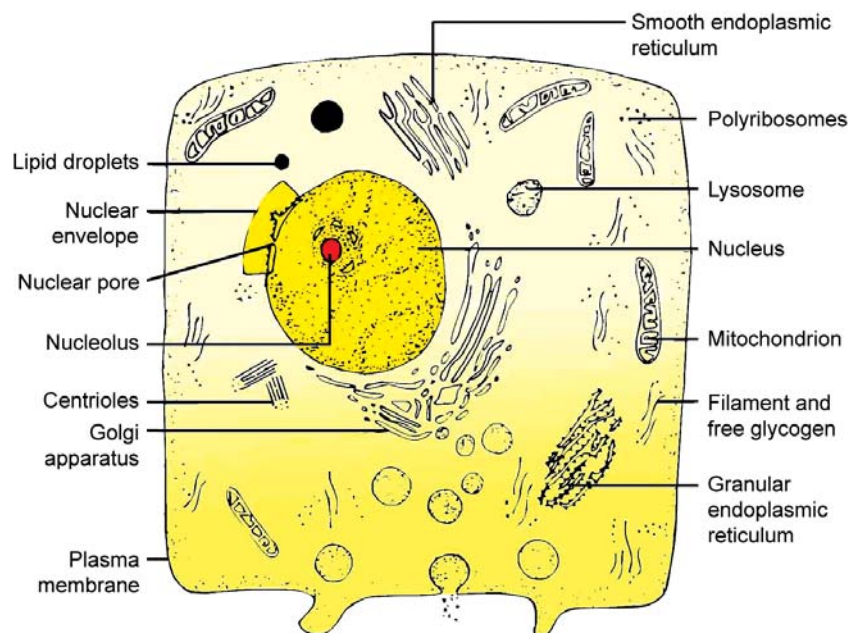


Figure 2.1: Structure of a typical cell

of the granular surfaced ER. At this place, the production of protein is initiated. Proteins are vital for all metabolic processes of the cells. Proteins contain amino acids, which form acids and bases.

Ribosomes

It is a cell organelle made up of ribosomal RNA and protein. Ribosomes may exist singly, or in clusters called polyribosomes, or on the surface of rough endoplasmic reticulum. In protein synthesis, they are the sites of messenger RNA (mRNA) attachment and amino acid assembly in the sequence ordered by the genetic code carried by mRNA. They translate genetic codes for proteins and activate mechanisms for their production. The type of protein produced is dependent on the messenger RNA. The mRNA carries the message directly from the DNA to the nucleus, and then to the RNA in the ER. After getting attached to the ribosomes, this molecule initiates the formation of amino acids.

Lysosomes

Lysosome is a cell organelle that is a part of the intracellular digestive system. Inside its limiting membrane, it contains a number of hydrolytic enzymes capable of breaking down proteins and certain carbohydrates. Lysosomes are small membrane covered bodies. They are present in all cells except the red blood cells and are more active in macrophages and leukocytes. They breakdown substances both inside and outside the cells.

Mitochondria

Mitochondria are membrane-covered organelles present in all cells and lie free in the cytoplasm. In them, many metabolic reactions take place, which generate energy and are a major source of adenosine triphosphate (ATP). They contain the enzymes for the aerobic stages of cell respiration and are thus the site of most ATP synthesis. Mitochondria lie near the area where energy is required.

GROWTH AND DEVELOPMENT

Growth is defined as a normal process in which increase in size or increase in weight of an organism takes place as a result of continuous division and differentiation of various types of tissues. It occurs by the synthesis of new protoplasm and multiplication of cells. Development is

defined as the process of growth and differentiation, or an increase towards maturity or full size.

These two processes are practically inseparable and rely on each other (they function side by side during the formation of the human face and oral cavity). As the tissues begin to differentiate at the age of 4 to 8 weeks in the embryonic period, they are most susceptible to defective development.

FERTILIZATION AND CLEAVAGE

Origin of tissue starts with fertilization of the egg. Fertilization occurs in the fallopian tubes by the union of the female germ cell, ovum (ova) and male germ cell, spermatozoa (sperm). This union after growth produces a zygote (Fig. 2.2). The cleavage of zygote takes place through mitosis gradually producing a ball of cells called morula in the uterine tube. Morula is a solid mass of cells, resembling a mulberry (Figs 2.2 and 2.3). By the end of the first week, morula grows and travels medially to the uterus. The uterine lining that is endometrium thickens and in order to nourish the fertilized ovum, its capillaries and glands develop. If the fertilized ovum does not reach the uterine cavity, the development of capillaries and gland is terminated by menstruation.

Morula increases in size to form blastocyst. Blastocyst later on becomes hollow and develops a small inner cell mass called the embryoblast. The outer layer of cell lining is called the trophoblast.

Embryoblast is formed by one-fourth of the cells of the egg cell mass. Embryoblast forms the embryo proper while the trophoblast forms the placenta. The cavity, which is present in between the inner cell mass and the outer layer of cell, forms the yolk sac (Primary yolk sac).

The commencement of the embryonic period is taken at the beginning of the third week after fertilization. By the end of the fourth week after fertilization, the heart and pericardium of the embryo becomes prominent. At the end of the sixth week after fertilization, the embryo becomes 22 to 24 mm in length. Development during the fetal period consists of growth and maturation of the structures that were formed during the embryonic period. The development in crown to rump (CR) length and to some extent the weight of the fetus may give an idea of some important developmental stages of the embryo (Table 2.1).

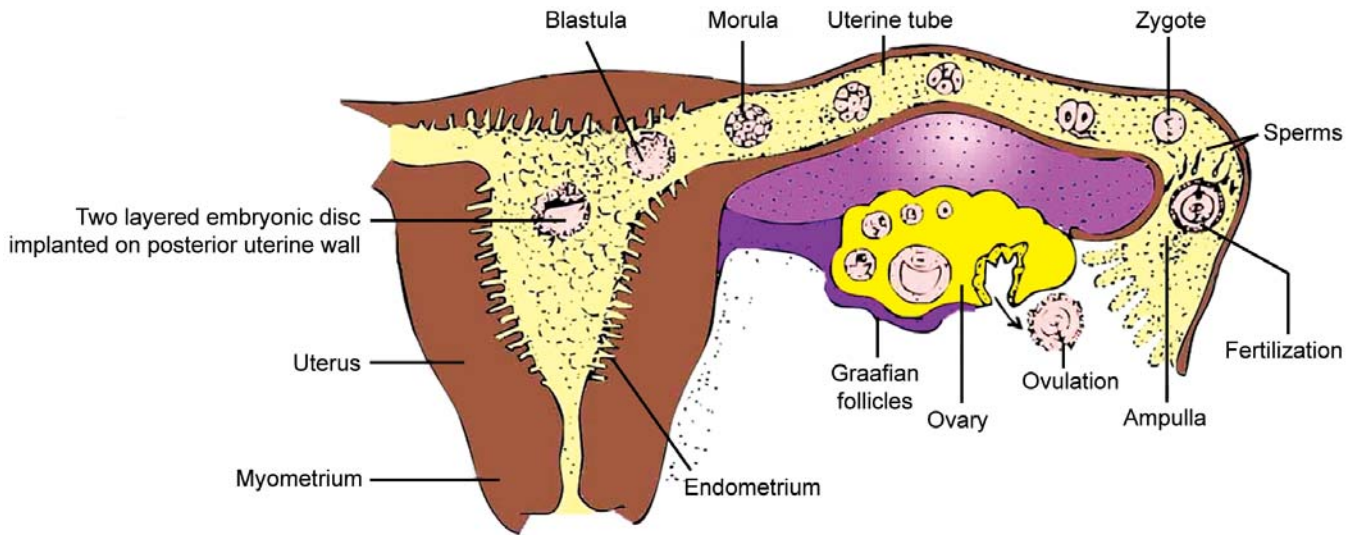


Figure 2.2: Uterus and uterine tubes showing path of sperm to distal tube, where fertilization of ovum occurs. Such produced zygote travels to uterus while undergoing cleavage and gets implanted on seventh day after conception

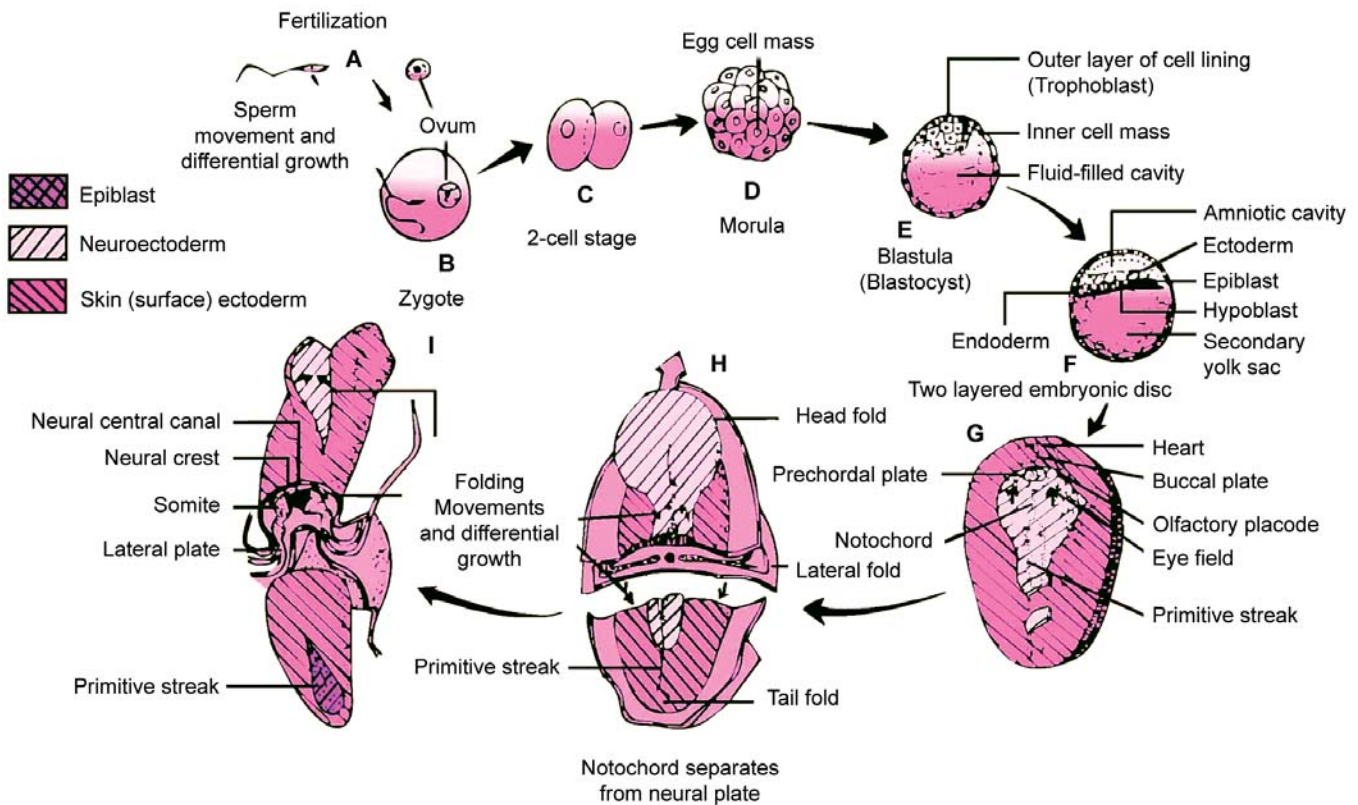


Figure 2.3: Development of embryo through neural tube formation. Small arrows within figures indicate where folding occurs

Table 2.1: Approximate age (in weeks), CR length (in mm) and weight of fetus (in grams)

Age (in weeks) Menstrual cycle	After Fertilization	Crown to Rump Length (CRL) (mm)	Fetal Weight (gms)
3	1	1.8	0.5
4	2	2.4	1.5
6	4	4	3
8	6	22	6
10	8	32	12
12	10	60	14
14	12	88	45

FORMATION OF GERM LAYERS

With the further development of blastocyst, the embryoblast differentiates rapidly to form a few layered germ disc (Figs 2.4 and 2.5A to E). Another small cavity (amniotic cavity) develops on the other side of inner cell mass. Amniotic cavity is lined by ectoderm (columnar cells) while the other sac (secondary yolk sac) is lined by endodermal cells (flattened cells).

Embryonic disc is formed in-between the amniotic cavity and the yolk sac. It consists of the common wall of the two adjacent sacs, that is both ectodermal and endodermal layers are present in the embryonic disk. In humans, the major portion of the egg cell mass forms the extra embryonic membrane.

Ectodermal cells of the embryonic disk on the dorsal surface form the neural plate, the lateral boundaries of which elevate to form the neural tube. Later on, it becomes the brain and the spinal cord.

Endodermal cells form a tube and become the gastrointestinal tract. This tube anteriorly develops pharyngeal pouches, lungs, liver, gallbladder, pancreas and urinary bladder.

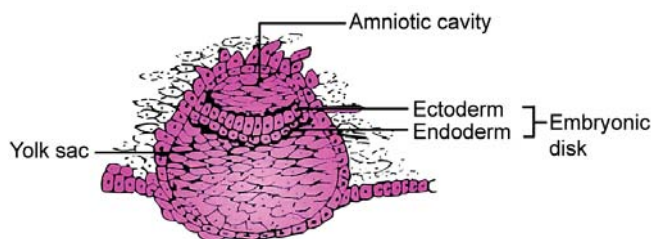


Figure 2.4: Developing ovum with different germ layers and amniotic cavity lined with ectoderm

Mesodermal layer develops between the ectodermal and endodermal layers. Mesodermal cells develop into muscles, skeleton, and blood cells of the embryo. Mesodermal cells are also present along the elongating digestive tube.

Thus all tissues of the body develop from the three layers - ectoderm, endoderm and the mesoderm (Figs 2.5A to E).

The prechordal plate is formed by a small enlargement of the ectodermal and endodermal cells near the margin of the embryo. This helps in differentiating the head and tail ends and the left and right halves of the embryo. The primitive streak, which is an elevation that bulges into the amniotic cavity, appears at the tail end. The primitive node or primitive pit is present at the head end of the primitive streak. Proliferation and migration of ectodermal cells between the ectoderm and endoderm forms a solid column till the prochordal plate. The notochord is formed as a result of canalization of this column; this helps support the embryo (Fig. 2.6).

During the third prenatal week, that is the third week after fertilization, neural folds appear from the lateral edges of the neural plate. These folds reach the midline first in the cervical region and then they close both anteriorly and posteriorly. Anterior tube shows three dilatations, which form the primary brain vesicles - the forebrain, midbrain and the hind brain. Cerebral hemisphere develops from the forebrain, which develops into the frontal, temporal and occipital lobes. Fifth cranial (trigeminal) nerve develops in the midbrain (Fig. 2.7).

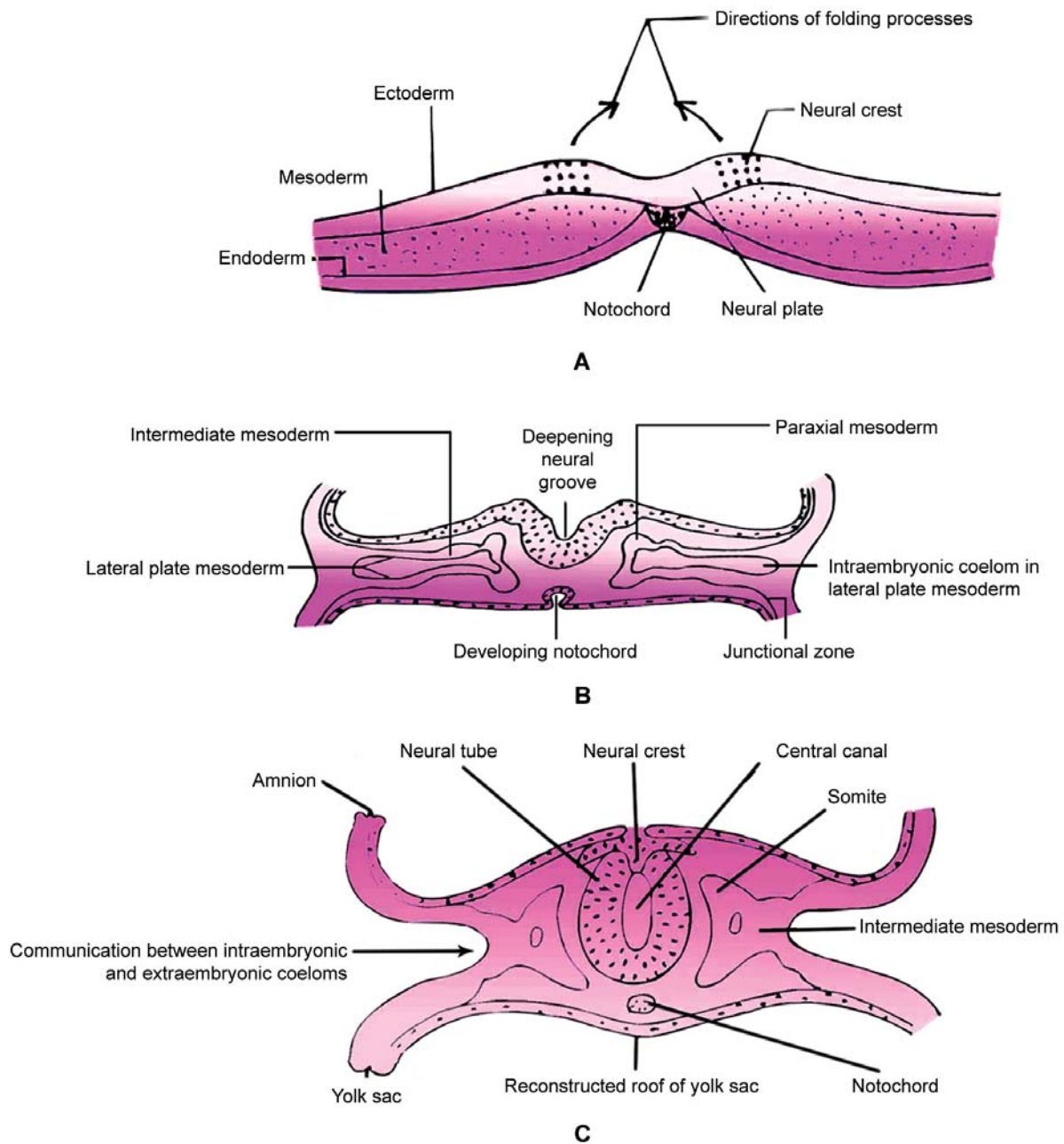
The blood vascular system originates from the angioblasts. These cells arise from the visceral mesoderm of the wall of the yolk sac. This occurs during the third week of prenatal life. The outer cells organize into elongating tubes and the inner cells become blood cells. The vessels begin to develop in the embryo and form a vascular network, which is connected to the placenta. By the fourth week the heart begins to beat. Other mesenchyme cells migrate into the pericardial area, which functions in the development of the heart tube. Cardiac muscle is differentiated later on by these cells. Heart enlarges and with this, growth and development of internal partitions start (Figs 2.8 to 2.10).

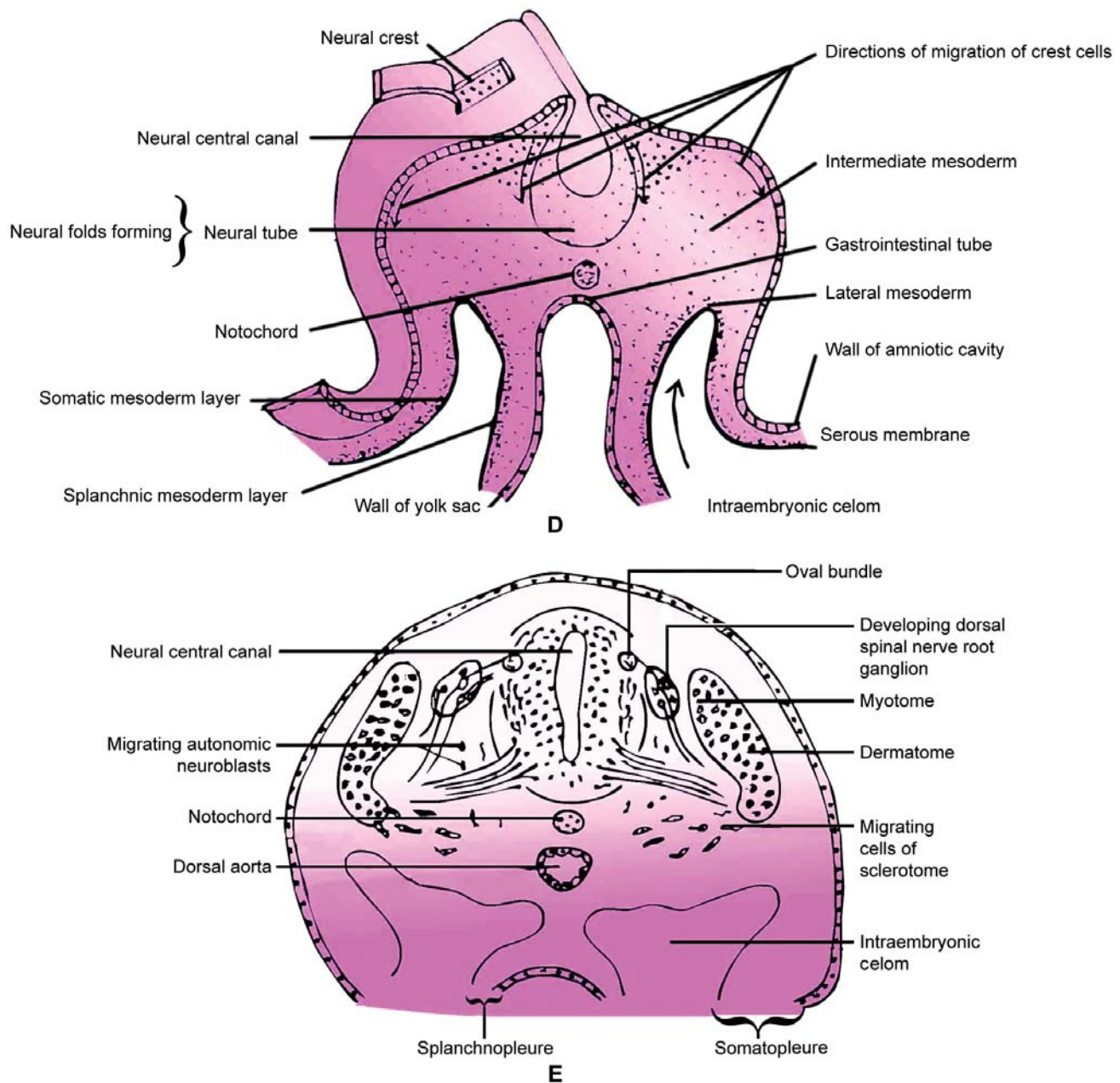
The contributions of the three layers in human embryo to the formation of the different systems and organs is as follows (Fig. 2.11).

Embryonic Ectoderm

Embryonic ectoderm consists of columnar cells which become cubical towards the periphery of the embryonic area. It gives rise to: (i) the enamel of teeth and the salivary glands; (ii) the epithelium lining the nose and paranasal sinuses, the roof of the mouth, the gums and the cheeks; (iii) epidermis and the lining cells of the glands which open on it, and the appendages of the skin, the hair and

the nails; (iv) practically the whole of the nervous system, including the cranial and spinal ganglia, the sympathetic ganglia and the posterior lobe of the hypophysis cerebri; (v) the anterior lobe of the hypophysis cerebri; (vi) the epithelium of the cornea, conjunctiva and lacrimal glands; (vii) the chromaffin organs; (viii) the lens; (ix) the plain muscle of the iris; and (x) the neuroepithelium of the sense organs.





Figures 2.5A to E: A series of schematic diagrams showing important stages in the development and differentiation of the neural plate and gastrointestinal tube, the neural crest, the notochord and the intraembryonic mesoderm and celom; **A to C.** Cross section through three-germ layer embryo showing similar structures in both head and trunk regions; **D and E.** Formation of neural and gastrointestinal tubes. They will separate from embryo surface after completion of fusion

Embryonic Endoderm

Embryonic endoderm consists of flattened cells which subsequently become columnar. It gives origin to: (i) the epithelial lining of the whole of the alimentary canal, with the exception of those portions already ascribed to the

ectoderm; (ii) the lining cells of the glands, which open into the alimentary canal, including the liver and the pancreas, but excluding the salivary glands; (iii) the epithelium lining the auditory tube and tympanic cavity; (iv) the epithelium of the thyroid and parathyroid glands

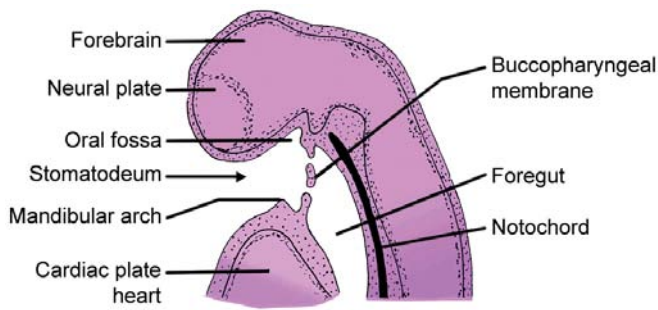


Figure 2.6: Sagittal section through head of four weeks old (3.5 millimeters CR length) embryo

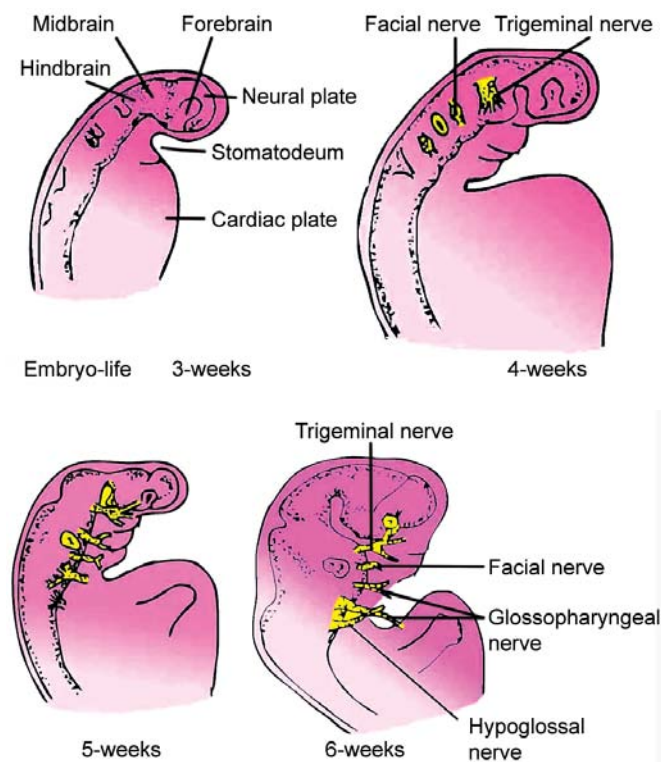


Figure 2.7: Developing cranial nerves

and the thymus; (v) the lining epithelium of the larynx, trachea and smaller air passages, including the alveoli and the air saccules; (vi) the epithelium of most of the urinary bladder and much of the urethra; and (vii) the epithelium of the prostate.

Intraembryonic Mesoderm

Intraembryonic Mesoderm gives origin to the remaining organs and tissues of the body. These include: (i) the teeth

with the exception of the enamel; (ii) all the connective and sclerous tissues; (iii) the whole musculature of the body, both striated and unstriated, with the exception of the musculature of the iris; (iv) the blood and the blood vascular and lymphatic systems; (v) the urogenital system with the exception of most of the urinary bladder, prostate and urethra; and (vi) the cortex of the suprarenal glands and the mesothelial linings of the pericardial, pleural and peritoneal cavities.

ORIGIN OF FACIAL TISSUES

A median strip of mesoderm cells present throughout the length of the embryo forms the neural plate (Fig. 2.6). From the ectoderm, along the margins of this neural plate, a group of cells undergoing extensive migration develop. These are the neural crest cells. They undergo extensive differentiation to give rise to various structures. Those cells migrating to the trunk region form neural, endocrine and pigment cells. Those that migrate to the head and neck form the skeletal and connective tissues like bone, cartilage, dentin and others except the enamel, which is formed by the ectoderm lining the oral cavity. The skin has an epidermis, which develops from the ectoderm and a dermis, which arises from the underlying mesoderm.

BRANCHIAL ARCHES: FORMATION AND DERIVATIVES

Branchial arches or pharyngeal arches are six in number, the fifth one being rudimentary. In the stomatodeum, a continuous process of mesodermal thickening occurs in the fourth week of intrauterine life (Fig. 2.7).

Due to continuous progressive separation of the primitive foregut from the future heart, six cylindrical structures are formed that are called the branchial arches. The ectodermal extension of the branchial arch is called the branchial cleft and the endodermal extension is termed the branchial pouch.

There is no specific name for each branchial arch except the first and second. The first one is called as the Mandibular and the second one is the hyoid arch (Table 2.2).

The nerve fibers from the mandibular division of the trigeminal nerve are derived from the mandibular arch. The seventh, ninth and tenth cranial nerves are derived from the second, third and fourth branchial arches respectively (Fig. 2.12)

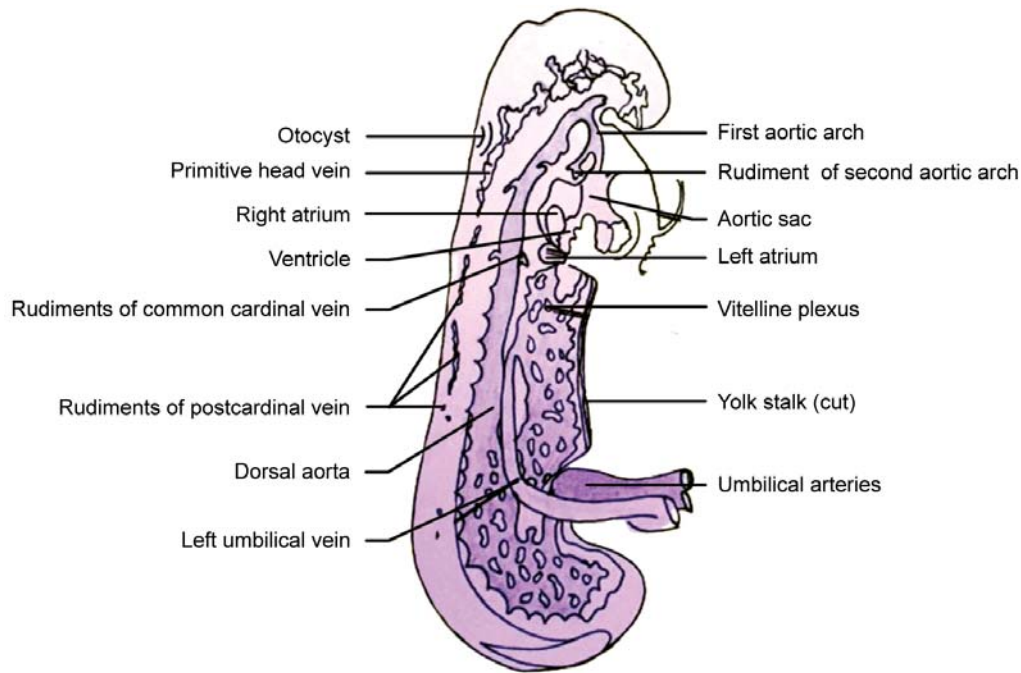


Figure 2.8: The blood vascular system of embryo with 14 paired somites. Age about 14 days, C.R. length 2.4 mm. The arteries and veins are in the process of development, hence no true circulation is possible at this stage, only the endothelial lining of the heart tube is seen

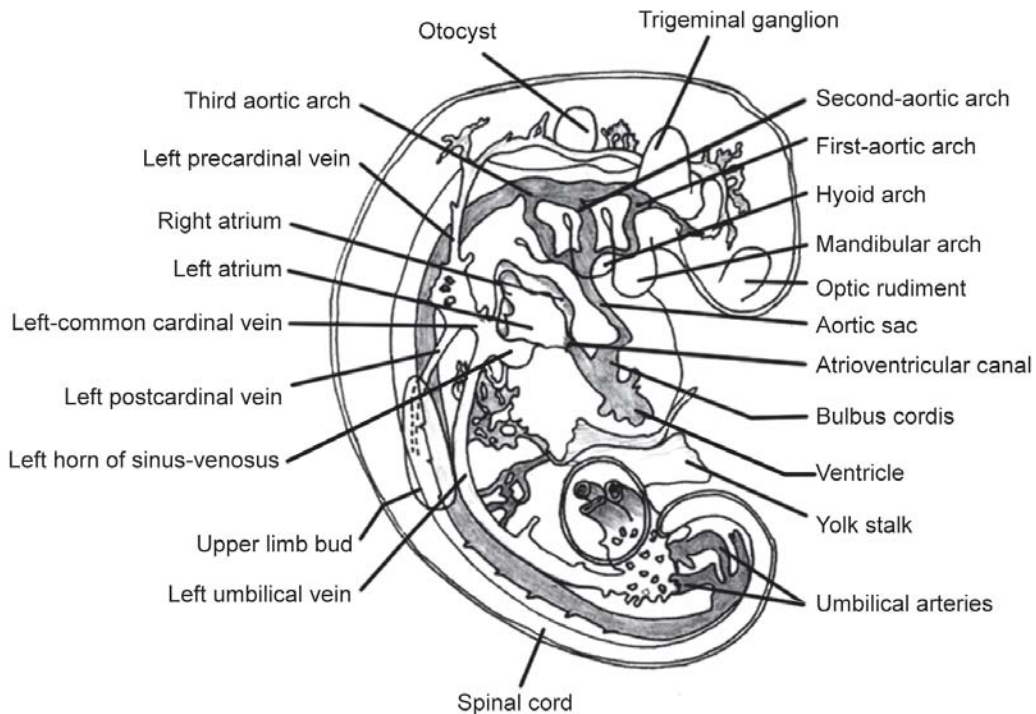


Figure 2.9: Profile reconstruction of the blood vascular system of embryo having twenty eight somites. C.R. length 4 mm, age about twenty six days. The endothelial lining of the heart chambers is shown and as the muscular wall has been omitted, the pericardial cavity appears much larger than the contained heart. The atrioventricular canal still connects the left atrium with the single ventricle

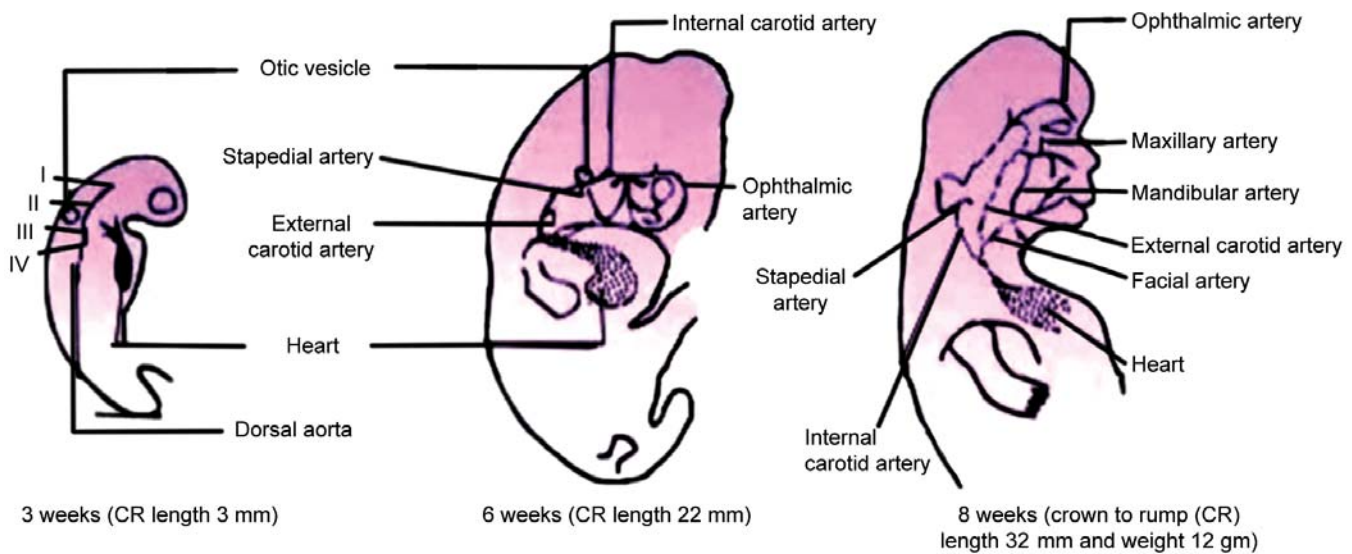


Figure 2.10: Development of arterial system of facial region with its relation to visceral arches (sagittal section)

Table 2.2: Derivatives of branchial arches

Arches	Muscles	Nerves	Skeletons
Mandibular	Anterior belly of digastric Muscles of mastication (masseter, medial pterygoid temporalis, lateral pterygoid and mylohyoid)	Mandibular nerve (A division of trigeminal nerve)	Meckel's cartilage Malleus, Incus (Figure 2.13)
Hyoid	Posterior belly of digastric Muscles of facial expression, Occipito-frontalis	Facial nerve (seventh Cranial N.)	Stapes, Styloid process, Stylohyoid ligament, Superior part of body of hyoid, smaller (lesser) cornu of hyoid (Figure 2.13)
Third	Stylopharyngeus	Glossopharyngeal (Ninth cranial N.)	Greater cornu of Hyoid (Figure 2.13)
Fourth	Muscles of pharynx except Stylopharyngeus, muscles of the palate except tensor palatine, cricothyroid	Superior laryngeal nerve	Thyroid cartilage arytenoids, cuneiform cartilages
Fifth Sixth	Rudimentary and transitory Muscles of larynx except cricothyroid	Rudimentary and Recurrent laryngeal nerve	Rudimentary and transitory Cricoid cartilage

The muscles of mastication and the anterior belly of the digastric are derived from the first arch. The muscles of facial expression and the posterior belly of digastric are derived from the second arch. Myoblasts from the third and fourth arches form the pharynx and soft palate. The connective tissue of the anterior two thirds of tongue arises from the first arch mesenchyme, whereas the

connective tissue of the posterior one third of the tongue arises from the third arch mesenchyme, but the covering of tongue is different. The anterior two-thirds is covered by ectoderm, whereas the posterior one third is covered by endoderm (Fig. 2.13).

The arterial system of facial region and facial skeleton develops gradually with visceral arches (Fig. 2.10).

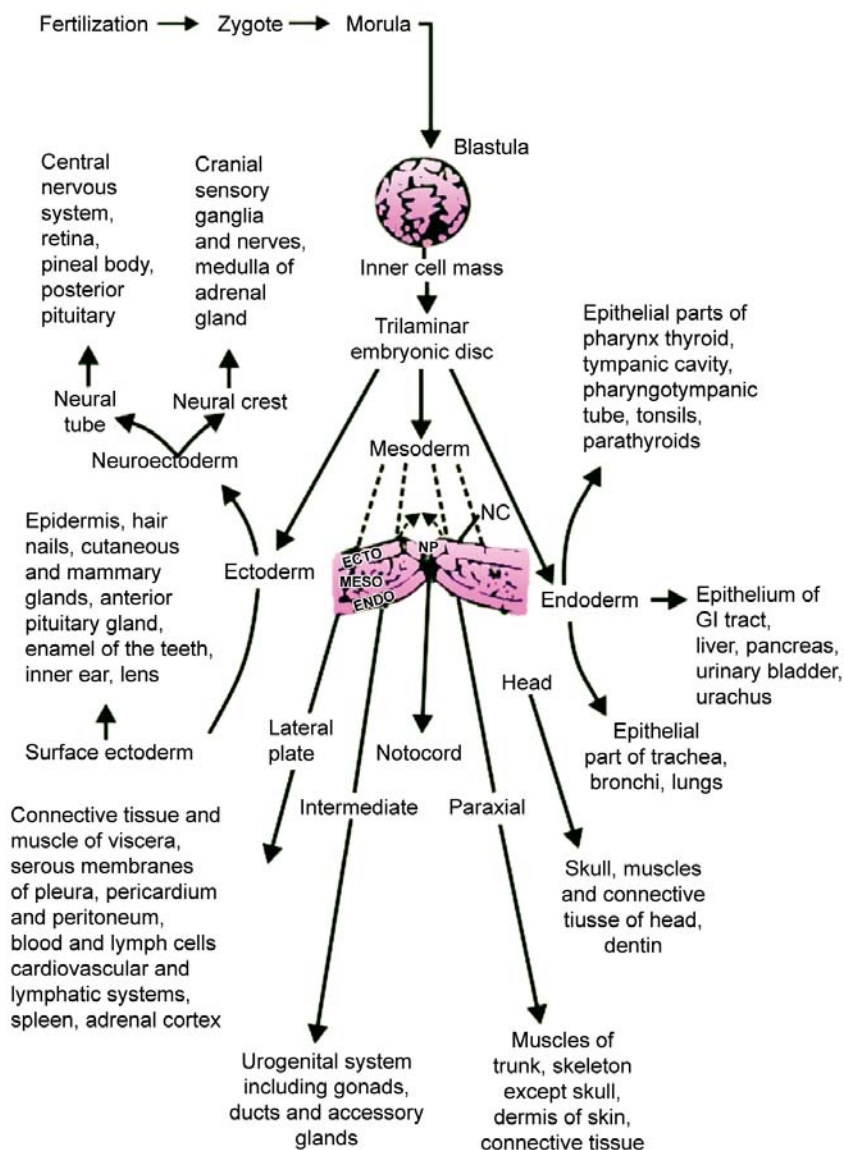


Figure 2.11: Derivatives of embryonic disk consisting of Endoderm (Endo), Mesoderm (Meso), Ectoderm (Ecto), Neural Crest (NC) and Neural Plate (NP). Dotted arrows = directions of folding processes

DEVELOPMENT OF FACIAL PROMINENCES

The branchial arches, which develop in the fourth week of intrauterine life, play very important role in the development of the future head and neck. The tissue surrounding the oral pit develops into the face. Frontal process is present above the oral pit and it covers the brain. Forehead develops from the frontal process.

Frontonasal Process

Frontonasal process develops from the crest cell resulting from the collection of neural crest cells in the future upper face.

Olfactory Placode

Olfactory placode is derived from the neural plate. It is the first structure to develop during the development of

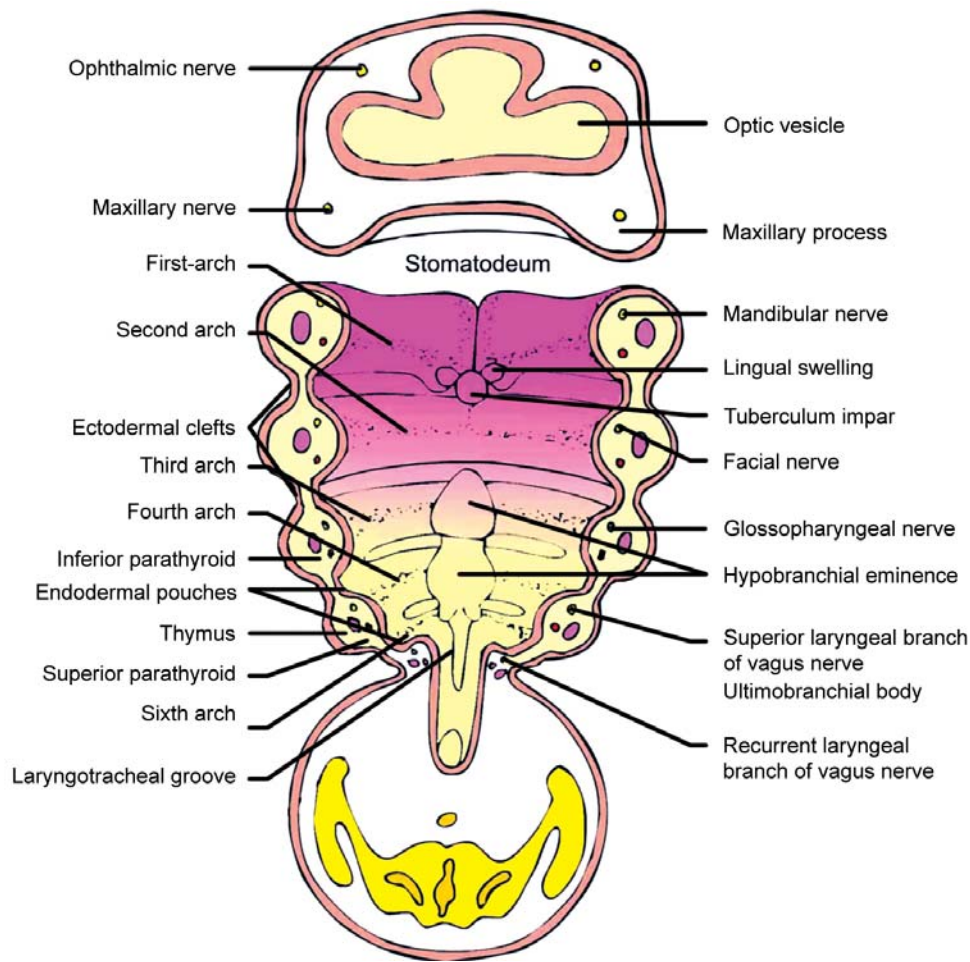


Figure 2.12: Formation of branchial or pharyngeal arches, dorsal aspect of the pharyngeal floor and section of lateral walls

face. Basically placode is a thickened band of ectoderm. The skull grows in all directions (Fig. 2.14).

Maxillary Process

The superior end of the mandibular arch gives a process called the maxillary process present on the lateral side of the oral pit (Fig. 2.14). Cheek develops from the maxillary process. Below the oral pit, the mandibular arch is present, which forms the lower jaw. Second branchial or hyoid arch is present inferior to the mandibular arch. Its muscle contributes to the face. Hyoid arch forms part of the external and the middle ear. Nasal placodes develop into nostrils as the tissue around these grow, resulting in two slit openings around the oral pit. Now the frontal area is known as the frontonasal process. Mouth slit widens to

the point at which maxillary and mandibular tissues merge. The upper lip is now composed of the medial nasal process and two lateral maxillary segments. From the medial nasal process develops the philtrum.

With the development of the lateral nasal prominence, the medial nasal prominence comes in its contact, so that all three processes contribute to the initial separation of the developing oral cavity and the nasal pit. This separation is called as the primary palate (Figs 2.15 and 2.16 A and B).

Nasal Placode

Nasal Placode is superior to the primitive foregut. There is bilateral localized thickening of the ectoderm which collapses readily and forms the nasal pits which are

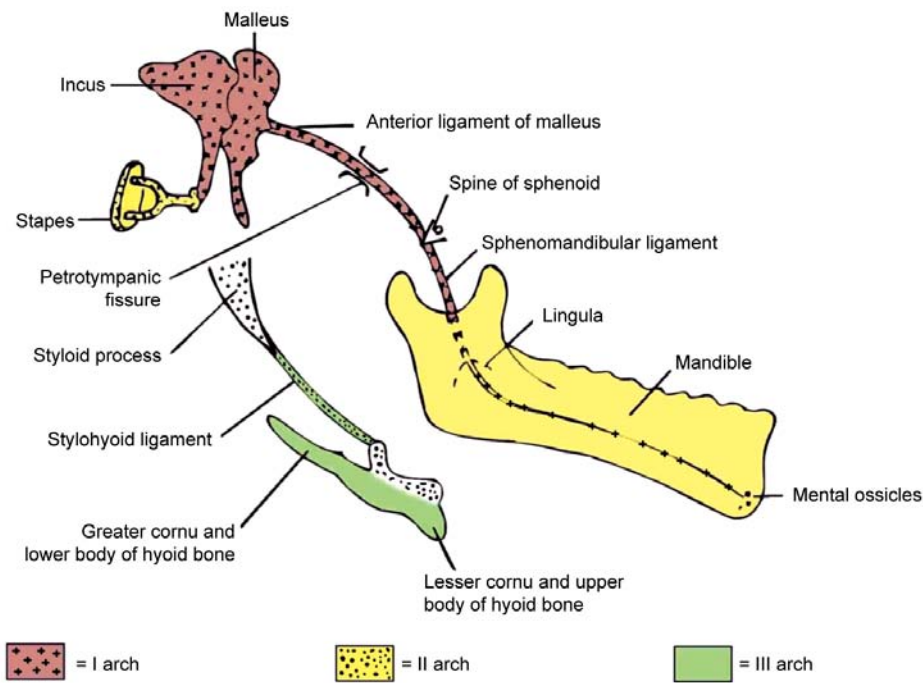


Figure 2.13: Skeletal derivatives (Osseous and Cartilaginous) of the I, II, III branchial arches

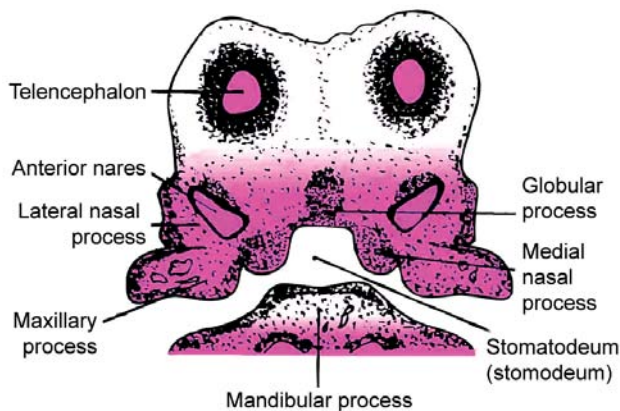


Figure 2.14: Developing processes of oral cavity in a frontal section of 35 days old embryo

converted into nostrils in future. Here frontonasal process is also divides by nasal pit and forms one medial nasal process and two lateral nasal processes. The initial separation of the oral cavity and the nasal pit is by the primary palate. It is formed by fusion of all the three processes (Figs 2.14 to 2.16 A and B).

DEVELOPMENT OF PALATE

Development of Primary Palate

Primary palate develops by ossification in a sheet of mesenchymal tissue superficial to nasal capsule. At about 28 days of intrauterine life olfactory placode in the form of localized thickening develop with ectoderm of the frontal prominence just above the opening of stomatodeum. Around placode the fast proliferations of the underlying mesenchyme forms the frontal eminence and produce a horse shoe shaped elevation (ridge) converting olfactory placode into the nasal depression (pit). The medial arm of elevation is called medial nasal process. The lateral arm of the horse shoe shaped elevation is called the lateral nasal process. The part of the frontal prominence where nose will develop and the above mentioned changes are taking place is called frontonasal process (region). The frontonasal process with medial nasal processes of both the sides develop into the primary palate, middle portion of the nose, middle portion of the upper lip and the anterior part of maxilla. The primary palate is formed from the frontonasal and medial nasal processes.

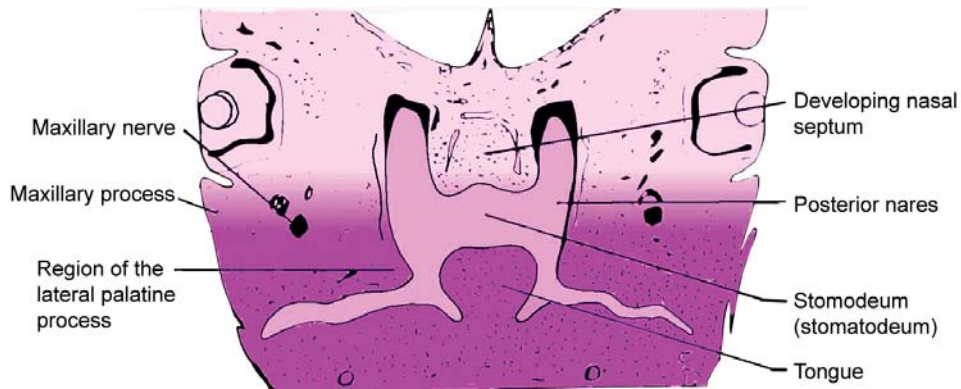


Figure 2.15: Developing anterior primary nasal cavity in a frontal section of thirty-seven days old embryo

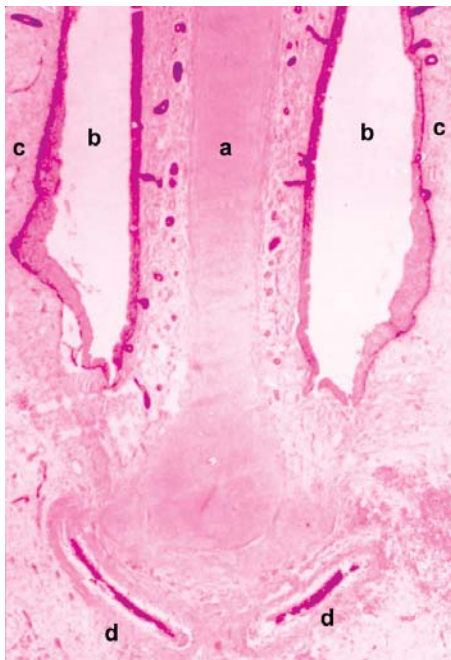


Figure 2.16A: Development of face -(fetus 40 days) -Sagittal section: central (medial) nasal septum (a) with nasal cavities (b) on both sides lined by epithelium x 30. (c) Lateral nasal processes (d) Palatal shelves

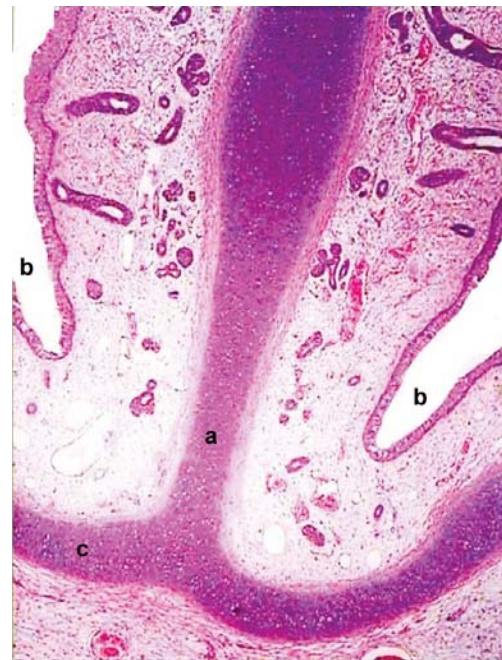


Figure 2.16B: Development of face (fetus six weeks) sagittal section: Central nasal septum (a) with nasal cavities (b) on both sides and palatal shelves (c) x 40

Development of Secondary Palate

At about sixth week of intrauterine life (IUL) a common oronasal cavity is surrounded anteriorly by the primary palate. It is mainly occupied by the developing tongue. Palate develops by ossification in a sheet of mesenchymal tissue superficial to nasal capsule. The distinctions between the oral and nasal cavities appear only by the development of the secondary palate. The palate is formed in two parts, primary and secondary.

The commencement of the development of secondary palate takes place between seventh and eighth week of

IUL. The completion takes place about the third month of IUL. The following three outgrowth appear in the common oronasal cavity.

- A. Nasal septum-which grows downward from the frontonasal process along the midline.
- B. Two palatine processes or shelves – One palatine process develop from each side (right and left) from the maxillary processes which develop and extend towards midline.

Both the shelves elevations develop by the side of developing tongue and remain and grow there. Between

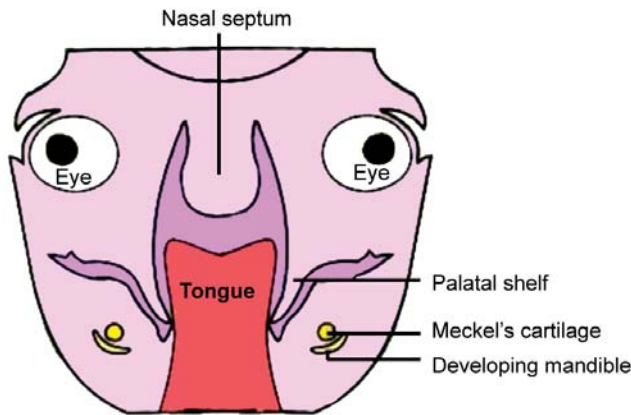


Figure 2.17: Seventh week embryo. Palatal shelves are developing downwardly on the sides of the tongue which later on come out and straighten above the tongue and face together in midline to form secondary palate

the palatine shelves the tongue occupies an elevated position. Both the palatine shelves are on the sides of the tongue, the medial margins being at the lower level than the tongue. Later on after seventh week of IUL both the palatine shelves elevate over the developing tongue leaving the tongue under them and then fuse with each other and with the primary palate (Figs 2.15 to 2.24).

The primitive common oronasal cavity is now divided into nasal and oral cavities by the fusion of the septum and the two shelves along the midline. Following three factors are responsible for this fusion and also for the closure of secondary palate.

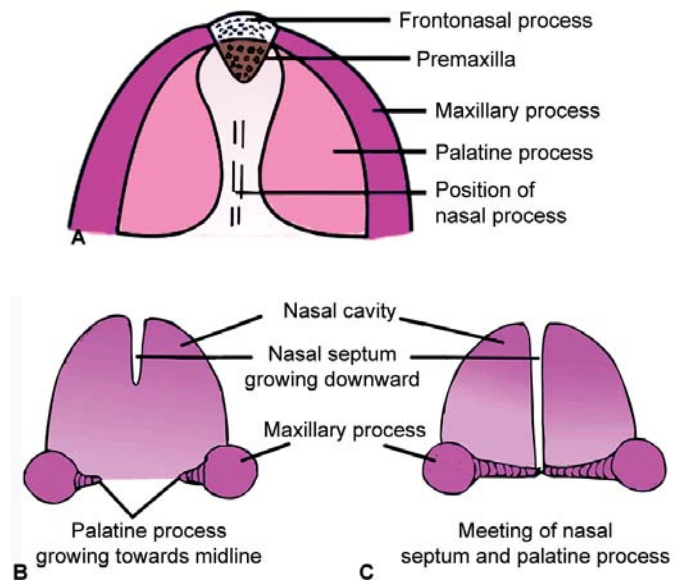
- A. The intrinsic force present in the palatine shelves helps the closure of the secondary palate.
- B. Growth pattern of the head which causes downward displacement of the tongue from between the palatine shelves.
- C. The presence of high concentration of glycosaminoglycans which attract water from the palatal shelves, which make shelves turgid due to the presence of contractile fibroblast in the palatine shelves.

In embryo between seventh and eighth weeks, the tongue and mandible in relation to upper facial complex are smaller. The lower lip is placed behind the upper lip. The upper part of the face is lifted away from the thorax by ninth week. The tongue and mandible grow forward and now lower lip is placed in the advanced position to the upper lip. The tongue is situated below the palatine shelves and all grow in this position.

THE PROCESS OF FUSION OF NASAL SEPTUM AND PALATINE PROCESSES

For the fusion of any process elimination of epithelial covering is essential. The epithelium of two palatine and nasal processes fuse as they meet and a midline union takes place. Twenty-four to thirty-six hours before epithelial contact DNA synthesis ceases. Basal epithelial cells are exposed as the surface epithelial cells are sloughed off. Rich carbohydrate surface coat of basal epithelial cells allow quick adhesion and fusion of the processes. The midline seam which consist of two layers of basal epithelial cells is formed which has to be removed to allow the ectomesenchymal continuity between the two fused processes. The seam is reduced into a thin layer then breaks up into isolated islands of epithelial cells. The basal lamina all around these epithelial cells is lost. These epithelial cells assume fibroblast like characteristics and change into mesenchymal cells.

Secondary palate is the precursor of the hard palate because it develops the hard palate and some part of the soft palate. The medial edge of the maxillary process is responsible for the development of the secondary palate (Figs 2.18 to 2.19). At the ninth week of intrauterine life, medial edges of the maxillary process come close to each other and fuse (Figs 2.17 to 2.22). Fusion of the two processes occurs because of the following.



Figures 2.18A to C: Development of secondary palate; A. Palatal view; B. Anteroposterior view of developing palatine process, nasal cavity, and nasal septum; C. Anteroposterior view of developed secondary palate, nasal septum and nasal cavity

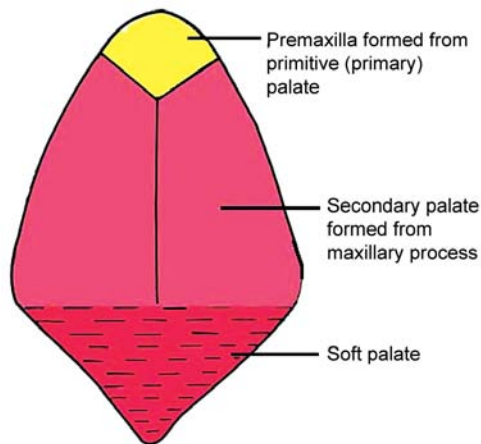


Figure 2.19: Embryological subdivisions of palate and lines of their fusion

1. Raised cyclic AMP level.
2. Cessation of cell division.
3. Production of glycoprotein.

Glycoprotein is responsible for adhesion of the two processes. The ossification of the midpalatine suture occurs at 12 to 14 years.

The growth of jaws is essential for transition from deciduous to permanent dentition. As the jaws grow in length, the permanent molars have space to develop, erupt and function. The shell-like bony enclosure protects each developing tooth (Figs 2.20 to 2.22).

DEVELOPMENT OF TONGUE

The development of tongue begins at about fourth week of intrauterine life (IUL) when the crown to rump length (CRL) is about 4 mm. Beneath the primitive mouth the pharyngeal arches meet in midline. Due to local proliferation of the mesenchyme in the floor of the mouth one elevation (swelling) appear (Fig. 2.25) in the midline which is called median tongue or tuberculum impar. Rudimentary tongue appear as a small median elevation called median tongue bud in the endodermal floor of the pharynx. It subsequently becomes incorporated in the anterior part of the tongue. This is followed by two elevations called lingual projections (bulges, swellings) or distal tongue buds one on either side of the median elevation appear on the endodermal aspect of the mandibular processes (Figs 2.15, 2.21 and 2.25).

The lateral lingual elevations and tuberculum impar quickly enlarge and merge and fuse with each other and form a large mass. Mucous membrane of anterior two third or buccal (presulcus) of tongue is formed from this mass. Along the ventral and lateral margins of this elevation a sulcus forms and deepens to form the linguogingival groove.

The posterior or root of the tongue develops from a second large median (midline) elevation called the hypobranchial eminence (copula of His), which is developed from the mesenchyme of the third arch. Hypobranchial eminence forms in the floor of the pharynx,

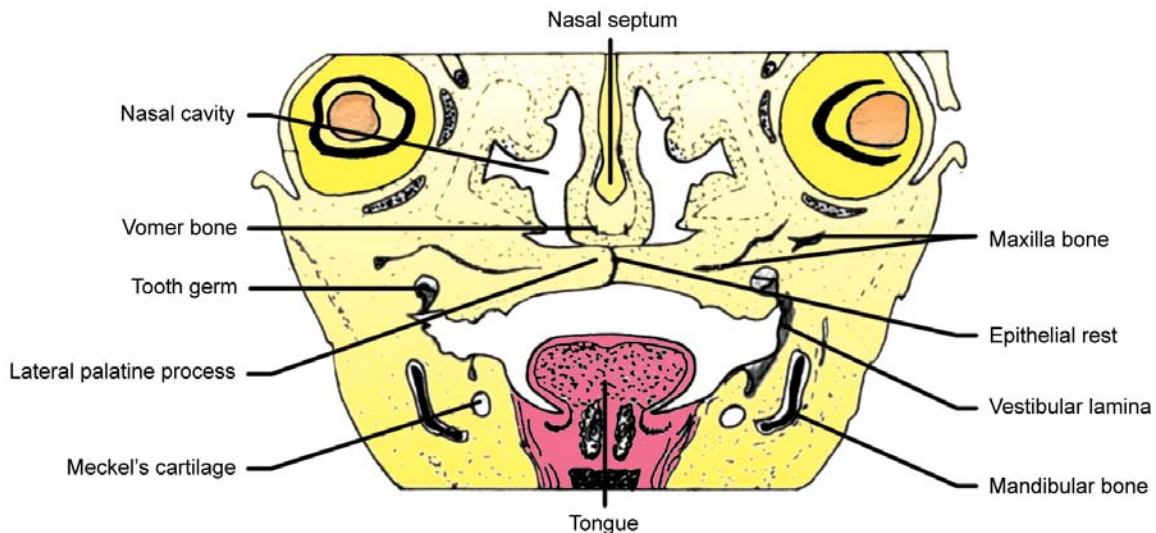


Figure 2.20: Middle part of secondary palate in a frontal section of 8 weeks embryo (30 mm crown to rump length)

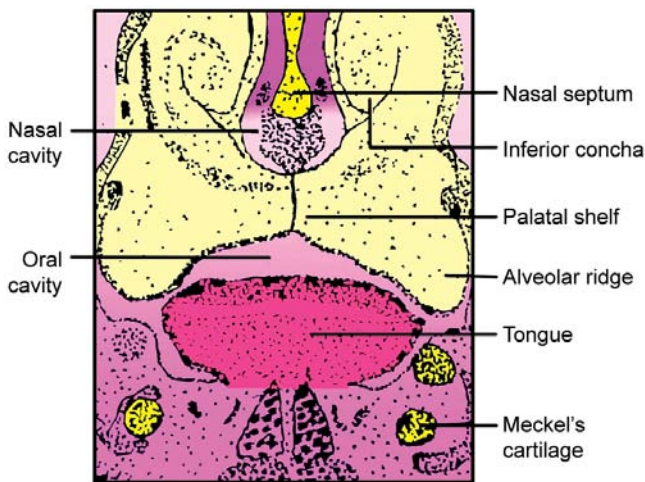


Figure 2.21: Head of embryo (10 weeks gestation, CR length 60 mm, weight 14 grams), frontal (coronal) section – horizontal palatal shelves are fused with each other and with nasal septum. Nasal cavity is separated from oral cavity by palatal shelves of secondary palate

and the ventral ends of the fourth, the third and, later, the second visceral arches converge into it. The mucosal covering of the root or posterior third of the tongue is formed by the hypobranchial eminence. From the floor of the mouth the tongue is separated by down growth of ectoderm around its periphery. Later on, this down-growth of the ectoderm around the periphery of tongue separates the tongue from the floor of the mouth. In this way lingual sulcus is formed and tongue becomes mobile.

The foramen cecum is found just behind the tuberculum impar. It is related to the development of the thyroid gland. Posterior to it, the hypobranchial eminence is found, which has two parts:

- a. Cranial part which is also called as copula, gives rise to posterior one third of the tongue
- b. Caudal part, gives rise to the epiglottis.

Muscles of Tongue

Muscles of tongue develop in the second month of IUL from the occipital myotomes (somites). These occipital myotomes (somites) migrate from the lateral aspects of the myelencephalon into the tongue area to form its musculature carrying with them their nerve supply of the twelfth cranial (hypoglossal) nerve.

Nerve Supply –The unusual composite development of the tongue explains its innervation.

1. Mucosa of anterior two-thirds of tongue derived from first arch which is supplied by the nerve of first arch

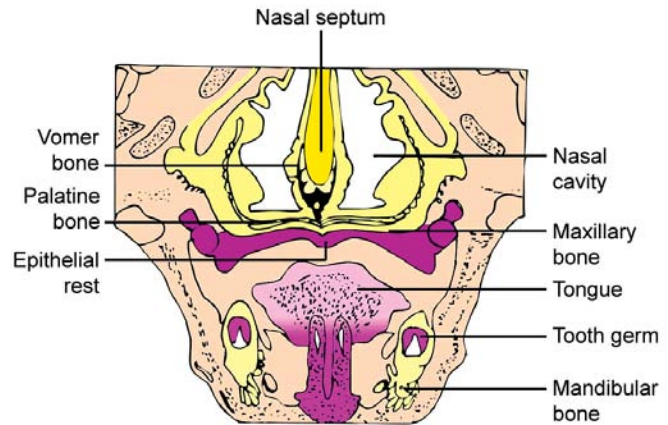


Figure 2.22: Middle part of secondary palate in a frontal section of 13-week fetus (71 mm CR length, weight 35 grams)

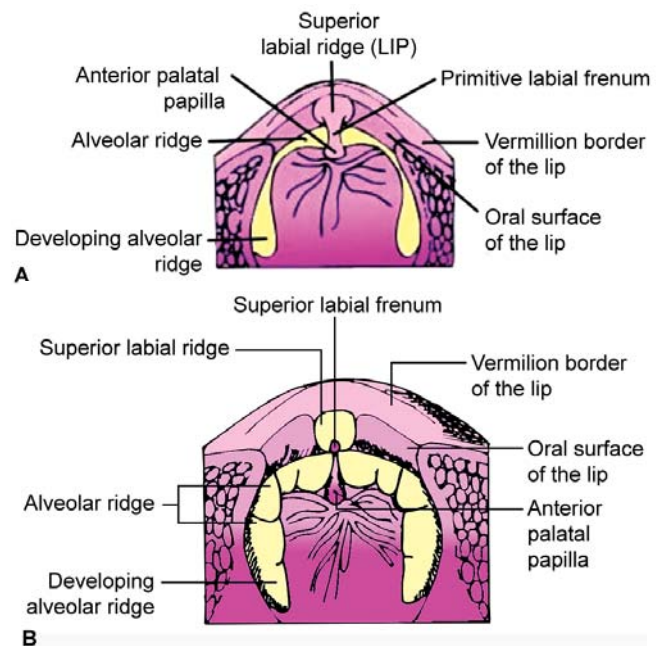


Figure 2.23: Lip and palate in an intraoral view of (A.) 3 months old (12 week of intrauterine life) (CR length 65 mm, weight 25 grams) fetus (B) 7 months old fetus

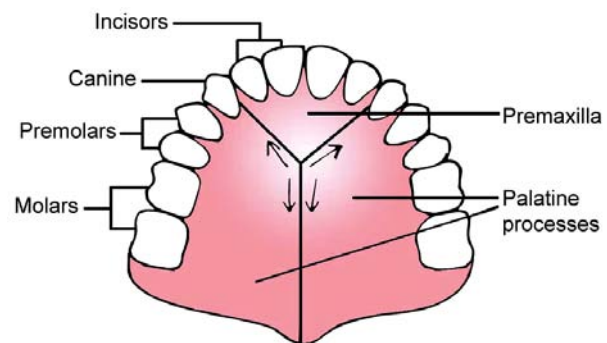
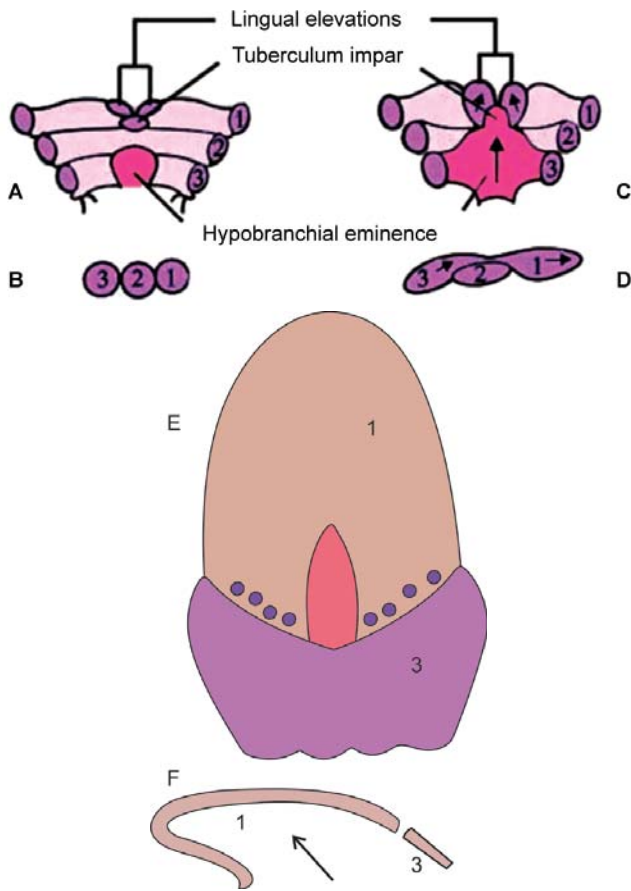


Figure 2.24: Embryonic processes forming the palate and the directions of fusion indicated by arrows



Figures 2.25 A to F: Development of tongue

- A. The floor of the primitive stomatodeum is made up of the branchial arches. In the mesenchyme of the first arch under the epithelium three elevations, one in the center called tuberculum impar and two lingual elevations, one on either side of the midline appear. In the third arch a mid line elevation called hypobranchial eminence appear.
- B. Sagittal section through the arches.
- C & D. Lingual elevations (projections) increase in size and along with tuberculum impar from the anterior two third of the tongue. The hypobranchial eminence grow over the second arch. Sagittal section is shown in (D)
- E. Final formation of tongue showing relative contributions of the first and third arches
- F. The sagittal section showing formation of tongue from first and third arch. The thick arrow showing the route of incoming occipital myotomes which form the muscles of tongue

which is derived from the fifth cranial (trigeminal nerve).

- a. The lingual nerve is derived from the posttrematic nerve of first arch (mandibular nerve)
 - b. The chorda tympani, the pretrematic nerve to the first arch.
2. Mucosa of posterior third (pharyngeal) part of the tongue which is derived from the third arch is supplied

by the ninth cranial (glossopharyngeal) nerve which is the nerve of third arch.

3. Motor supply of the muscles of the tongue – The muscles of the tongue being myotomic in origin, are supplied by the hypoglossal (twelfth cranial) nerve.

The sulcus terminalis is distinguished at the age of nine weeks of IUL (CRL 52 mm, fetal weight 13 gm). The vallate papillae appear at about the same age, increasing in number up to 170 mm stage.

GROWTH OF FACE AND JAWS

For the purpose of understanding, the growth of skull has been described under the following headings.

1. Growth of Face - Upper jaw (maxilla) and Lower jaw (mandible)
2. Growth of Cranium - Vault and Base
3. Sinuses

The growth of the face is completed after the growth of cranium (Figs 2.26A to F)

Development of Maxilla and Mandible

Skull is divided into following three components

- i. The cranial vault
- ii. The cranial base and
- iii. The face

In the beginning of the second month of fetal life, the skull consist of the following

- a. The cartilaginous chondrocranium, which forms the base of skull.
- b. The membranous desmocranium, which forms the lateral walls and roof of the brain case.
- c. The cartilaginous visceral part which consist of skeletal rods of the branchial arches.

Maxilla

Maxilla is connected with many bones by the help of sutures. These bones are frontal, zygomatic temporal nasal, palatine, perpendicular plate of ethmoid, lacrimal, and the maxilla of opposite side.

Development of Maxilla

The development of maxilla starts at sixth week of IUL. Maxilla develop from the tissues of the first branchial arch. The maxilla form within the maxillary process.

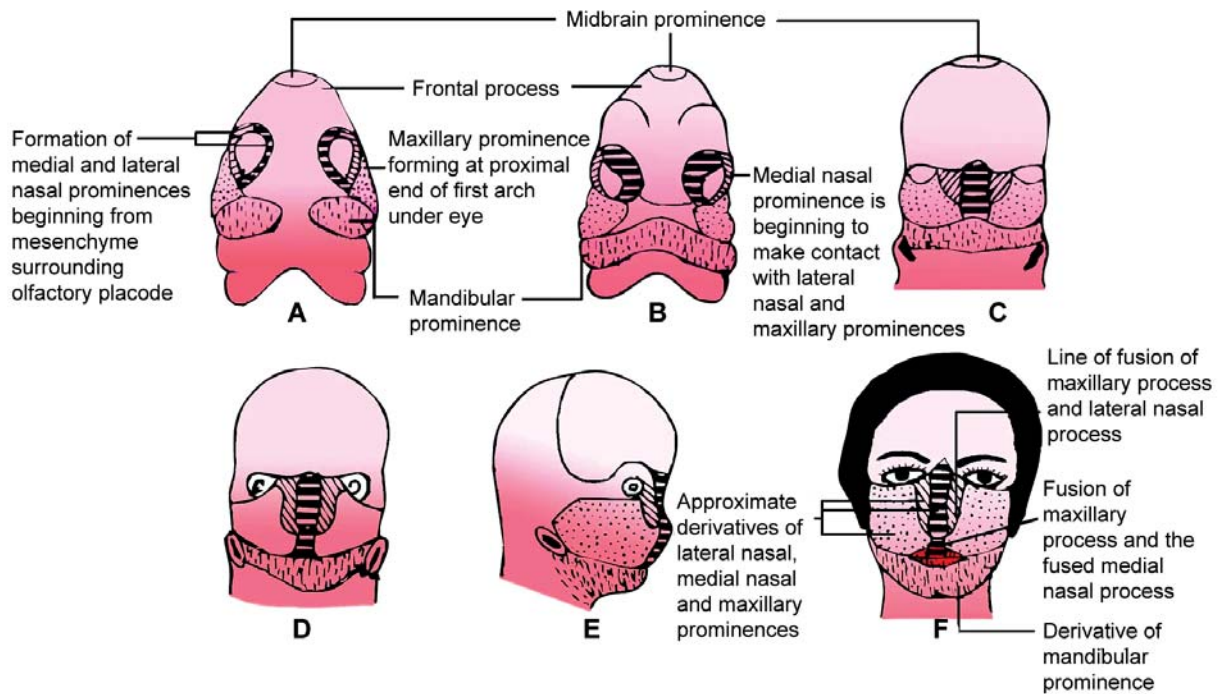


Figure 2.26: Schematic development of human face showing contribution of various facial prominences (A) Embryo 4 to 6 mm CR length (approximately 30 days), (B) Embryo 8 to 11 mm CR length (approximately 40 days), (C) Embryo 16 to 18 mm CR length (approximately 47 days), (D) & (E) Embryo 23 to 28 mm CR length (approximately 55 days), (F) Adult face. (Note: Prominence is also called as process)

Maxilla develop from a center of ossification in a sheet of mesenchymal tissue which is superficial and closely associated to the nasal capsule. In maxilla also (like mandible) the center of ossification appears in the angle formed by the two nerves. Center of ossification appear between the angle formed by anterosuperior dental nerve as it comes out from the inferior orbital nerve. Bone formation, from this center spreads towards the developing zygoma posteriorly under the orbit. Anteriorly it spreads towards the future incisor region. For development of frontal process ossification spreads upwards. For the infraorbital nerve a bony canal also develops. By the downwards extension of bone lateral alveolar plate for the tooth germs is formed. To develop the hard palate ossification spreads into the palatine processes. The main body of the developing maxilla and the junction of the palatal process give rise to the medial alveolar plate. A trough of bone around the maxillary tooth germs is formed with medial alveolar plate along with its lateral counterpart.

In the formation of maxilla secondary cartilage also help. A secondary cartilage called zygomatic or malar cartilage develops in the zygomatic process. After third month of IUL there is no indication on the facial aspect of

the upper jaw of a premaxilla (os incisivum). Rarely a suture or somewhat like a suture is observed on the floor of the nasal cavity. During sixteenth week of IUL maxillary sinus develops as a shallow groove on the nasal aspect of the developing maxilla.

At birth (1) the frontal process of maxilla is well developed (2) Body of the bone is not well developed. It consists of under developed alveolar process with the tooth germs. (3) Zygomatic and palatal processes are present but are very small (4) As maxillary sinus is rudimentary (about the size of pea) the body of the maxilla is comparatively small (5) A suture line or cleft is present at birth in the anterior region of the palate, which is diverging to each side and form the incisive fossa. This suture runs into septum between the lateral incisor and canine teeth or very rarely between canine and first premolar. Until the age of thirty years the palatal indication of separation between the os incisivum and the rest of the maxilla may persist (Figs 2.24 and 2.27).

The direction of growth of maxilla is downward and forwards

A. Maxilla grows in height by the following.

1. Continuous apposition of the alveolar process.



Figure 2.27: The region of developing deciduous canine showing early developing maxilla

2. Apposition on the inferior palatal surface.
 3. Resorption of nasal floor.
- B. Maxilla grows in width by the following.
1. Growth of median palatal sutures.
 2. Appositional growth of maxilla.

Development of Mandible

In the sixth week of intrauterine life (CR length 22 mm, weight 6 gm), the mandible develops as an intramembranous bone by the side of the Meckel's cartilage. It develops as a bilateral thin plate of bone. The mandible develops from the tissues of the first branchial arch and develops within the mandibular process. The mandible is formed in dense fibromembranous tissues which lies lateral to the inferior alveolar nerve and its incisive branch and the lower portion of Meckel's cartilage. Meckel's cartilage is also known as ventral mandibular cartilage.

Meckel's cartilage has a close positional relationship or proximity to the developing mandible. Meckel's cartilage does not make any contribution in development of mandible. At sixth week of IUL each half of mandible is ossified from one centre which appears near the mental foramen. Meckel's cartilage extends as a solid hyaline cartilaginous rod, surrounded by a fibrocellular capsule extending from the midline of the fused mandibular processes the developing otic capsule. Both the cartilages one of each side do not meet at midline but are separated by a thin band of mesenchyme. The mandibular branch which is a branch of trigeminal nerve lie in close relationship to the Meckel's cartilage about two third along the length of the cartilage.

During sixth week of IUL on the lateral aspect of Meckel's cartilage a condensation of mesenchyme occurs in the angle formed by the division of the inferior alveolar nerve and its incisive and mental nerve branches.

Intramembranous ossification starts in this condensation at the seventh week of IUL. From this centre ossification spreads and body of the mandible and the ramus are formed. (Figs 2.28 to 2.33) Rapidly the ossification spreads in the following directions.

- a. Anteriorly to the midline
- b. Posteriorly towards the place where the mandibular nerve divides into the lingual and inferior alveolar branches. The new bone formation takes place anteriorly by the lateral side of the Meckel's cartilage. In this way a trough is formed which consist of lateral and medial plate which unite.

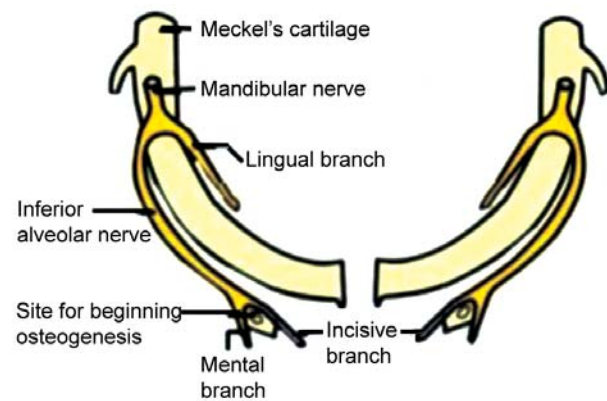


Figure 2.28: Mandible formation showing site for beginning osteogenesis where the inferior alveolar nerve divide into incisive branch and mental branch

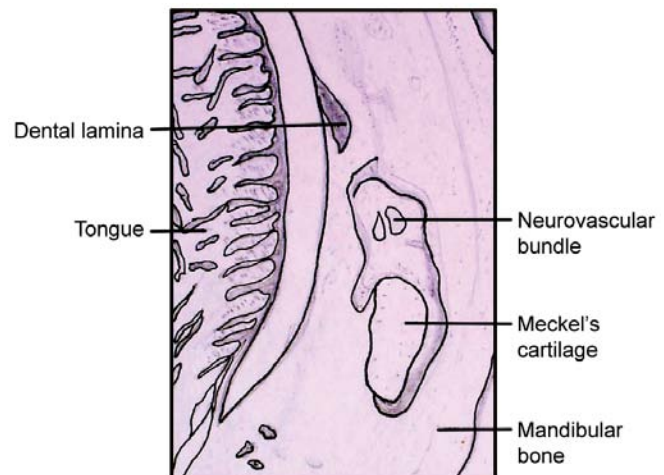


Figure 2.29: A small portion of mandibular bone formation is seen intramembranously on the lateral aspect of Meckel's cartilage in a transverse section through early developing mandible (eighth week of development)

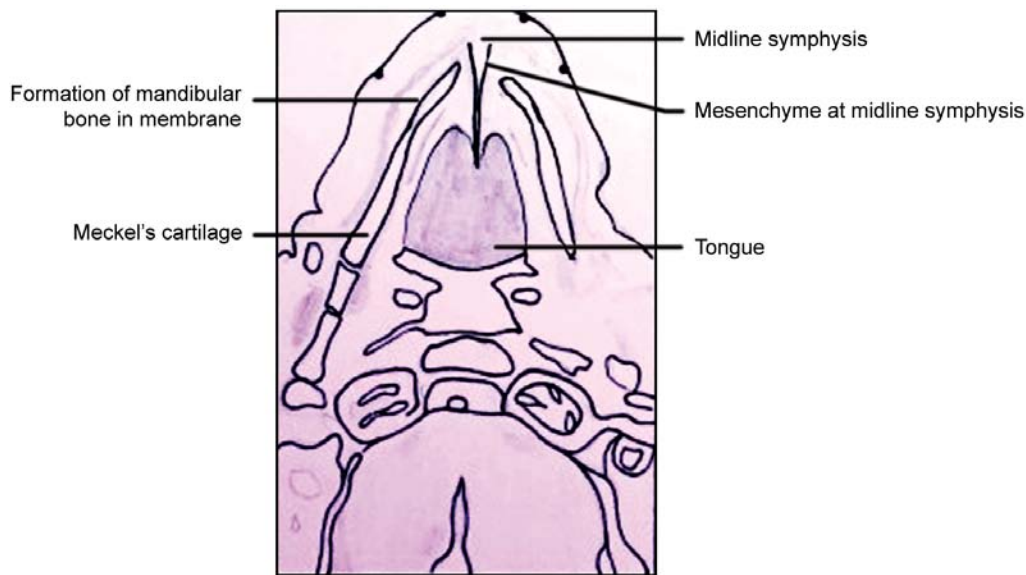


Figure 2.30: Horizontal section through developing mandible during eighth week of intrauterine life showing formation of mandible in membrane around Meckel's cartilage

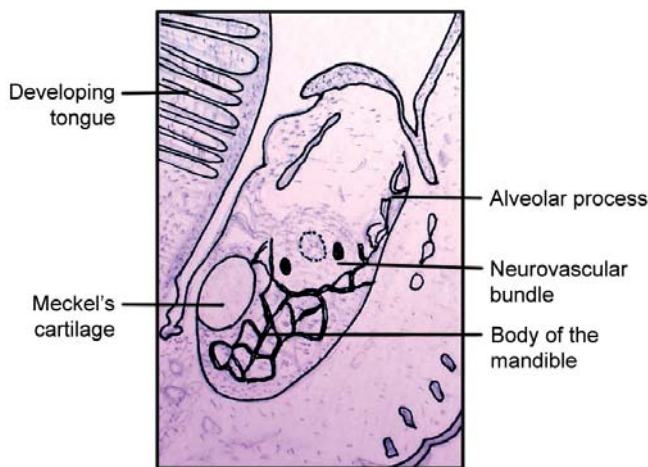


Figure 2.31: A later stage in the development of body of mandible showing the increase in size of mandible through continued bone formation

The troughs of bone of both the sides meet at midline. Until shortly after birth the two centres of ossification remain separate at the mandibular symphysis. The lateral and medial plates join and trough is soon converted into a canal. The crypts for the developing teeth is also formed. By tenth week of IUL the part of Meckel's cartilage below the incisor teeth is surrounded and invaded by bone.

Along with the lateral aspect of Meckel's cartilage a backward extension of ossification forms a trough which

later converts into a canal which contains the inferior alveolar nerve. In the condensed mesenchyme the backward extension of ossification proceeds to the point where the mandibular nerve divides into the inferior alveolar and lingual nerves. In this canal in the bone mandibular nerve extends to midline and medial and lateral alveolar plates of bone. Above this canal wide trough develop in relation to developing tooth germs in such a way that the tooth germs occupy a secondary trough. This wide secondary trough of bone have small compartments divided by small partitions. Each compartment is occupied by individual tooth germ which is covered by growing bone. In this manner the body of the mandible is formed.

Up to birth further growth of the mandible is greatly effected the appearance of following three secondary (growth) cartilages and the development of muscular attachments.

A. Condylar cartilage – It is a cone or carrot shaped mass of cartilage. It develops during twelfth week of IUL. It extends from the head of the mandible downwards and forwards through the ramus. It contributes to the growth in height of the ramus. It is largely invaded and replaced by bone by the middle of fetal life. At twentieth week (about middle of IUL) by endochondral ossification this mass of cartilage is converted to bone. Only a thin layer of cartilage remains in the condylar

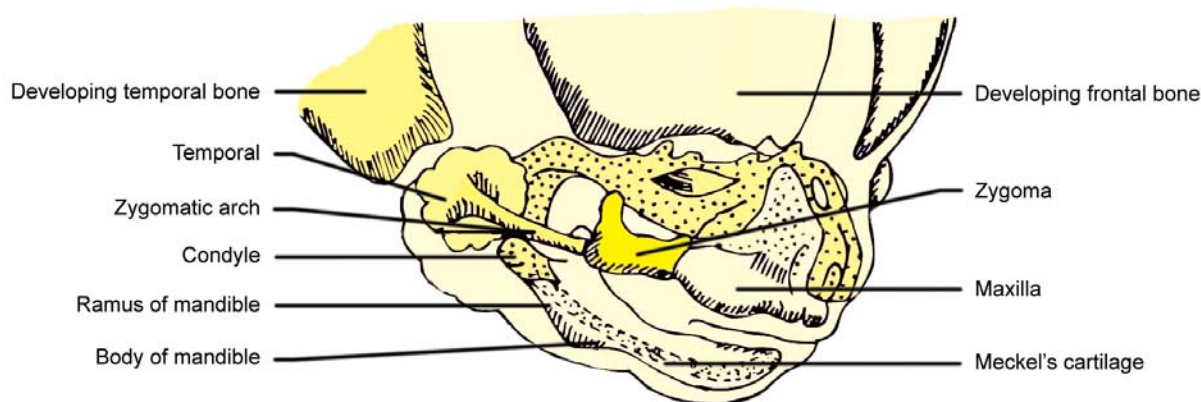


Figure 2.32: Developing facial skeleton of ten weeks old fetus (CR length 60 mm, weight 14 gm) showing relationships of various bones and cartilages with each other

head. Its upper end persists as a zone of proliferating cartilage beneath the fibrous articular surface of head providing a mechanism of growth (like epiphyseal cartilage of long bone) until the third decade (at the end of second decade of life).

- B. The coronoid cartilage – It appears along the anterior border and top of the coronoid process at about fourth month of IUL. It is a transient growth cartilage. It disappears before birth
- C. The symphyseal cartilage – One or two cartilage nodule appear on each side at the symphysis menti. Usually they are two in number, one on each side, but rarely they can be four in number, two on each side

They appear in the connective tissue between the two ends of Meckel's cartilage but do not have any relation with it. During seventh month of IUL these may ossify to form a variable number of small ossicles, called mental ossicles, which are present in the fibrous tissue of the symphysis. Before the end of first year of life these ossicles unite with the bone.

Development of Ramus

Posteriorly by a rapid spread of ossification into the mesenchyme the ramus of the mandible is formed away from the Meckel's cartilage. In adult mandible the place of angulation is identified by the lingula. Lingula is the place where inferior alveolar nerve enters the body of the mandible. At tenth week of IUL by membranous ossification with little direct involvement of Meckel's cartilage the rudimentary mandible is developed.

Fate of Meckel's (Ventral Mandibular) Cartilage

- A. The dorsal and posterior end of Meckel's cartilage form the rudimentary of both malleus and incus of the inner ear and its fibrocellular capsule remains as sphenomandibular ligament. The cartilage is totally lost and resorbed completely from the sphenoid to the area where mandibular nerve divide into alveolar and lingual branches.
- B. Meckel's cartilage is completely resorbed from the lingula forward to the division of the alveolar nerve into its incisor and mental branches.
- C. From the lingula and the division of alveolar nerve into its incisor and mental nerve to the midline Meckel's cartilage makes a small contribution to the mandible by means of endochondral ossification.

In a nutshell the characteristics of development of mandible are as follows.

- a. It is a membrane bone.
- b. It develops in relation to the nerve which is derived from characteristics of arch.
- c. It develop almost totally independent of Meckel's cartilage.
- d. The mandible has neural, alveolar and muscular elements.
- e. Its growth is helped by development of secondary cartilages.
- f. At birth mandible is in two separate halves which are united by fibrous tissue in the median plane which is called symphysis menti.

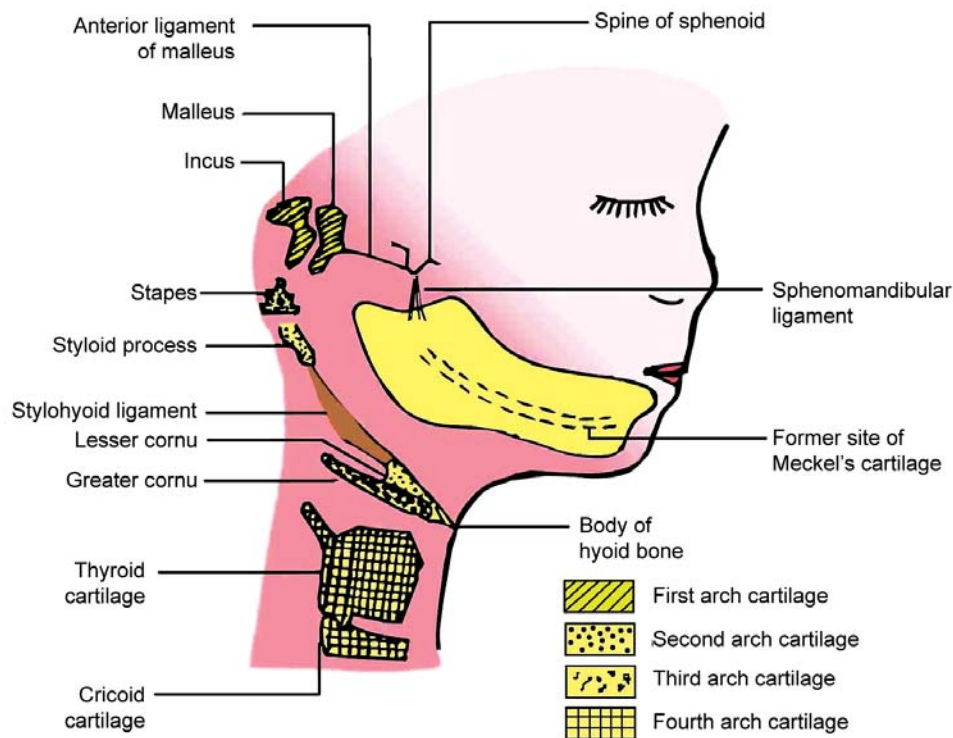


Figure 2.33A: Development of fetal mandible and its relationship to Meckel's cartilage, which serves as a template and scaffolding for ossification and other branchial arch derivatives

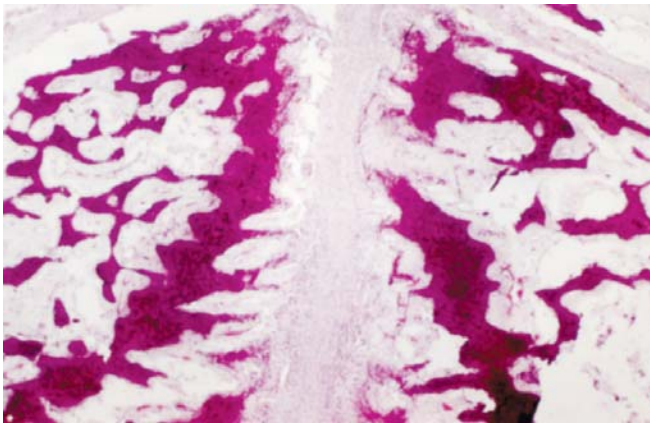


Figure 2.33B: Development of mandible in 16 week old fetus. The symphysis menti that is the joint between the two rami is fibrous in nature. Trabeculae of mandibular bone are present on both sides (H & E x 50)

- g. The main growth center of the mandible is the condyle.
- h. At the birth it is fibrocartilaginous in nature. Later it is converted into a bony structure.
- i. The direction of growth of the condyle is upwards and backwards. This results in the downward and forward growth of the mandible.

- j. The mandible ossifies from two centers located medially to the Meckel's cartilage.

The length of the mandible increases by the following.

1. Appositional growth along the posterior border of the ramus.
2. Resorption of the anterior border of the ramus

The height of the mandible increases by appositional growth of the alveolar border. The width of the mandible increases by appositional growth of outer surface and posterior border

Common Developmental Features Between Both the Jaws

1. Development of both begin from a single center of membranous ossification which is related to a nerve and to a primary cartilage.
2. Both develop secondary cartilages to help in their growth.
3. Both form a neural element related to the nerve.
4. Both develop the alveolar process which contains developing teeth.

Facial processes and prominences are given in Table 2.3.

Table 2.3: Facial processes and prominences produce following parts of mouth and face

Processes and prominences	Parts of mouth and face produce
1. Maxillary process	<ul style="list-style-type: none"> • Upper alveolar process • Hard palate • Zygomatic bone
2. Mandibular process	<ul style="list-style-type: none"> • (Lower) alveolar process • Body of mandible • Lower lip
3. Median nasal process	<ul style="list-style-type: none"> • Frenum • Nasal septum • Philtrum • Premaxilla
4. Lateral nasal process	<ul style="list-style-type: none"> • Lateral cartilage of nose • Lacrimal and other nasal bones
5. Stomodeum	<ul style="list-style-type: none"> • Anterior part of soft palate • Hard palate • Lip • Teeth

Growth of Cranium

From the gestation period, growth of the cranium occurs by endochondral, interstitial and sutural growth. Growth of cranium occurs in all directions (Fig. 2.34).

Basically the skull contains the mandible, two maxillae and cranium. Various types of cranial bones are set together with the help of sutures to form a cranium. For better understanding, the growth of cranium is studied in two parts.

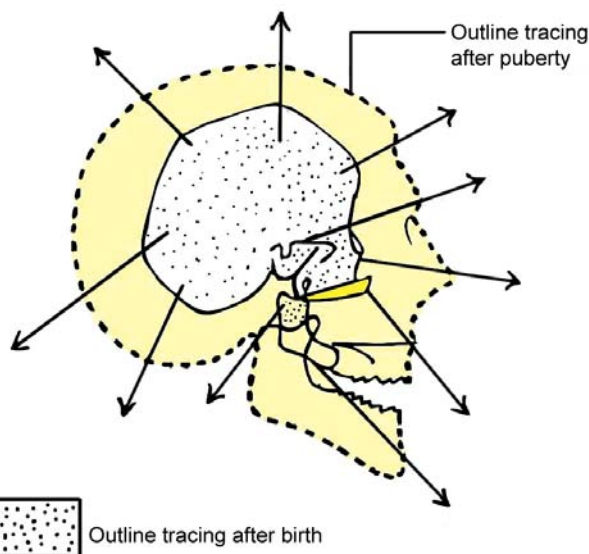


Figure 2.34: Growth pattern of cranium and face. Skull grows in all directions as indicated by arrows

Cranial Vault

The cranial vault consists of one frontal bone, two parietal bones, one occipital bone, two temporal bones, and one sphenoid bone. All these are membranous bones. They are fused together through sutures and form a single structure. The growth of the cranium is because of the growth of the brain. The growth of the brain precedes the growth of the cranium. About 90 percent growth of the brain is completed by the age of five years.

Cranial vault grows with the help of apposition of various individual bones, proliferation and ossification of suture lines and selective resorption. The height of the cranium increases by the sutural growth of parieto-occipital, parietosphenoidal and parietotemporal sutures. The length of the cranium increases by sutural growth of parietosphenoidal, parietotemporal and interparietal sutures.

Cranial Base

The type of growth that takes place in the cranial base is synchondrosis that is cartilaginous growth. This is the type of growth in which cartilage is first formed, which is subsequently converted to bone before adulthood. The actively participating areas are - intraoccipital, sphenoid and ethmoidal and sphenoccipital.

Growth of Paranasal Air Sinuses

The paranasal air sinuses are the air filled spaces in the skeletal bones around the nasal cavity. One of the functions of sinuses is to reduce the weight of bone and skull. The sinuses grow secondary to the bone. Sinuses grow by the process of expansion of the middle ear into the neighboring bone and evagination of nasal chambers.

The growth of the maxillary sinus is dependent upon the eruption schedule of maxillary teeth. For the first time it is seen in an eight-week-old embryo and it further develops just before birth. It expands and modifies in form till eruption of all permanent teeth.

The face develops during the short period between fourth to seventh prenatal weeks. The environmental factors can cause a facial defect before the fourth week. In intrauterine life, therefore, special care should be taken to avoid irritation, chemical, hormonal or dietary imbalance and stress, etc. before the fourth week of the intrauterine life.

CLINICAL CONSIDERATIONS

During the development of various structures in the human body, multiple types of defects are seen, which can occur because of malunion or nonunion of whole or a part of one process to that of another process. In the facial and palatal developmental defects, environmental factors play an important role up to the fourth week of intrauterine life. Most of the face forms from the tissues present on the surface of the brain, hence defects of the anterior brain may give rise to congenital defects of face.

Facial Clefts

Among congenital malformations, cleft lip and cleft palate are most common. According to the latest estimates, they occur in one out of every 1500 births in the white population and in one out of 3000 births in the black population in the US, and one out of 5000 births in the Indian subcontinent.

Cleft can be unilateral or bilateral and it can be complete or incomplete, with or without cleft of lip and alveolus (Fig. 2.35). The etiological factor involved in cleft lip is nonunion of maxillary process and globular process, and in cleft palate it is nonunion or incomplete union of two palatine processes.

The oblique facial cleft results from malunion of the maxillary process and lateral nasal process. The lateral facial cleft results from the failure of fusion of maxillary process and mandibular arch. The incidence of cleft is more in children of (a) an epileptic mother who takes phenytoin therapy, (b) a pregnant mother exposed to hypoxia and (c) a smoker mother.



Figure 2.35: Cleft lip, palate and alveolus of left side (as seen indirectly in intraoral mirror). (Courtesy: Dr SC Pandey, Lucknow)

Fissural (Inclusion, Developmental) Cysts

Fissural cysts are cystic cavities, which arise along the fusion of various bones or embryonic processes and are lined by epithelium.

Median anterior palatine cyst (Nasopalatine duct cyst, median anterior maxillary cyst). This cyst is present at the midline of maxillary alveolar process, near the incisive canal. It arises due to proliferation of epithelial remnants of the nasopalatine duct, which is an embryological structure consisting of a duct or cord of epithelial cells lying inside the incisive canal. It is the most common maxillary developmental cyst.

Globulomaxillary cyst The globulomaxillary cyst is an inverted, pearshaped structure found inbetween maxillary lateral incisor and canine. The globulomaxillary cyst is observed within the bone at the junction of all three facial prominences. The facial prominences are one medial nasal process and two maxillary processes.

Palatal cysts of the neonate (Epstein's pearls, Bohn's nodules) These are tiny multiple cysts observed on the palate of neonates and fetuses. They are mostly present along the junction of the hard and soft palate. They arise from the epithelial remnants of developing palatal salivary glands.

Thyroglossal tract cyst It is a rare developmental cyst which may form anywhere along the embryonic thyroglossal tract between the foramen caecum of the tongue and the thyroid glands. It arises from remnants of this tract.

Median mandibular cyst It is found in the midline of the mandible. It is very rare.

Nasopalatine cyst (Nasolabial cyst; Klestadt's cyst) It is a rare fissural cyst and may secondarily involve bone. It arises from the proliferation of entrapped epithelium at the junction of the globular process, the lateral nasal process and maxillary process.

Benign cervical lymphoepithelial cyst (Branchial cleft cyst; lateral cervical cyst; benign cystic lymph node) They originate from the remnants of the branchial arches or pharyngeal pouches. They occur on the lateral aspect of the neck.

Epidermoid and dermoid cysts They are derived from embryonic germinal epithelium.

Nasolabial cysts They are very rare and develop from the gastric or intestinal mucosa occurring in the oral cavity due to misplaced embryonal zones.

Heterotopic oral dermoid cyst It develops from oral vestibule and nasal cavity. It causes a depression on the alveolar process.

Developmental Anomalies of Tongue

Median rhomboid glossitis (Central papillary atrophy of the tongue) It results from persistence of the tuberculum impar. It is characterized by a red, smooth zone as seen in the midline, slightly anterior to the foramen caecum.

Bifid tongue (Cleft tongue) It is seen when the two lateral processes of the tongue are not fused completely. It is very rare.

Macroglossia (Enlarged tongue) It is due to an over-development of the musculature of the tongue.

Microglossia (Small tongue) It is very rare and due to an under-development of the tongue

Ankyloglossia It is due to the fusion of tongue with the floor of the mouth. Mostly it is partial and very rarely complete.

Fissured tongue (Scrotal tongue) In this there are numerous small fissures or grooves on the dorsal surface of the tongue.

Syndrome: Treacher Collin's Syndrome

Edward Treacher Collin, a British ophthalmologist (born in 1862 and died in 1919) described this syndrome. It is also called mandibulofacial dysostosis. It is an inherited disorder characterized by the presence of a dominant gene. The features of this syndrome are defective development of ears, secondary palate and maldevelopment of various facial prominences.

Hemifacial Microsomia

Microsomia indicates the structural development, which is abnormally smaller than normal. It is the third most common craniofacial malformation. It has been reported when the drug thalidomide was taken during early pregnancy by the mother. It is characterized by underdevelopment of the muscles of mastication, external ear, parotid gland and temporomandibular joint. When the abnormalities of vertebrae are also involved, the condition is called as oculoauriculo-vertebral syndrome.

Environmental teratogens also affect the development of normal cells, tissues and organ systems.

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Development of Teeth

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INTRODUCTION

The development of the tooth involves many complex biological processes, including epithelial mesenchymal interactions, morphogenesis and mineralization. In human beings, 20 deciduous and 32 permanent teeth develop from the interaction between the oral epithelium cells and the underlying mesenchymal cells. The basic developmental process is similar for all teeth but each developing tooth develops as an anatomically distinct unit. Vitamins, minerals and hormones affect tooth development. Vitamin A is important for epithelial growth, vitamin C for connective tissue development and vitamin D is essential for calcification. A brief description of development of teeth in the form of flow charts has been given (Figs 3.1 and 3.2).

PRIMARY EPITHELIAL BAND

The oral ectoderm is neural crest or ectomesenchyme in origin. It is lined by stratified squamous epithelium. The initial oral cavity develops after the rupture of the buccopharyngeal membrane at the fourth week of intrauterine life. The interaction between the oral epithelium and the underlying mesenchymal cells results in tooth development.

After thirty-seven days of development, a continuous band of thickened epithelium forms around the mouth in both the future upper and lower jaws. This occurs from the fusion of separate plates of thickened epithelium. These bands of the epithelium are roughly horseshoe-shaped structures. These correspond in position to the future dental arches in the presumptive upper and lower jaws (Fig. 3.3).

The primary epithelial band forms as a result of a change in orientation of the plane of the dividing cells (Figs 3.4 A to C and 3.5).

Dental Lamina

At the sixth week of gestation period, certain areas of basal cells of the oral ectoderm proliferate more rapidly than the adjacent cells. The primary epithelial band forms two subdivisions called the dental lamina and the vestibular lamina. The dental lamina is a band of epithelium that has invaded the underlying ectomesenchyme along both the horseshoe-shaped future dental arches (Figs 3.4 to 3.6).

Deciduous dentition develops directly from the dental lamina at the eighth week of fetal life, whereas the permanent molars develop from a distal extension of the dental lamina.

The initiation of the permanent first, second and third molars occurs at the fourth month of intrauterine life, one year after birth and five years after birth respectively. The lingual extension of dental lamina is called successional lamina. Successional lamina is responsible for the development of permanent incisors, canine and premolars. The successional lamina is active from the fifth month in utero (for the permanent central incisor) to ten months of age (second premolar).

Fate of Dental Lamina

After initiation of tooth development, the dental lamina degenerates. Total functional activity period of the dental lamina is around five years. Sometimes it takes more than five years, when the initiation of tooth development is delayed. After functional activity, the remnants of the dental lamina may persist in the jaw or gingiva in the form of islands or epithelial pearls. These are known as cell rest of Serres. At any specific portion the dental lamina functions for a shorter period, as only a relatively short time elapses between initiation of tooth development and degeneration of dental lamina. It goes on degenerating in anterior teeth and activating in the posterior teeth. In the

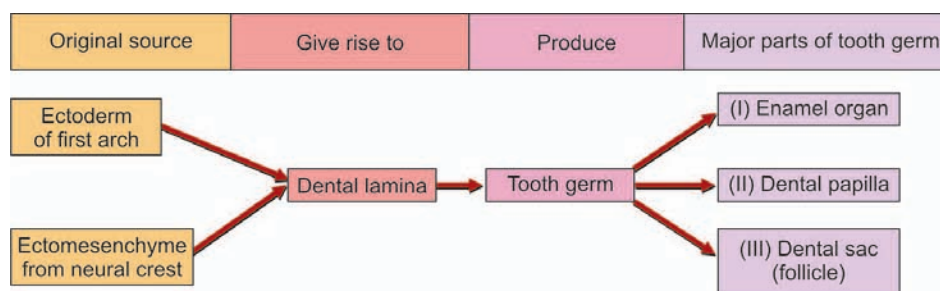


Figure 3.1: Development of tooth germ

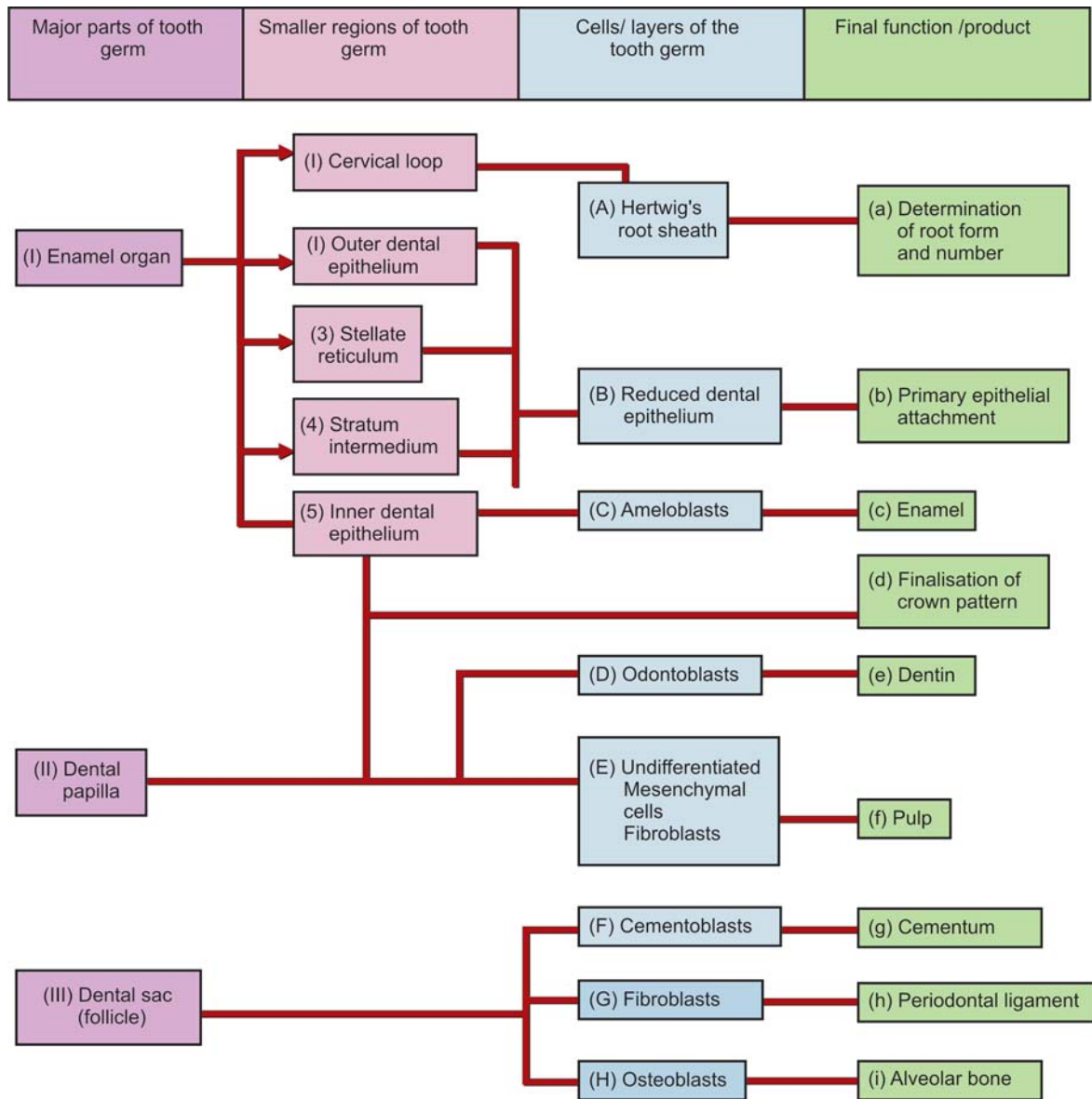


Figure 3.2: Development from tooth germ to complete tooth formation

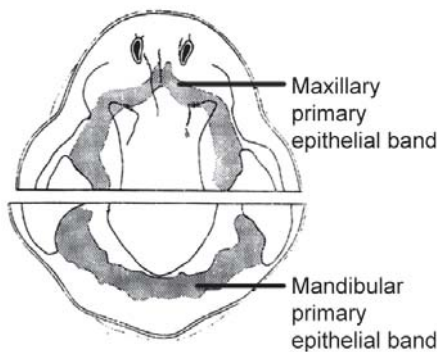
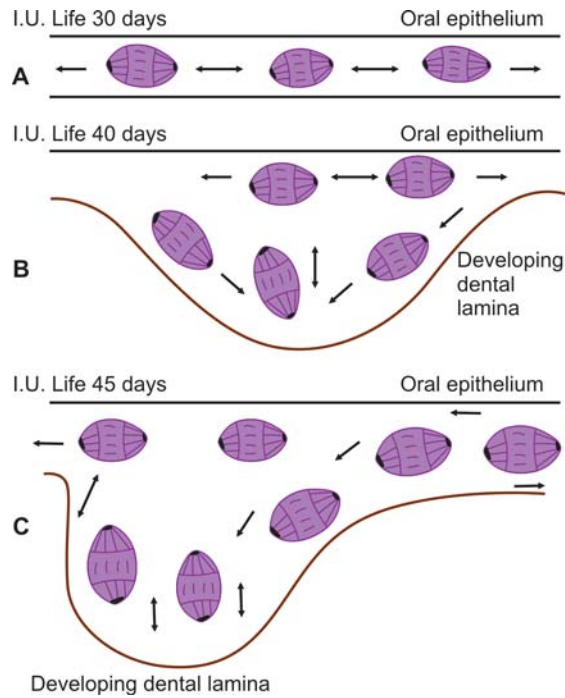


Figure 3.3: Reconstructed early oral cavity. The position of the maxillary and mandibular primary epithelial bands is shown by shaded areas. (Adapted from Nery, E.B., Kraus, B.S., and Croup, M.: Arch. Oral Biol. 15:1315, 1970.)



Figures 3.4 A to C: Section through oral epithelium showing the change in the plane of cleavage of cells of primary epithelium resulting in its thickening, producing dental lamina from 30 to 45 days of intrauterine life (IU Life).

A. Cell division of oral epithelium at 30 days of IU life.

B. Cell division of developing dental lamina at 40 days of IU life

C. Cell division of developing dental lamina at 45 days of IU life.



Figure 3.5: Initiation of tooth development. Embryo about 12 mm in length (fifth week). Thickened oral epithelium as seen under high magnification. Some cells are undergoing mitosis both in epithelium and mesoderm

incisor area the dental lamina is first to appear and disappear after initiation of tooth development, and in the third molar area it is the last to appear and disappear.

As the teeth continue to grow, the connection of teeth with the dental lamina is lost by mesenchymal invasion.

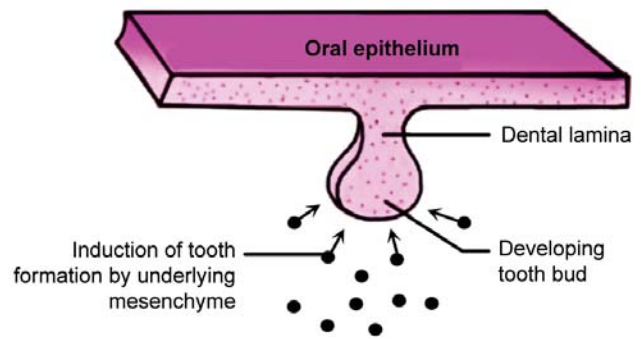


Figure 3.6: Developing tooth bud with dental lamina from oral epithelium

Vestibular Lamina

Facial (labial and buccal) to dental lamina another thick band of epithelium develops in the maxillary and mandibular dental arches. It is called as the vestibular lamina or the lip furrow band. It develops somewhat later and independently (Figs 3.7 and 3.8). It later hollows out and forms the oral vestibule between the alveolar portions of the jaws and the lips and cheeks. The oral epithelium in the lower jaw forms an epithelial invagination separating the tongue from the developing alveolar process termed the linguoalveolar sulcus.

DEVELOPMENTAL STAGES OF TOOTH

Tooth formation is a continuous process. It is characterized by a series of stages. Each tooth develops through successive bud, cap and bell stages. During these

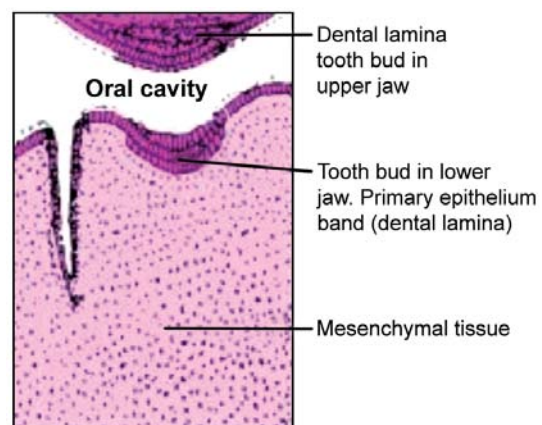


Figure 3.7: The oral epithelium thickens and invaginates into the mesenchymal tissue to form primary epithelium band (dental lamina) for the development of tooth (bud stage)

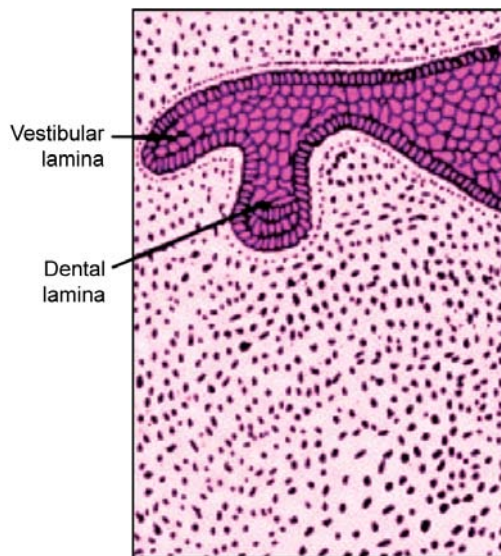


Figure 3.8: The primary epithelial band divides into two processes, the vestibular lamina and the dental lamina (embryo sixth week, CR length 10 mm)

stages, the tooth germs grow and develop into specialized cells which form the enamel, dentin and the cementum. The number of deciduous teeth is twenty, that is ten maxillary and ten mandibular. Dental lamina plays an important role in the development of teeth. The division of ectodermal cells of the dental lamina at the sites of development of the deciduous dentition results into the formation of ten knob-like structures in each jaw, five on each side of each jaw. This is the initial stage of the life cycle of a tooth. In total, twenty such structures called enamel organs develop. These knob-like structures develop rapidly and grow into the underlying ectomesenchyme (Fig. 3.8). These enamel organs represent the tooth bud of deciduous dentition. First these enamel organs develop in the mandibular anterior region. These enamel organs increase in size, as the cells continue to proliferate. They take the shape of a cap with their outer surface towards the oral cavity.

Inside the depression of the enamel organ, that is, inside the cap, the ectomesenchyme cells increase in number. They look more dense and represent the beginning of the dental papilla. The ectomesenchymal cells and fibers that surround the dental papilla and enamel organ develop. This is known as the dental sac or dental follicle. Formation of enamel is from the enamel organ, dentin and pulp from dental papilla and periodontium from dental sac or dental follicle. The shape of the enamel organ

continues to change. The dental lamina breaks up and the tooth bud loses its connection with the epithelium of the primitive oral cavity (Figs 3.9 A to I). Interaction of the first arch epithelium and neural crest cell results in the development of tooth. The development of tooth is controlled by genes through molecular signals.

The anatomy of all the teeth is different from each other, but they pass through similar stages of tooth development. For better understanding of the development of the tooth, it is described under the following stages:

- A. Bud stage - Initiation
- B. Cap stage - Proliferation
- C. Bell stage -
 - a. Early - Histodifferentiation
 - b. Advanced - Morphodifferentiation

The name of the stages is based on the shape of the epithelial part of the tooth germ that is epithelial enamel organ (Figs 3.6 to 3.28).

Bud Stage (Initiation)

Bud stage is the initial stage of tooth development (Figs 3.6 to 3.11). The basement membrane separates the epithelium of dental lamina from the ectomesenchyme (Fig. 3.5). Ten small, round or ovoid swellings develop superficial to the basement membrane called as tooth buds. Tooth buds are the precursors of enamel organs. The epithelium of the tooth bud forms the enamel. The epithelial cells do not show any change in shape and function. The supporting ectomesenchymal cells are densely packed under the lining epithelium and around the epithelial bud.

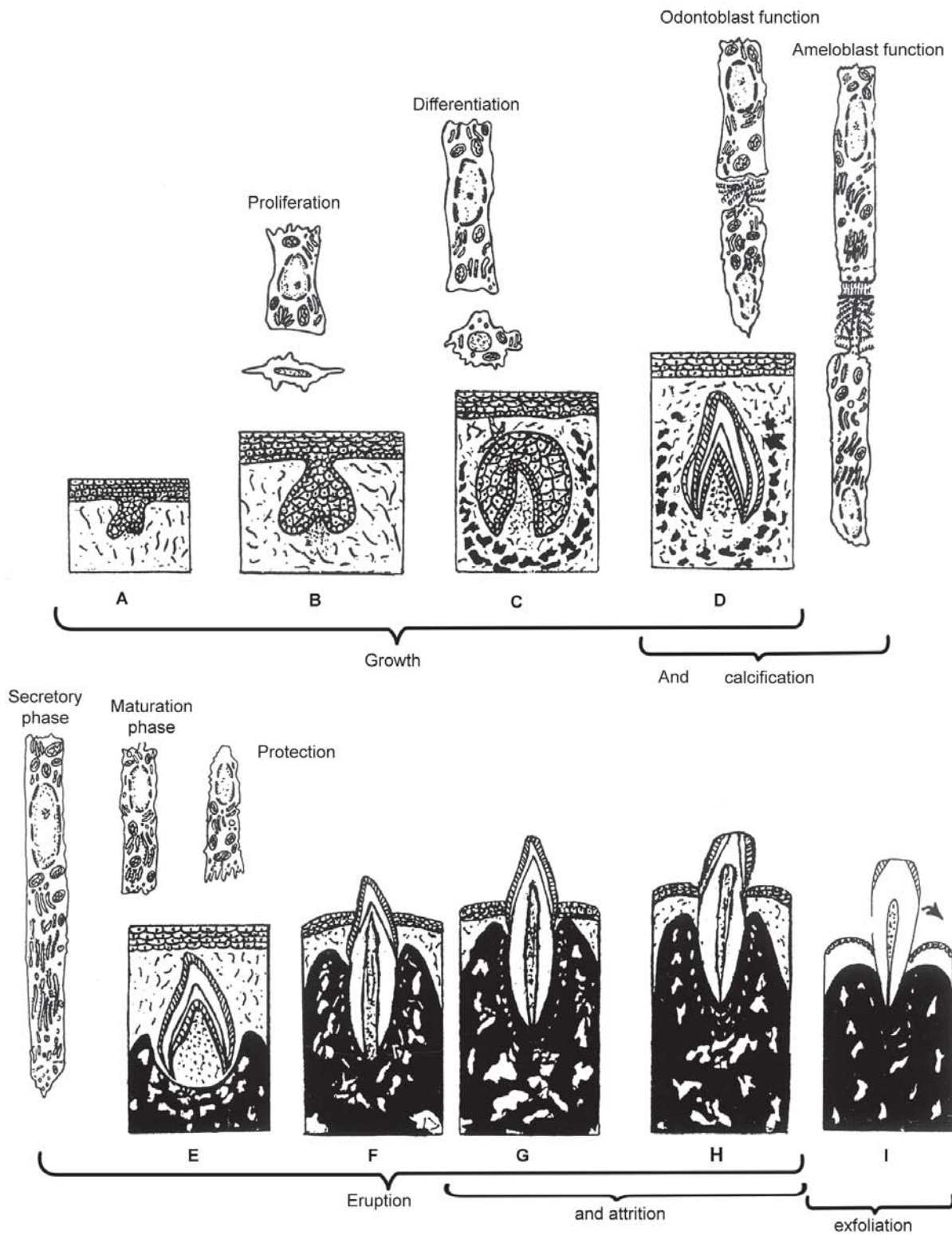
The enamel organ of bud stage contains two types of cells.

1. Polygonal cells, which are centrally situated
2. Low columnar cells, which are peripherally situated.

The ectomesenchyme of the enamel portion of the enamel organ is divided as a result of the increased mitotic activity. The centrally situated cells rapidly divide and grow and are condensed and form the dental papilla. Tooth pulp and dentin are formed from dental papilla. The ectomesenchyme that surrounds the tooth bud and dental papilla forms the dental sac. Cementum and periodontal ligament are formed from the dental sac.

Cap Stage (Proliferation)

The epithelial bud continues to proliferate into the ectomesenchyme. Immediately adjacent to the epithelial



Figures 3.9 A to I: Life -cycle of a tooth. A. Initiation (Bud Stage), B. Proliferation (Cap Stage), C. Morphodifferentiation, Histodifferentiation (Bell Stage), D. Apposition, E. Before eruption F. After eruption, G. and H. Attrition, recession of pulp, I. Weakening and loss of periodontal support leading to exfoliation

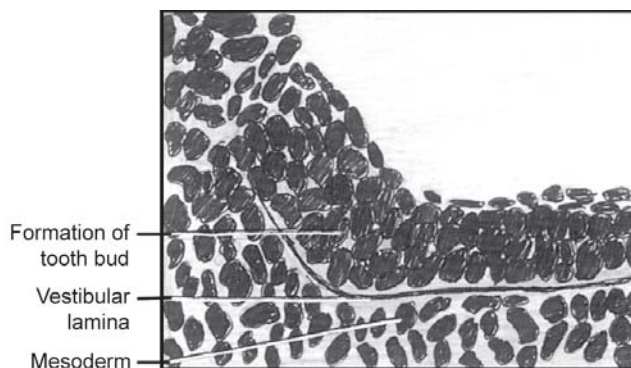


Figure 3.10: Tooth development - Bud stage (Proliferation stage). Embryo about 15 mm in length (sixth week). Cells of the bud stage under high magnification

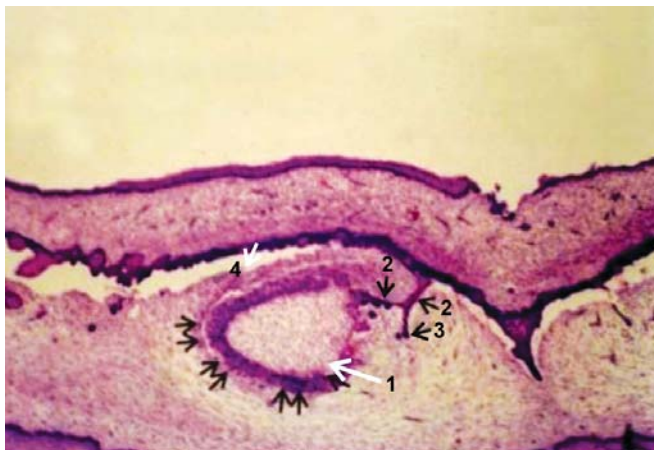


Figure 3.11: Low power photomicrograph showing earliest stage of tooth germ development: Bud stage in 14 weeks old fetus. Oval expanded tooth primordium or enamel organ (1) is seen arising from the dental lamina, (2) The enamel organ is surrounded by dense condensation of ectomesenchyme all around at this stage. (3) Successional dental lamina, (4) Primary epithelial Band, Condensed ectomesenchyme is indicated by double arrows. (H & E x 50)

ingrowth, the cellular density increases. This process is known as the condensation of the ectomesenchyme. When the embryo is 20 weeks old (180 millimeters), deciduous dentition is at various stages of development. After continuous division and differentiation, the size and shape of the enamel organ changes from knob-like to cap-like. This developmental stage is called as the cap stage (Figs 3.12 to 3.14). Invagination on the inner surface of tooth bud results from unequal division leading to cap stage. Cap stage is characterized by the outer and inner enamel epithelium and the stellate reticulum.

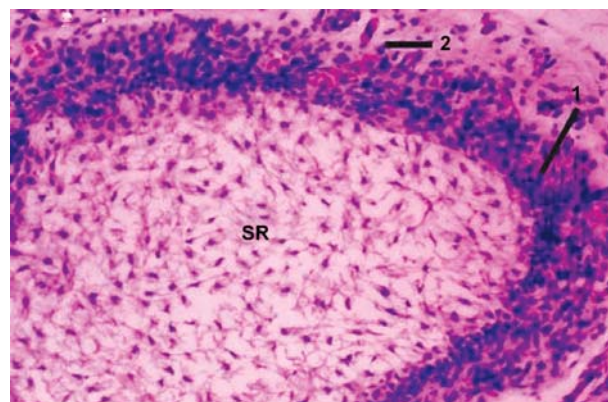


Figure 3.12: The center of enamel organ showing stellate reticulum. The cells are polyhyderal in nature and show separation from each other due to fluid collection and form a network. (1) Peripheral enamel epithelium, (2) Ectomesenchyme condensed around the enamel organ (H & E x 100)

Outer and Inner Enamel Epithelium

The cells of the outer enamel epithelium are cuboidal and cover the convexity of the cap whereas the cells of the inner enamel epithelium are tall, columnar and cover the concavity of the cap. Basement membrane separates the inner enamel epithelium from the dental papilla and outer enamel epithelium from the dental sac. Hemidesmosomes anchor the cells to the basal lamina.

Stellate Reticulum (Enamel Pulp)

Polygonal cells present in the center of the enamel organ between the inner and the outer enamel epithelium begin to separate by accumulation intercellular fluid. Osmotic force on the enamel organ is exerted due to presence of glycosaminoglycans contained in the ground substance. As a result, water is drawn into the enamel organ from the dental papilla and polygonal cells change into star shaped cells, forming a cellular network called the stellate reticulum. The proteinaceous fluid-containing albumin gives a cushion-like consistency to the stellate reticulum that supports and protects the delicate enamel-forming cells (Fig. 3.12).

Dental Papilla

The proliferating epithelium of the enamel organ exerts influence on the ectomesenchyme (neural crest cells) to proliferate. The ectomesenchymal cells are partly covered by the invaginated portion of the inner enamel epithelium.

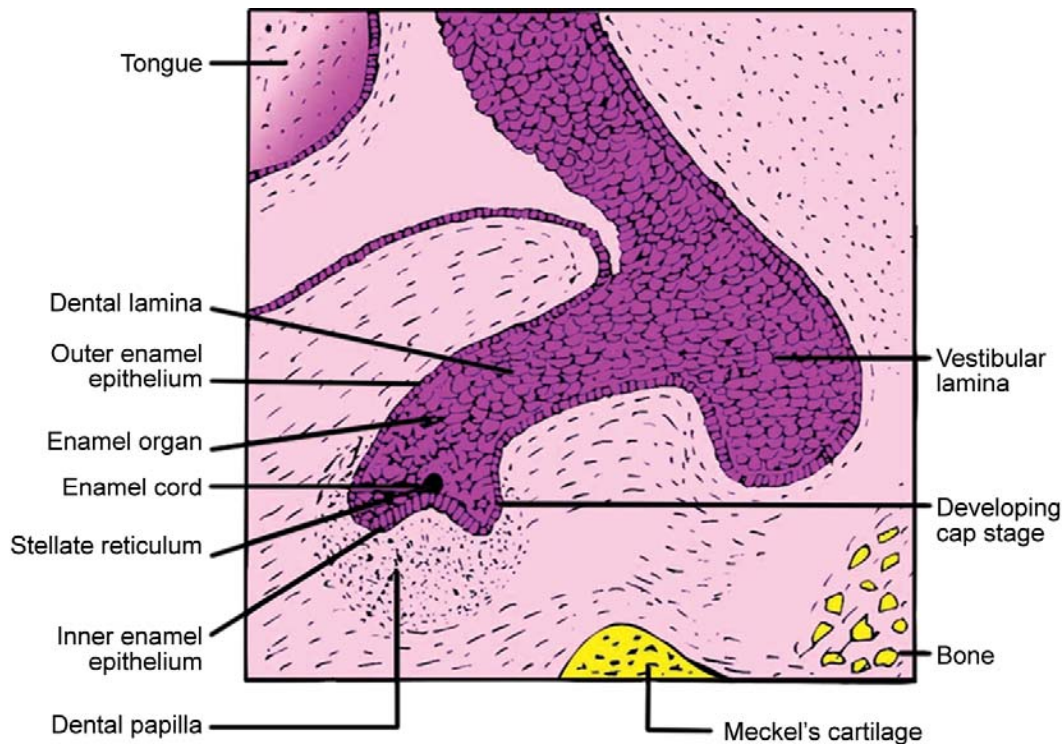


Figure 3.13: Labiolingual section of early cap stage of tooth development with surrounding structures

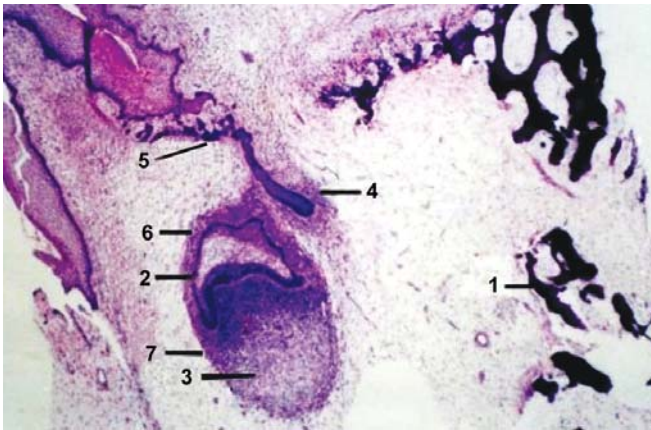


Figure 3.14: Cap stage of tooth germ development. The tooth bud of a deciduous tooth showing invagination of dental papilla (3) on the inferior aspect of enamel organ (2) giving rise to cap shape to the tooth germ. Successional lamina (4) with very early primordium of permanent tooth is growing posterior to the dental follicle (7). (5) Dental lamina, (6) Dental sac, 1= Alveolar bone. (H & E x 20).

On condensation, it is called as the dental papilla. It is the primordium of the pulp and is responsible for the formation of dentin and pulp. In dental papilla, mitotic cell division takes place along with proliferation of new

blood capillaries. The peripheral cells near the inner enamel epithelium increase in size and differentiate to form the odontoblasts.

Dental Sac (Dental Follicle)

Along with the development of enamel organ and the dental papilla, in their surrounding areas at the margins, cell division takes place resulting into condensation and fibrous development in this zone. This results in the formation of the dental sac. The cells of dental sac are responsible for the formation of the cementum and the periodontal ligament.

The entire tooth and its supporting structures are formed by epithelial enamel organ, the dental papilla and the dental sac.

Dental sac is the capsular structure consisting of circular arrangement of fibers (Fig. 3.15). These fibers with root development are differentiated into various types of periodontal fibers, which on one end are embedded in the alveolar bone and the other end in the developing cementum. During the development of the root end of tooth, these fibers are further embedded into the alveolar bone and cementum.

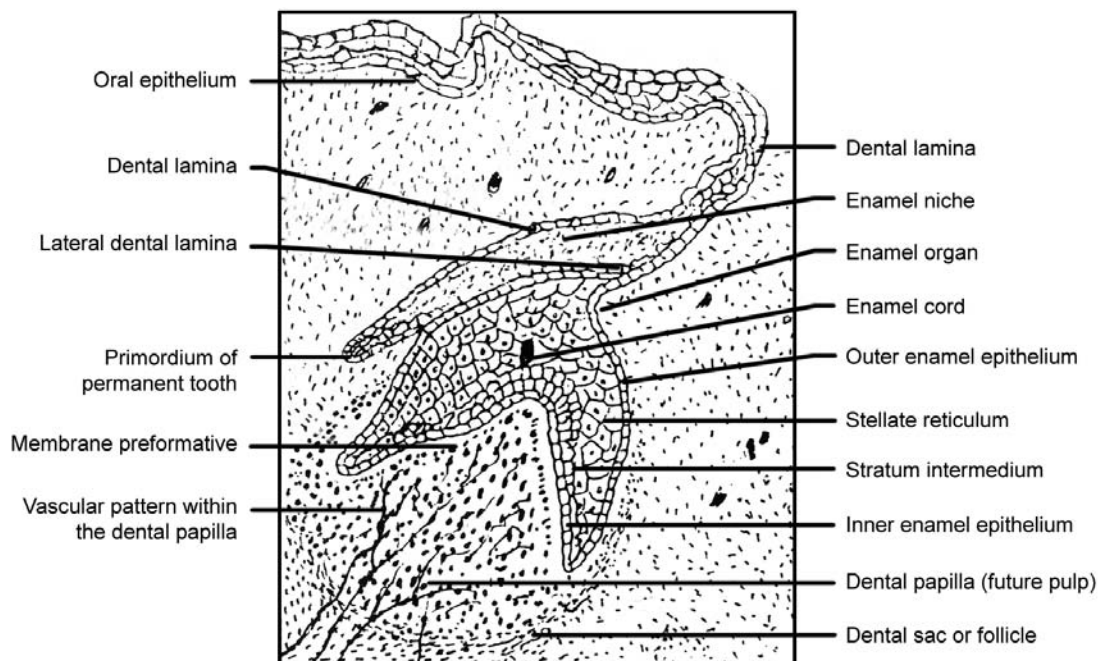


Figure 3.15: Labiolingual section of early bell stage of tooth development showing various structures. (Embryo CR. length 120 mm) 14 weeks, (weight 110 gm)

Bell Stage (Histodifferentiation and Morphodifferentiation)

With the division of the ectomesenchymal cells of inner portion of enamel organ and the deepening of the invagination of the epithelium, the margins continue to grow and the enamel organ assumes a bell stage (Figs 3.15 to 3.28). The developmental changes begin late in the cap stage. These changes continue during the transition of the tooth germ from cap stage to the bell stage. These changes are called as histodifferentiation. Similar epithelial cell mass transforms itself into a morphologically distinct component. The shape of the crown is determined in the bell stage and controlled by the genes, their signaling molecules and growth factors. The bell stage is characterized by the following four layers of epithelial cells (Fig. 3.18)

- a. Inner enamel epithelium
- b. Stratum intermedium
- c. Stellate reticulum
- d. Outer enamel epithelium

The development of teeth occurs in various developmental stages. Anterior teeth are at a more advanced stage than posterior teeth, as anterior teeth erupt earlier (Figs 3.19 and 3.20).

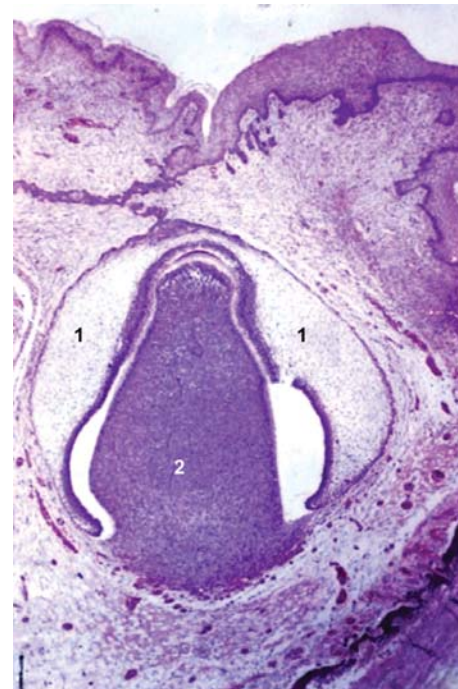


Figure 3.16: Bell stage of early advanced tooth germ development: Enamel organ shows (1) thinning at the apex of tooth and deep invaginations of cervical loops at the sides of dental papilla (2) resembling shape of a bell. Very early dentinogenesis and enamel formation are also seen (H & E x 20)



Figure 3.17: Developing teeth at the age of 14 weeks. Developing tooth; (1) at the bud stage of development (2) at the advanced bell stage of development; (3) dental lamina (4) oral epithelium; (5) dental ridge; (6) epithelial enamel organ; (7) dental papilla (9) primordium of permanent tooth successional tooth germ (bud stage) (10) bone of mandible (11) Hertwig's epithelial root sheath in the region of the root diaphragm

Inner Enamel Epithelium

Inner enamel epithelium consists of a single layer of tall columnar cells, which differentiate into specialized cells called ameloblasts, before amelogenesis (Fig. 3.21). These are characterized by high glycogen content. The diameter and length of ameloblasts is five microns and 40 microns respectively. Ameloblasts are attached to each other by junctional complexes and to stratum intermedium by the desmosomes. Inductive influences of ameloblast on the underlying ectomesenchymal cells of dental papilla result in development of odontoblasts (Fig. 3.21).

Stratum Intermedium

Stratum intermedium is the layer of squamous cells present in between the inner enamel epithelium and stellate reticulum (Fig. 3.22). These cells are intimately attached by desmosomes and gap junctions. They have a high degree of metabolic activity due to developed cytoplasmic organelles, acid mucopolysaccharides, glycogen deposits and an enzyme, alkaline phosphatase. Stratum intermedium is essential for the development of enamel because it contains new ameloblasts and is essential for the formation and calcification of enamel. It is absent in the root part of the tooth. Inner enamel epithelium and stratum intermedium are considered as a single functional unit.

Stellate Reticulum

The star-shaped cells of the stellate reticulum have long processes, which anastomose with the processes of adjacent cells. There is continuous expansion in the size of the stellate reticulum because of increased amount of intercellular fluid. Just prior to the beginning of enamel formation, at the height of the cusp or incisal edge, the stellate reticulum collapses and gets mixed up with the cells of the stratum intermedium. This decreases the distance between the ameloblasts, which are centrally situated, and the blood capillaries situated near the outer enamel epithelium. This change, which starts at the height of cusps shows gradual cervical progression (Figs 3.15 to 3.30).

Outer Enamel Epithelium

In the initial stages of development of enamel organ, the cells of the outer enamel epithelium are single-layered, and cuboidal in shape. Before enamel formation begins, outer enamel epithelium is folded. The capillary network develops in between the folds from the dental sac and provides a rich blood supply to the avascular enamel organ. This rich nutritional blood supply is required for the intense metabolic activity of the avascular enamel organ. The dental papilla is mesenchymal in nature. Under inductive influences of epithelium, the dental papilla develops the odontoblasts. The development of odontoblasts occurs and laying down of dentin starts before the inner enamel epithelium lays down the first layer of enamel matrix (Figs 3.21 to 3.24).

Dental Lamina

The dental lamina proliferates lingually at its deep end and gives rise to the enamel organ of the permanent teeth. It happens in all teeth except the permanent molars.

Dental Papilla

The dental papilla is covered by the enamel organ. The mesenchymal peripheral cells of the papilla differentiate into specialized cells called odontoblasts, which produce dentin. First, they are cuboidal-shaped and are then elongated to become columnar in shape and produce a thin layer of predentin. (Figs 3.23 and 3.24). Thereafter, the inner enamel epithelium produces enamel (Figs 3.20 and 3.27). The organizing influence of the epithelium helps the peripheral cells of the mesenchymal dental

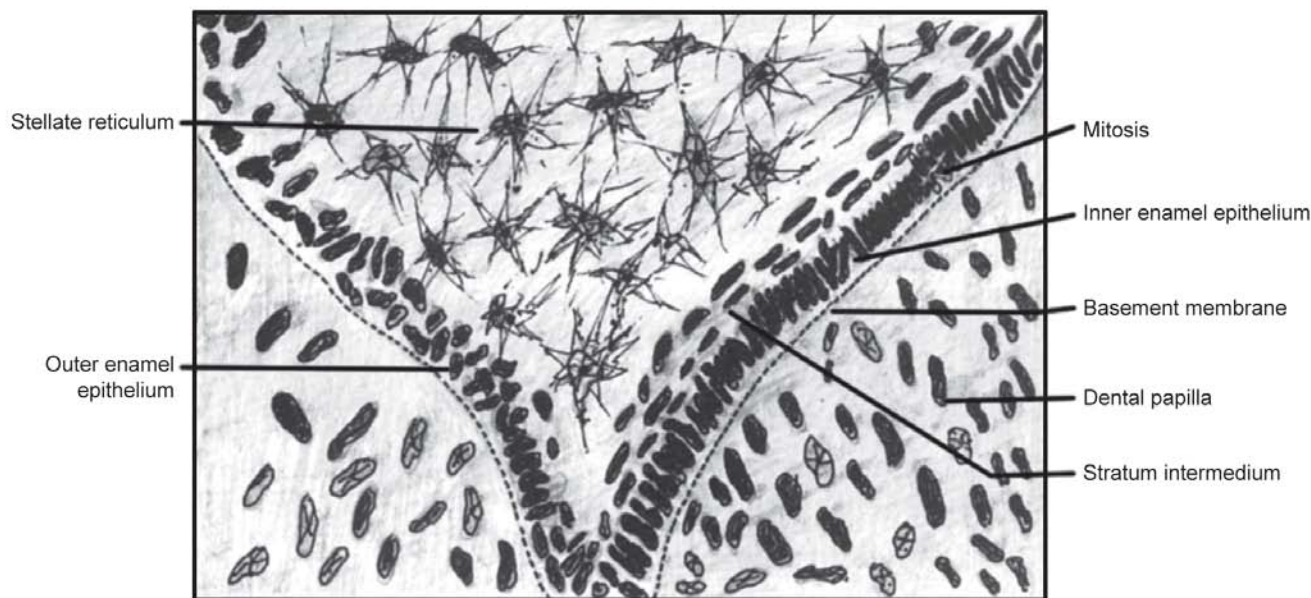


Figure 3.18: Various layers of epithelial enamel organ (under high magnification)

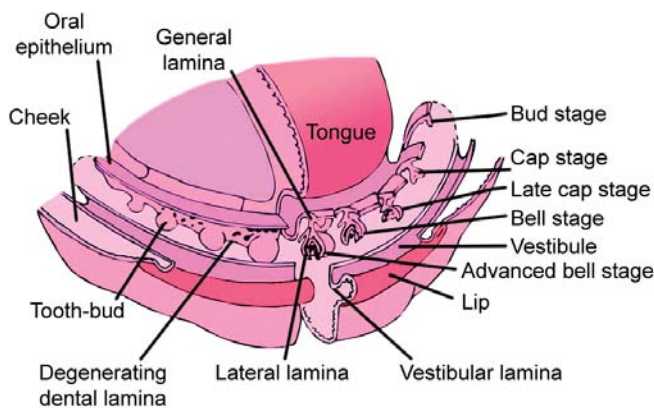


Figure 3.19: Development of deciduous teeth at different stages in alveolar process of 14 weeks old embryo (CR length 120 mm, weight 110 gm). Anterior teeth are at more advanced stages of development than posterior teeth as anterior teeth erupt earlier. Developing stages of teeth have been shown by longitudinal sections in the right half

papilla to differentiate into odontoblasts. These cells change shape from cuboidal to columnar. Membrana performativa is the basement membrane, which separates the enamel organ and dental papilla before dentin develops.

Advanced Bell Stage

In advanced bell stage, two more features of tooth development are also seen (Figs. 3.17, 3.27, 3.31 to 3.34) -

1. Future dentinoenamel junction - forms from the boundary present between the inner enamel epithelium and odontoblasts. The first layer of dentin is formed along the future dentinoenamel junction and formation proceeds pulpally and apically. After the formation of first layer of dentin, enamel is laid down over the dentin by the ameloblast and enamel formation proceeds occlusally.
2. Hertwig's epithelial root sheath - develops from the cervical portion of the enamel organ.

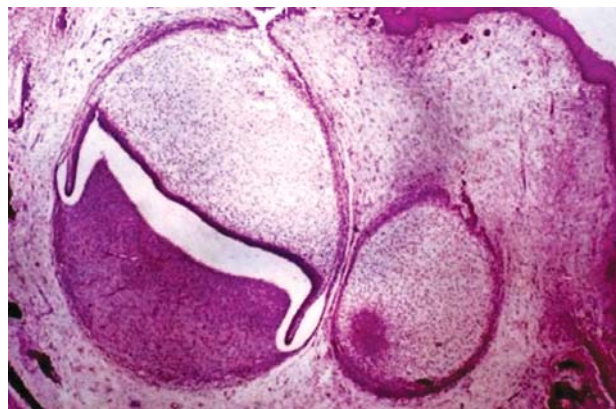


Figure 3.20: Multiple dental follicles in different stages of development in fetus (H and E x 20)

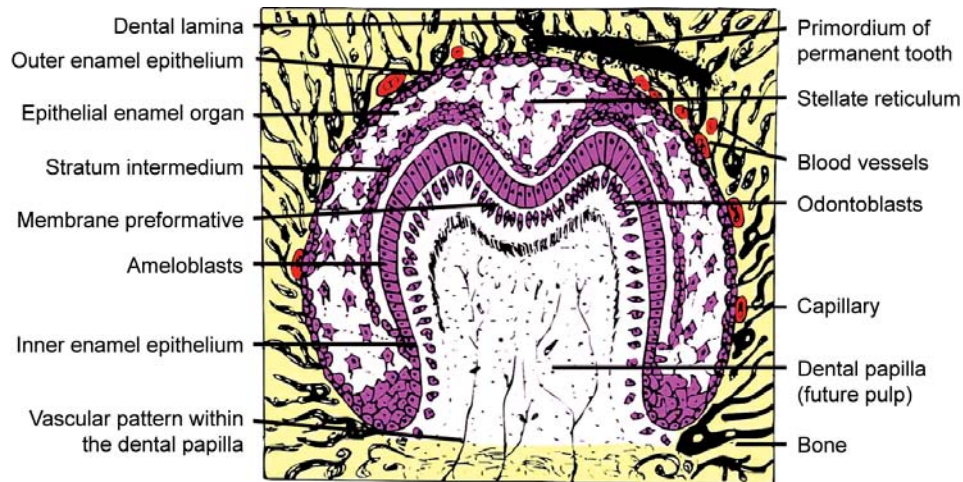


Figure 3.21: Bell stage: Labiolingual section through deciduous mandibular first molar tooth germ. Embryo about 160 mm CR length (18 weeks, fetal weight 320 gm).

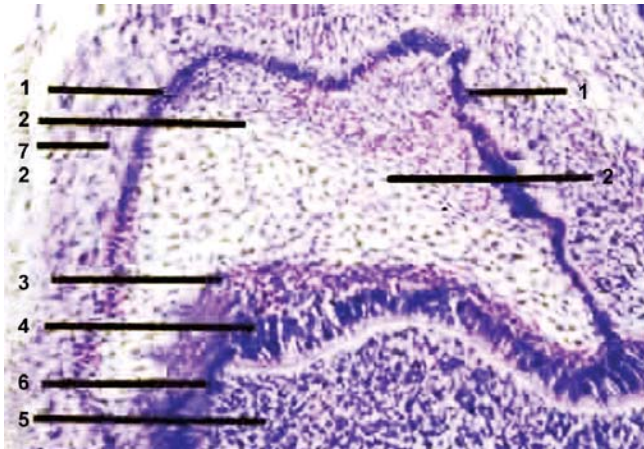


Figure 3.22A: Enamel organ showing well-developed stratum intermedium layer situated above the inner enamel epithelium (1) Outer enamel epithelium (2) Stellate reticulum (3) Stratum Intermedium (4) Inner Enamel Epithelium (5) Dental papilla (6) Basement membrane between enamel organ and dental papilla, (7) Condensation of ectomesenchyme around organ. (H & E x 100)

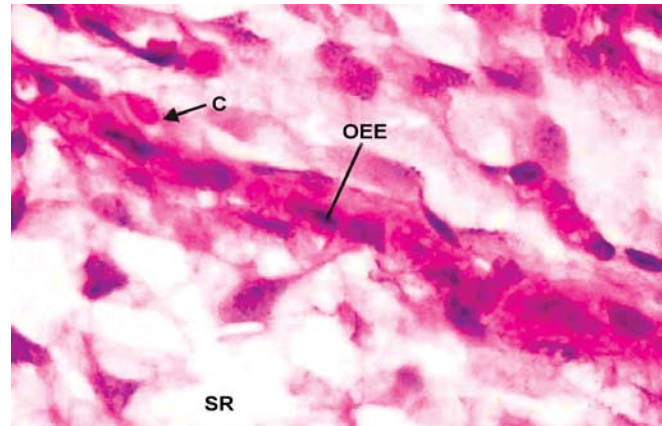


Figure 3.22B: Capillaries (C) are in intimate contact of cells of outer enamel epithelium (OEE). Stellate reticulum (SR). x 400

HERTWIG'S EPITHELIAL ROOT SHEATH AND ROOT FORMATION

After enamel and dentin formation has reached the future cemento-enamel junction the development of roots begins. The enamel organ forms Hertwig's epithelial root sheath (Root Sheath of Hertwig). Hertwig's epithelial root sheath is double layered. It contains only the outer and the inner enamel epithelia and not the stratum intermedium and stellate reticulum. The root sheath initiates formation of dental root and determines the number, shape, length and dimensions of the roots (Figs 3.31A and B, 3.32A and B).

The development of root does not begin till the enamel and dentin reach the future cemento-enamel junction. In development of root, enamel organ performs a very important function by forming Hertwig's epithelial root sheath. Hertwig's epithelial root sheath is molded and assumes tall shape of root by initiation of radicular dentin formation with the help of differentiation of radicular cells into the odontoblasts. Enamel on radicular portion is not formed because of the absence of stratum intermedium. The remnants of the epithelial root sheath are embedded in the periodontal ligament of erupted teeth and are called as epithelial rests of Malassez.

Before root formation starts, the epithelial diaphragm is formed by the root sheath. During root formation, epithelial

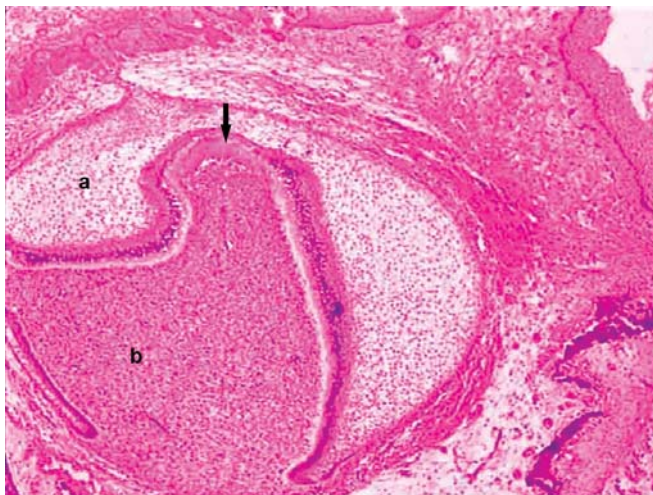


Figure 3.23A: Advanced bell stage of tooth germ development: Dental follicle with bell shape of the enamel organ (a) due to invagination by the dental papilla (b) very early dentinogenesis is indicated by the arrow $\times 40$

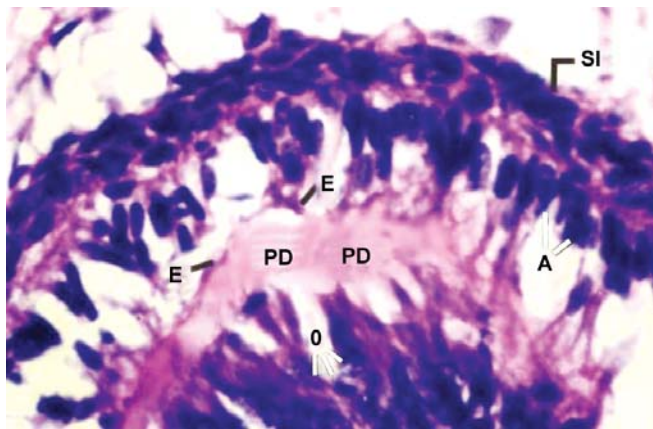


Figure 3.23B: Bell stage of tooth germ development: Very early dentinogenesis: High power view of dentin laid down by highly activated odontoblasts. (O) Odontoblastic processes are seen entering the predentin (PD) (pink matrix of dentin) which will later form dentin tubules. Mineralization of dentin has not started. The ameloblastic (A) layer is highly active with elongated cells. Nuclei are toward base of cells. Tiny specks of enamel (E) are being formed at the surface of dentin. Stratum intermedium (SI) is markedly reduced in thickness

diaphragm stays at about the same level. Towards the future cemento-enamel junction, the outer and inner enamel epithelium bends in a horizontal plane resulting in constriction of the wide opening of the tooth germ. As the development of the root continues, crown of the tooth moves occlusally and the plane of the diaphragm remains

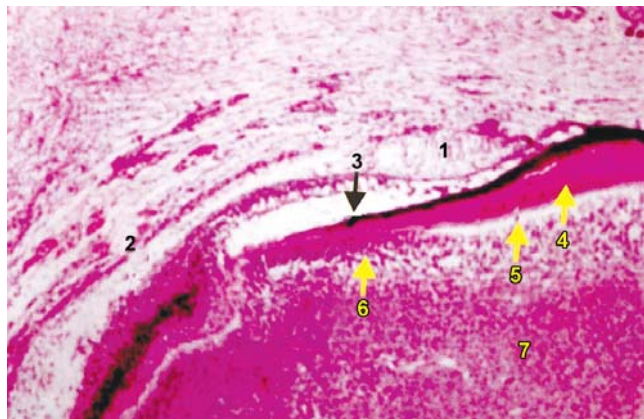


Figure 3.24: Tooth development: (1) Enamel organ, (2) Highly active multilayered secretory ameloblastic layer, (3) Enamel, (4) Dentin, (5) Predentin, (6) Columnar multilayered odontoblastic layer, (7) Dental papilla (H & E $\times 20$)

unchanged. The cells of the epithelial diaphragm proliferate along with the cells of the pulp. There is no growth of the free end of the diaphragm into the connective tissue but the epithelium proliferates coronal to the epithelial diaphragm. As the differentiation of the odontoblast continues, the root length increases towards the apex from the cemento-enamel junction. As the root length increases, the tooth moves occlusally. After a few layers of radicular dentin are formed, the continuous double layers of the Hertwig's epithelial root sheath are invaded by the proliferating connective tissue cells of the dental sac causing it to break up into a network of epithelial strands. This causes the epithelium to move away from the dentinal surface, causing the connective tissue cells to come in contact with the outer dentinal surface and differentiate into cementoblasts, which deposit cementum onto the dentinal surface. Because of this rapid sequence of proliferation and destruction of Hertwig's root sheath, it is never present as a continuous layer on the developing root surface (Figs 3.31 to 3.34).

During the last stages of root development, the growth of the epithelial diaphragm lags behind the growth of the pulpal connective tissue. In this way, width of the apical foramen is decreased first, then there is decrease in the width of the diaphragmatic opening. The opening of apical foramen is further decreased by apposition of dentin and cementum at the root apex.

In multirooted teeth, the shape of Hertwig's epithelial root sheath is different from that of the single rooted teeth (Figs 3.35 to 3.38). In multirooted teeth, the division of root trunk occurs in two or three roots. The cervical

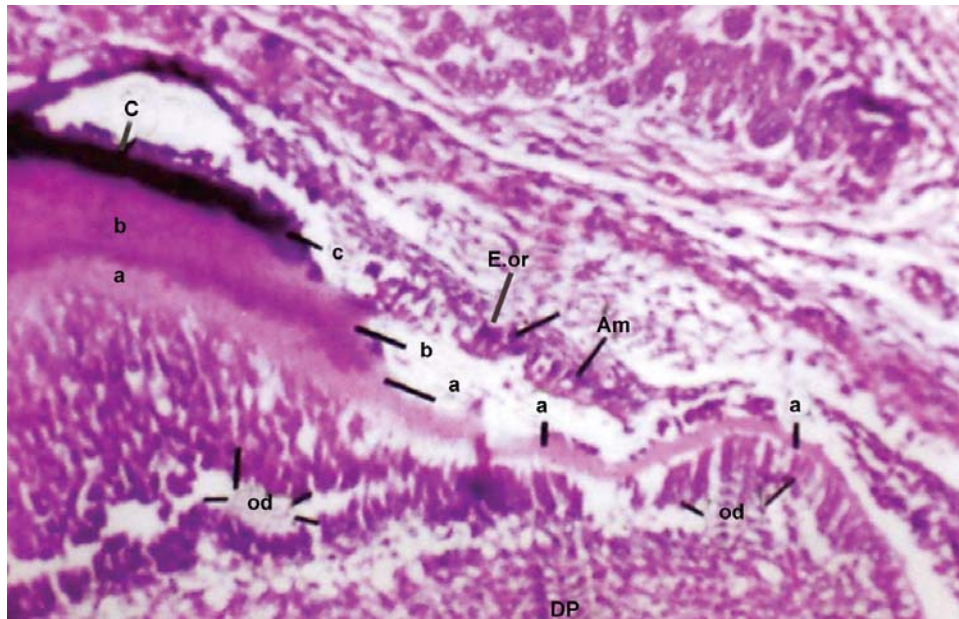


Figure 3.25: Tooth development: Various stages of dentin and enamel laying: First dentin is laid down by highly active tall columnar odontoblasts (od) at the top surface of dental papilla (DP). The odontoblasts have formed multiple layers of cells indicating their high proliferative activity. Initially, predentin (a) is laid down which then gets calcified forming mineralized dentin (b). Enamel (c) gets formed only after a small amount of dentin is laid down. Enamel organ (E or) gets atrophic but ameloblasts (Am) become active, tall and columnar. Tooth germ in 14 weeks old fetus (Undecalcified section H & E x 100)

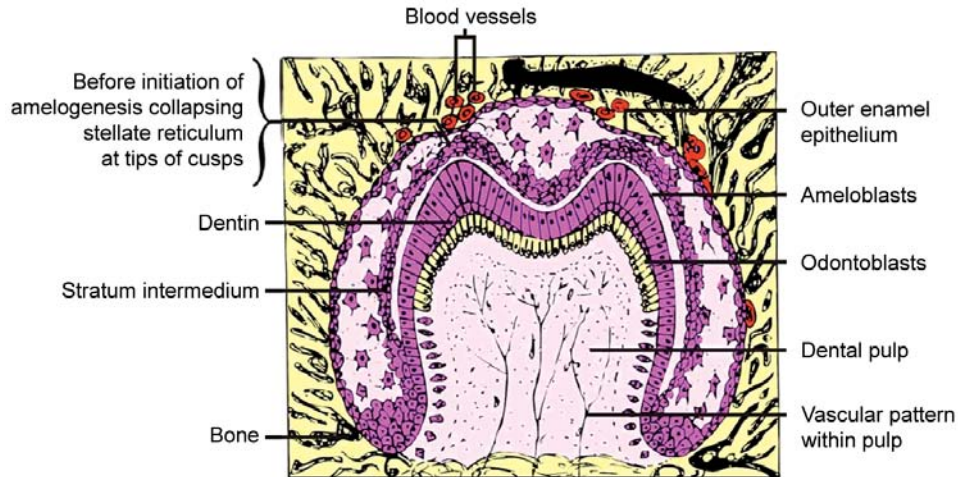


Figure 3.26: Advanced bell stage - Dentinogenesis (Appositional stage). Labiolingual section through deciduous mandibular first molar. Embryo about 190 mm in length (20 weeks, fetal weight 460 gm) showing dentin formation

expansion of enamel organ occurs in such a way that the horizontal diaphragm gives rise to tongue-like extensions, which grow inward. Two such extensions are found in the tooth germs of upper first premolars and lower molars and three such extensions are found in the tooth germs of

maxillary molars (Figs 3.35 to 3.38). Sometimes, the continuity of the Hertwig's epithelial root sheath is broken by the presence of small blood capillaries, leading to the development of accessory root canal openings from the pulp.

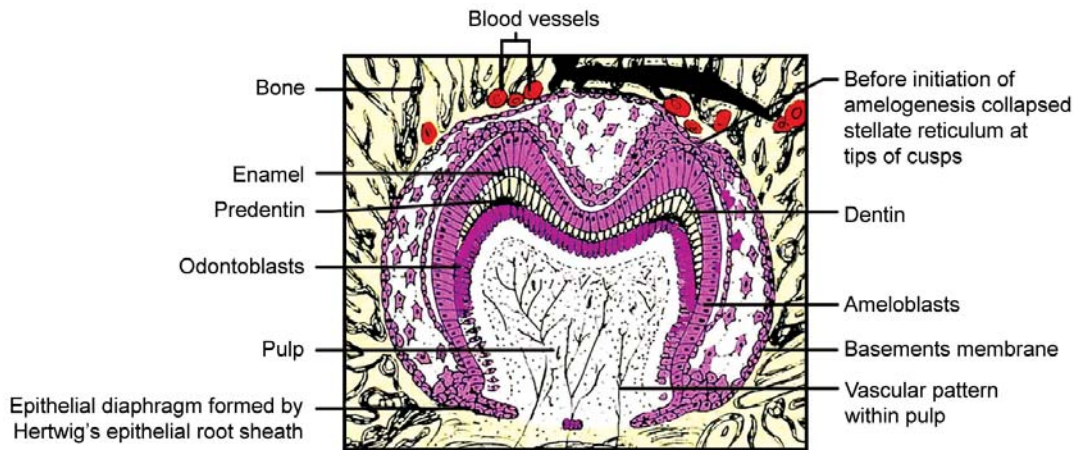


Figure 3.27: Very advanced bell stage (Dentinogenesis-Amelogenesis stage). Labiolingual section through deciduous mandibular first molar. Embryo about 210 mm CR length (22 weeks), fetal weight 630 gm showing formation of enamel, dentin and epithelial diaphragm. Outer enamel epithelium is laid in folds in which vascularity increases to provide nutrition to ameloblasts to form enamel

Enamel Pearl

Sometimes the cells of the Hertwig’s epithelial root sheath are differentiated and produce ameloblasts. Later, they are converted into droplets of enamel. Such type of droplets, which are adherent to the dentin are called enamel pearls. These are seen in roots of permanent molars, especially in the furcation area, during the development of roots.

Enamel Knot, Enamel Cord, Enamel Septum, Enamel Navel and Enamel Niche

In some of the tooth germs, following transient structures are observed. All these structures- enamel cord, enamel

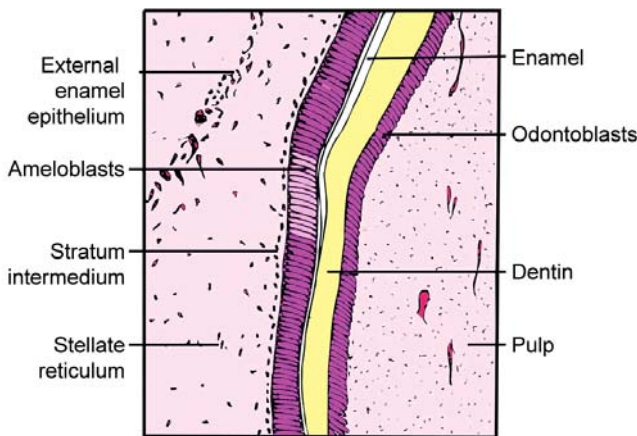


Figure 3.28: Region of a tooth germ at a very advanced bell stage: (Dentinogenesis-Amelogenesis stage) showing enamel and dentin formation (x150)

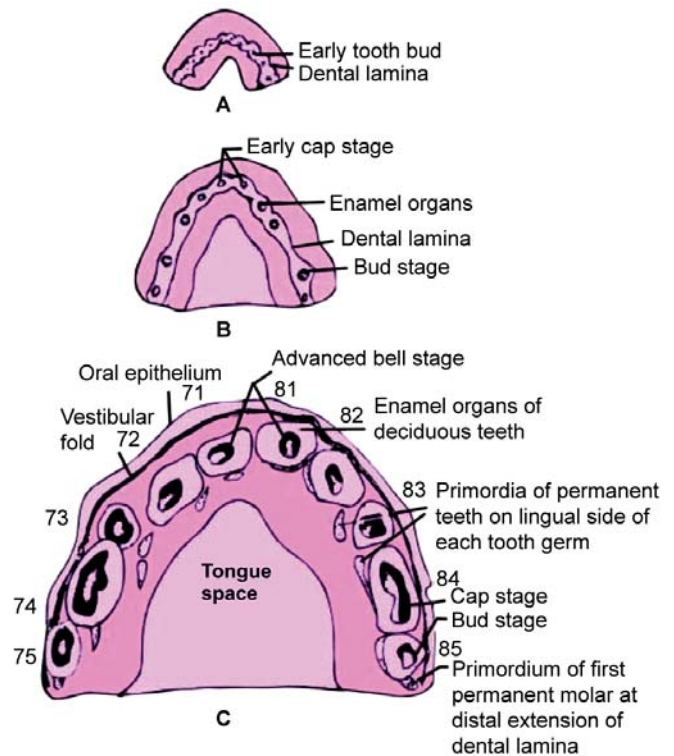


Figure 3.29: Development of deciduous teeth in mandible: A. 28 mm CR length (eighth week) embryo – early bud stages of all the teeth. B. 40 mm CR length (tenth week). Early cap stage in anterior teeth and bud stage in posterior teeth; C. 120 mm CR length embryo (14 weeks, weight 110 gm). Advanced Bell stage in anterior teeth and Cap stage in posterior teeth. 71 to 75 and 81 to 85 are developing deciduous teeth according to the FDI system of nomenclature. Primordia of permanent teeth are developing on the lingual side of deciduous teeth

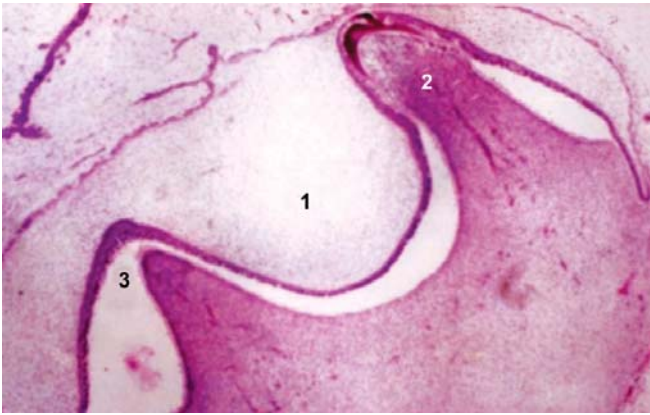


Figure 3.30: Tooth germ of a molar in 16 week old fetus. Two of the four cusps are clearly visible. One cusp (2) shows beginning of enamel laying down, while the other cusp (3) does not (1) Enamel organ (H & E x 20)

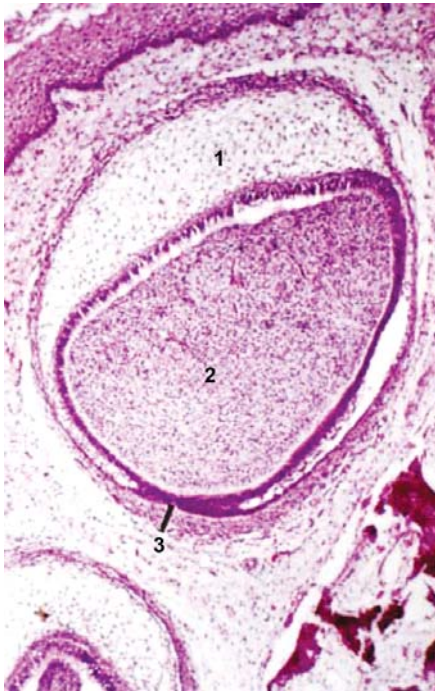


Figure 3.31A: Hertwig's epithelial root sheath (Root sheath of Hertwig): Bell stage of tooth germ showing formation of root sheath of Hertwig. (1) Enamel organ, (2) Dental papilla, (3) Formation of root sheath of Hertwig (H & E x40)

knot and enamel niche, are temporary and disappear before the beginning of amelogenesis. Probably their function is to act as a reservoir of dividing cells for the growing enamel organ.

Enamel Knot

Enamel knot is a localized thickening at the center of the enamel organ. The enamel knot slightly deepens into dental papilla. Because of this, a knob-like enlargement is seen around which labial and lingual grooves are present (Fig. 3.39).

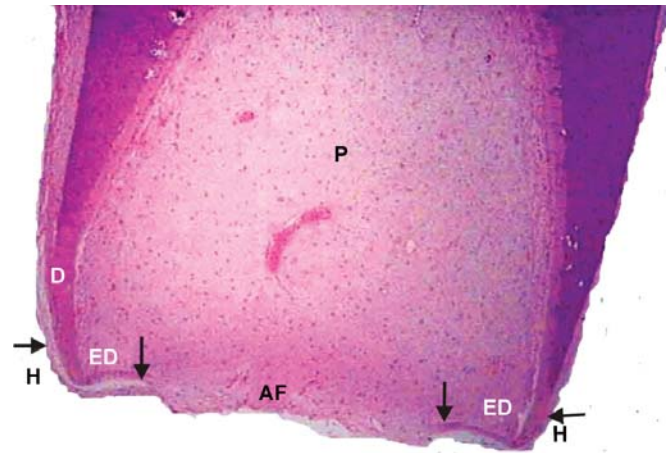


Figure 3.31B: Hertwig's epithelial root sheath envelops the dentin and is responsible for formation of dental root. It is formed by outer and inner epithelia of enamel organ. Epithelial diaphragm (ED) is formed by right angle bend of the epithelial cells column. Epithelial diaphragm is responsible for formation of apical foramen. Root sheath of Hertwig (H), Dentin (D), Pulp (P), Apical foramen (AF) x 40

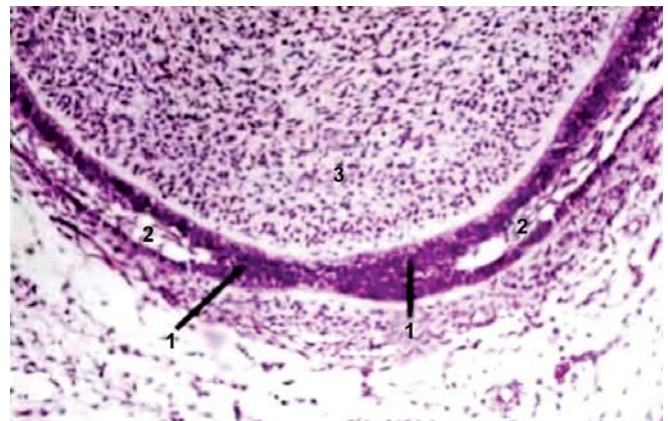


Figure 3.32A: Root sheath of Hertwig: The terminal portions of cervical loops in the Bell stage of tooth germ show loss of stellate reticulum. Outer and inner enamel epithelia come close to each other. (1) Root sheath of Hertwig, (2) Stellate Reticulum of cervical loops, (3) Dental papilla (H & E x 100)

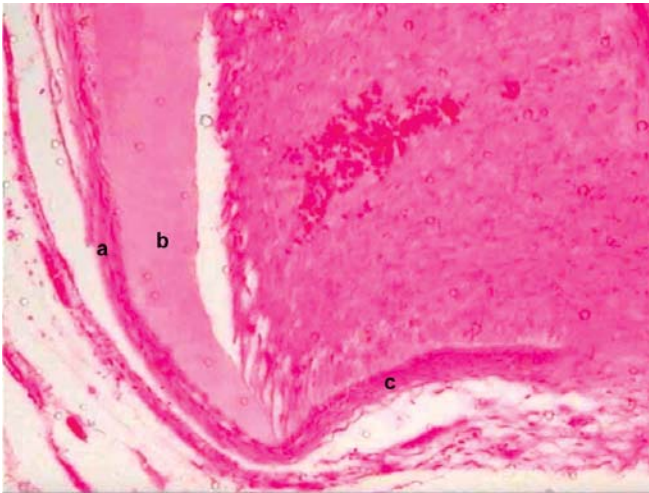


Figure 3.32B: Root sheath of Hertwig: There is a column of epithelial cells (a) which can be clearly traced from the distal part of the root outside the dentin (b). This column is bending at right angle to form the epithelial diaphragm (c) which takes part in the formation of apex foramen x 200

Enamel Cord

Usually the enamel knot is continuous with the enamel cord. Enamel cord is a continuous vertical strand of cells running from the knot to the external dental epithelium (Figs 3.15 and 3.39).

Enamel Septum

When the extension of enamel cord meet the outer enamel epithelium then it is called as enamel septum. It divides the stellate reticulum into two parts.

Enamel Navel

Small depression in the outer enamel epithelium at the meeting point of enamel cord is called as enamel navel. It resembles the umbilicus.

Enamel Niche

In histological sections, enamel niche is the apparent structure created by the plane of histological sections cutting through a curved dental lamina so that mesenchyme appears surrounded by the dental epithelium. This is created because the dental lamina is a sheet and not a single strand containing a concavity filled with connective tissue. Sometimes in histological sections it appears that the tooth germ has a double attachment to the oral epithelium by two separate strands (Fig. 3.15).

LIFE CYCLE OF A TOOTH

Life cycle is the complete cycle of a tooth from the beginning of development to eruption, arrangement, attrition and finally exfoliation of the tooth (Figs 3.9 A to I). The developmental stages of a tooth are described earlier in this chapter. The further stages of life-cycle are described as follows.

Eruption

Eruption is described as the emergence of teeth into the oral cavity. Every tooth in the dentition has a different eruption schedule. The deciduous mandibular central incisor is the first tooth to erupt into the oral cavity. The last tooth to erupt is the permanent maxillary third molar.

The factors that are responsible for eruption are the followings.

1. Vascular pressure exerted around and beneath the root.
2. Continuous growth of the root.
3. Bone remodeling
4. Traction of the periodontal membrane
5. Deposition of dentin
6. Narrowing of pulp
7. Functional movement of the muscles

Attrition

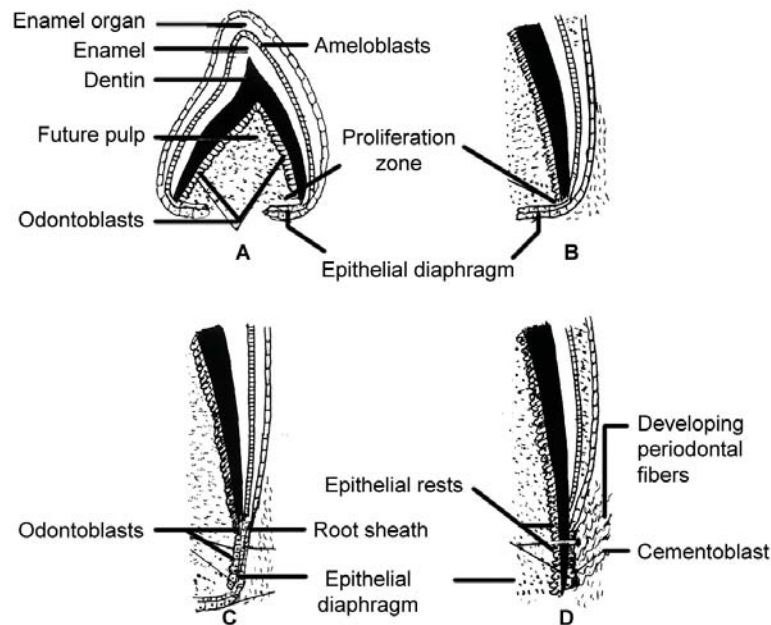
It is a physiological process characterized by wearing away of a tooth during tooth-to-tooth contact as in mastication. The surfaces involved are incisal, occlusal and proximal. Basically attrition is an aging process and it continues throughout life.

Regressive Changes of Pulp

Regressive changes of the pulp are characterized by a reduction in the cellular component of the pulp associated with a decrease in the number of odontoblasts. But the teeth are clinically symptomless and give normal response to the vitality test. As the age of an individual advances, the cellular component of the pulp decreases gradually and the number of odontoblasts is reduced.

Resorption of Teeth

Resorption of the teeth occurs under normal conditions. Resorption can occur on external surface or internal surface called as external resorption or internal resorption respectively. The most frequently involved part of the tooth



Figures 3.33 A to D: Stages in root development shown by section through tooth germ of mandibular canine A. Epithelial diaphragm and proliferation zone of pulp; B. Higher magnification of cervical region. Elongation of Hertwig's epithelial root sheath coronal to diaphragm. In elongated pulp differentiation of odontoblast is placed; D. Dentin formation has taken place in area of proliferation. Root sheath is broken up into epithelial rests and is separated from dentinal surface by connective tissues. Cementoblasts and developing periodontal fibers are also seen

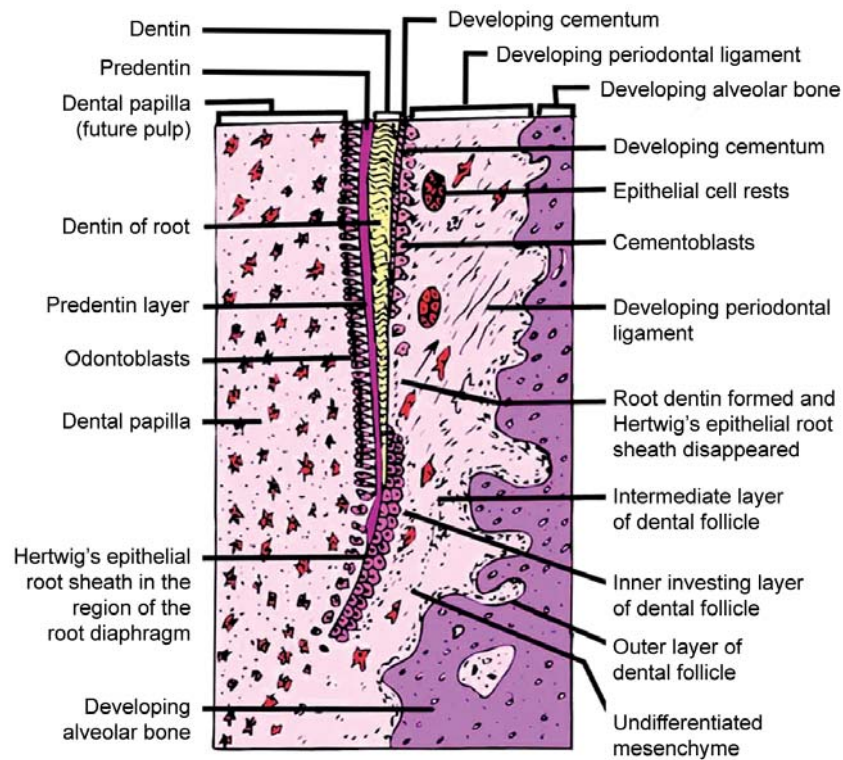
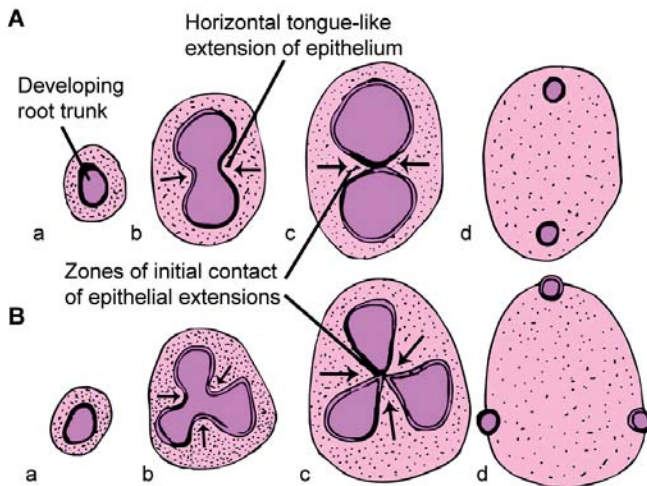
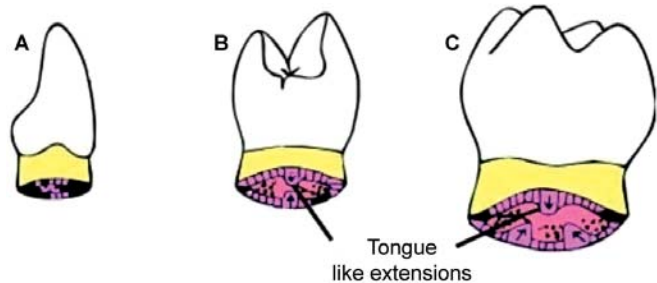


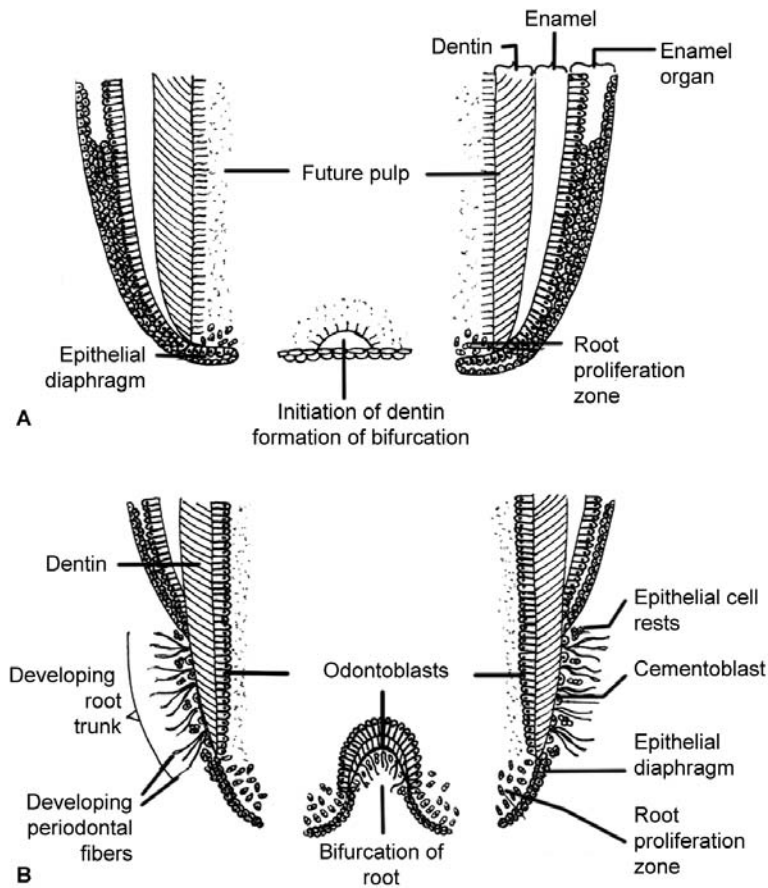
Figure 3.34: The structures of apical region of the developing root, periodontal ligament and alveolus



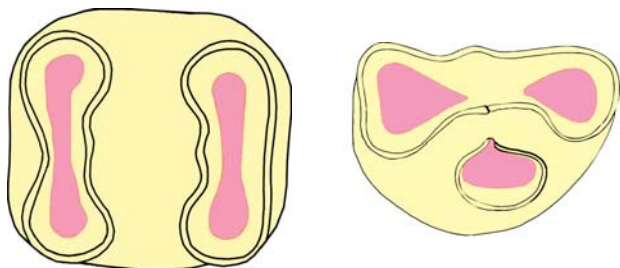
Figures 3.35 A and B: Multiple root development. Epithelial diaphragm horizontally extends and makes contact and fuse together to divide developing root into; A two or; B. three roots (a) developing root trunk; (b) Horizontal tongue like extensions of epithelium; (c) fuse together and vertically divide developing root into two or three roots; (d) root apices



Figures 3.36 A to C: The root formation: A single root; B. two roots; C. three roots



Figures 3.37A and B: Stages in development of root of two-rooted tooth. Diagrammatic mesiodistal section of lower molar; A. Initiation of dentin formation at bifurcation; B. Formation of two roots in progress



Figures 3.38 A and B: Stages of root development two years before eruption; A. Apices of developing two rooted tooth B; Apices of developing three rooted tooth

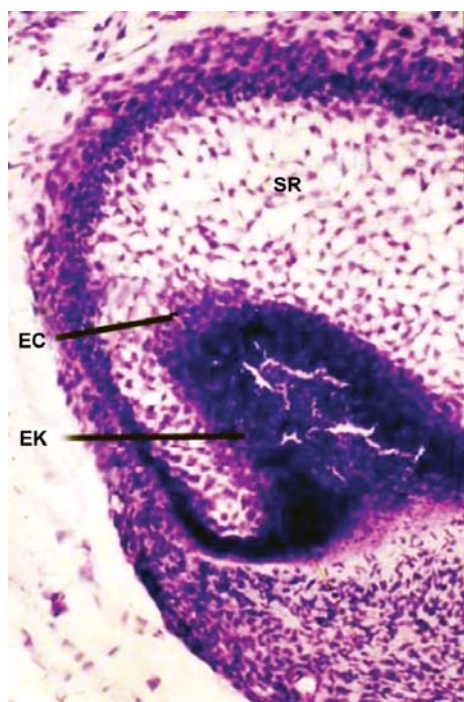


Figure 3.39: Cap stage of enamel organ showing enamel cord (EC) which is a tight column of cells extending vertically into stellate reticulum (SR) and arising from the enamel Knot (EK) (H & E x 100)

is the root and the intensity of resorption is mixed in nature. Generally, resorption starts from the cemento-enamel junction or at the site of the root apex.

Exfoliation of the Teeth [Shedding]

Exfoliation is the last stage in the life cycle of a tooth.

In deciduous dentition, shedding occurs because of the resorption of their roots. It is brought about by pressure exerted by the erupting permanent teeth. The factors which are responsible for the shedding of deciduous teeth are the following.

1. Pressure of the erupting permanent teeth
2. Activation of the osteoclastic and odontoclastic cells
3. Increasing masticatory forces due to development of the muscles leads to weakened dentition and helps in shedding.

Aging in Teeth and Periodontium

With aging, teeth show (a) attrition, resulting in reduced cusp height and inclinations (b) hypermineralization of calcified tissues (c) recession of pulp with fibrosis and (d) reduced number of cells and vascularity. In periodontium age changes are (A) gingiva show (a) reduced keratinization, (b) reduced vascularity (c) loss of elasticity and calcification of periodontal fibers (B) Loss of alveolar bone support to the tooth.

All changes in periodontium lead to gradual loosening and result in the exfoliation of teeth.

Histophysiologic Phases

There are a number of physiological growth processes that occur in the development of a tooth or odontogenesis. They are continuous and overlap with each other.

Initiation

Initiation or induction is a feature of the bud stage of odontogenesis. The cells present in the dental lamina have the capacity to develop enamel organ for the future development of teeth. The epithelial ectomesenchymal interaction is essential for initiation induction. The mechanism of such a process is not known (Figs 3.4 to 3.9). Different teeth are initiated at different times. Sometimes the teeth may develop in abnormal locations. The developmental stages in these cases are similar to the normal developmental stages.

If there is a lack of initiation, it results in the absence of either a single tooth or many teeth.

Abnormal initiation may result in the development of single or supernumerary teeth.

Proliferation

Proliferation is characterized by regular changes in the size and shape of the developing tooth germ. In this stage, the tooth germ has a strong affinity to differentiate into various structures. This is illustrated on the basis of the part of tooth germ, which continues to grow in tissue culture (Figs 3.10 and 3.14).

Histodifferentiation

Histodifferentiation shows maximum development in the early bell stage of tooth development. It succeeds the proliferative stage. During histodifferentiation, the formative cells of the tooth germ undergo definite morphologic as well as functional changes and acquire their functional roles. The cells differentiate and give up their capacity to multiply (Figs. 3.15 to 3.25). Certain interactions that occur between tissues during the development of a tooth are shown in Figure 3.40. In this stage, the peripheral cells of the dental papilla differentiate into odontoblasts producing dentin. This indicates that the presence of dentin is essential for the development of enamel, or in the absence of dentin, enamel does not form. The cells no longer multiply as they take up their new functions (Figs. 3.15 and 3.16).

Morphodifferentiation

Proliferation is essential for morphodifferentiation. The basic anatomic form of the tooth is established in the late bell stage after active histodifferentiation. In this stage, the dentinoenamel and the cemento-enamel junctions are developed by the continuous deposition of enamel, dentin and cementum from ameloblasts, odontoblasts and cementoblasts respectively, resulting in the establishment of a complete morphologic pattern, size and shape of a tooth (Figs. 3.26 and 3.27). If any disturbance occurs in morphodifferentiation, it affects the form and size of the tooth, but the functions of the ameloblasts and the odontoblasts are not affected.

Because of the disturbance, new parts may be differentiated, or a suppression of the part may occur, or a peg-shaped or malformed tooth may form, or a central incisor with a notch at the edge may form.

Apposition

After the morphologic pattern of a tooth is established, an additive growth of the hard dental tissues occurs. Apposition is characterized by the rhythmic, layer-like deposition of an extracellular matrix of enamel and dentin. The pattern of apposition of enamel, dentin and cementum is different, and it will be described separately in respective chapters. Appositional growth is characterized by regular and rhythmic deposition of the extracellular matrix. Periods of activity and rests are present during tooth development. These alternate one after the other.

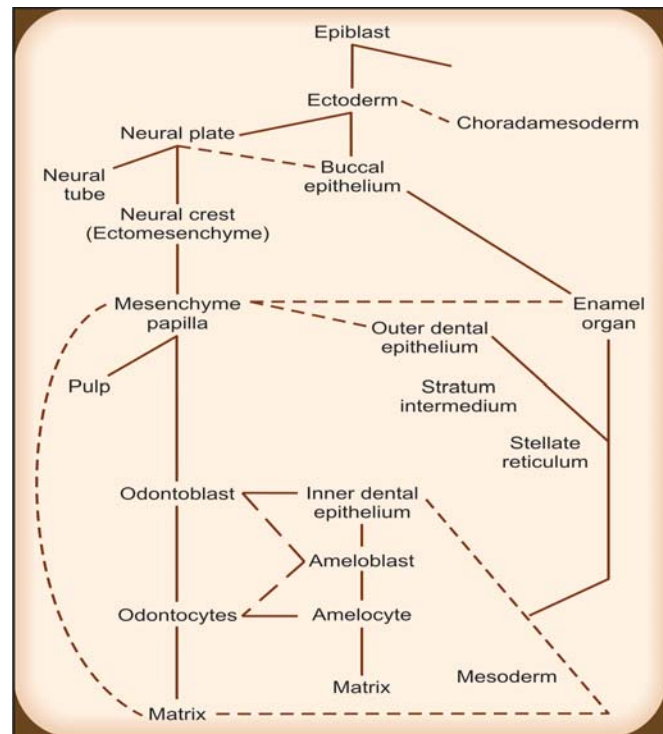


Figure 3.40: Outline of development of tooth. Broken lines – known or suspected interactions that occur between tissues. Data suggesting placement of these lines derive from transplantations and in vitro studies. Words “amelocyte” and “odontocyte” are employed only to indicate that these cells may possess different capabilities for interaction with other tissues after their overt differentiation (Courtesy: Dr. Koch William E., Chapel Hill, NC.)

There may be some genetic and also environmental factors that disturb the normal synthesis and secretion of organic matrix of enamel. This may lead to enamel hypoplasia.

CLINICAL CONSIDERATIONS

1. Sometimes, the teeth may develop in abnormal locations other than in the oral cavity, for example in the ovary and in the hypophysis.
2. In partial anodontia or oligodontia only few teeth are present in the oral cavity. Partial anodontia is characterized by the lack of initiation of teeth, for example, permanent maxillary lateral incisors, third molars and mandibular second premolars may be missing. Anodontia is complete lack of tooth.
3. Supernumerary (single or multiple) or additional teeth may also develop due to abnormal additional initiation. Most commonly the mesiodens is seen between the maxillary central incisors.

4. Osteodentin is an atypical dentin that develops due to deficiency of vitamin A. The ameloblast is not differentiated properly, as a result the proliferation of mesenchymal cells is disturbed.
5. The eruption of teeth is delayed in persons having hypopituitarism and hypothyroidism. Small clinical crown but normal anatomic crown is seen in these type of patients.
6. Hutchinson's incisor is seen in the patients suffering from congenital syphilis. It is characterized by "screw driver" or notched edge of maxillary central incisor. Mulberry molars are also seen in patients of syphilis due to disturbed morphodifferentiation stage.
7. Enamel hypoplasia is a condition in which there is disturbance in the synthesis and secretion of organic matrix of enamel. Two main factors are responsible for this condition:
 - Genetic
 - Environmental

If the calcification is defective and organic matrix is normal, then the condition of enamel or dentin is said to be hypocalcified or hypomineralized. Both conditions happen when the apposition stage of tooth development is disturbed. Splitting of one tooth germ result in a production of two similar teeth and union of two tooth germ together before mineralization results in a fused teeth.

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Enamel

- Introduction
- Physical properties
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 - Rod sheaths
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 - Other structures
- Age changes
- Development
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 - Outer enamel epithelium
 - Stellate reticulum
 - Stratum intermedium
 - Inner enamel epithelium (ameloblastic layer)
 - Cervical loop
- Life cycle of ameloblasts
 - Morphogenic stage
 - Organizing and differentiating stage
 - Formative stage
 - Maturative or mineralizing stage
 - Protective stage
 - Desmolytic stage
- Amelogenesis (enamel formation)
- Clinical considerations

INTRODUCTION

Enamel is an ectodermal derivative and is the most highly mineralized tissue known. It is the hardest tissue of the body, which covers entire surface of the anatomical crown of all the teeth. Enamel provides shape and contour to the crown of teeth. It is also known as Substantia adamantia.

Enamel is composed of bending rods, which in turn are composed of crystals. Minute spaces or gaps exist where crystals do not form between rods. This causes enamel to be variable in density and hardness. So some areas of enamel may be more prone to penetration by small particles. This characteristic leads to tooth destruction by dental caries. After enamel is completely formed, no more enamel can be deposited. Final shaping of the cusp is performed in enamel. Enamel forms strong and suitable covering on the anatomical crown of the teeth.

PHYSICAL PROPERTIES

Enamel is the most highly mineralized tissue of the body. It forms an outermost protective covering of variable thickness over the anatomic crown of the tooth. Mature functional enamel is the hardest mineralized epithelial tissue of the body. Because of its high mineral content and crystalline arrangement, enamel is brittle and subject to fracture by trauma or heavy sharp-edged forces. Other hard tissues like dentin, bone, cementum and cartilage also are mineralized connective tissues but mature enamel is the only tissue that is totally acellular.

An important physical property of enamel is its permeability, which has been demonstrated by radioactive tracers and dyes. It acts like a semi-permeable membrane, which permits complete or partial passage of certain molecules like ^{14}C - labelled urea, ^{131}I , etc. Enamel is permeable to some fluids, bacteria and their products.

The enamel attains a maximum thickness of 2.5 mm on the cusp of crown and a minimum of about 100 microns at the neck of the tooth and bottom of the pits and fissures. The hardness of enamel enables it to withstand the blunt, heavy masticatory forces. The nature of its structure and the hardness render it very brittle having a low tensile strength. The color of the enamel varies from bluishwhite to yellowish or grayish white. The color is determined by differences in the translucency and thickness of enamel. The translucency is affected by variations in the degree of calcification and homogeneity of the enamel. Thinner areas of the enamel appear yellowish as the underlying

yellowish dentin is visible through the enamel, and where thickness of enamel is more, it appears slightly grayish. The specific gravity of enamel is 2.8. The density of enamel is 2.8 to 3 g/ml. It has a Knoop hardness number (KHN) of 343 while dentin has 68 KHN and cementum has 40 KHN. Enamel resists masticatory impact of about 10 to 20 kg per tooth.

Hydroxyapatite crystal of enamel is 300 times larger than dentin.

CHEMICAL PROPERTIES

Enamel is composed of both inorganic and organic substances. Mature enamel is a highly mineralized crystalline structure, containing by weight 96 percent inorganic matter and 4 percent organic matter and water. The inorganic content is mainly hydroxyapatite crystal and is present 92 to 98 percent by volume of total inorganic matter. Other mineral elements such as strontium, magnesium, lead and fluoride are present in very small amounts. The remaining constituents of enamel are organic contents, mostly tyrosine rich amelogenin protein and non-amelogenin proteins and water; these total approximately 4 percent by weight.

The organic content forms a fine network between the inorganic crystals. Enamel is composed of the same mineral crystals that are present in dentin, cementum and bone. Like bone, cementum and dentin, mineral crystals of enamel are not replaced once deposited. By volume, organic matter is equal to inorganic matter. The relative space occupied by the entire enamel and the organic framework is almost equal. By weight, inorganic matter is 24 times heavier than the organic matter.

During development of enamel, the staining reactions of enamel matrix for histological study resemble keratinizing epidermis. The specific studies have shown sulfhydryl groups and keratin. Protein that contains a high percentage of serine, glutamic acid and glycine is isolated. By radiological diffraction studies, it has been shown that the molecular structure of proteins is like cross-beta proteins. Histochemical studies of developing teeth have revealed that the enamel forming cells (ameloblasts) also contain a polysaccharide-protein complex. When calcification becomes a prominent feature, acid mucopolysaccharide enters the enamel itself. Enamel of erupted teeth can transmit and exchange radioactive isotopes originating from saliva and the pulp. The organic

components of enamel are the proteins amelogenins and amelins, which are two major classes of calcium binding proteins. In enamel there are two main groups of proteins (A) amelogenins (90%) and non- amelogenins (10%). Important proteins of nonamelogenin group are (a) enamelin (b) ameloblastin and (c) tuftelin. During enamel maturation, amelogenins disappear at a much faster rate than amelins. Amelins have a very high affinity for binding apatite crystals. They are similar to the protein keratin that is found in the skin. The even distribution of enamelin between and on the rods helps in permeability of enamel.

STRUCTURE

Enamel is composed of the following.

1. Enamel rods (prisms)
2. Rod sheaths
3. Inter rod substance (cement)

Enamel Rods

Structurally enamel is composed of millions of enamel rods or prisms. An enamel rod is a long, thin structure extending from the dentinoenamel junction to the surface of enamel. The enamel rod follows a tortuous course; thus the length of an enamel rod may be greater than the thickness of enamel. Each rod is formed by four ameloblasts. One ameloblast forms the rod head, a part of two ameloblasts form the neck, and the tail is formed by a fourth ameloblast. Each ameloblast contributes to four different rods (Figs 4.1 A and B).

Human enamel is composed of rods that in the transverse section are shaped with a rounded head or body section and a tail section. The rounded portion of each prism lies between the tail of two adjacent prisms. Such interlocking arrangement provides enamel with additional strength and stability. The heads or bodies of the rods are closer to the occlusal or incisal surface, while the tail points in the cervical direction (Fig. 4.2). In a cross-section of human enamel, many rods resemble fish-scales (Figs 4.3 and 4.4).

Direction of rods In general, the rods are directed at right angles to the dentinoenamel junction and the tooth surface. In cervical regions of deciduous and permanent teeth, the directions of enamel rods are different (Fig. 4.5).

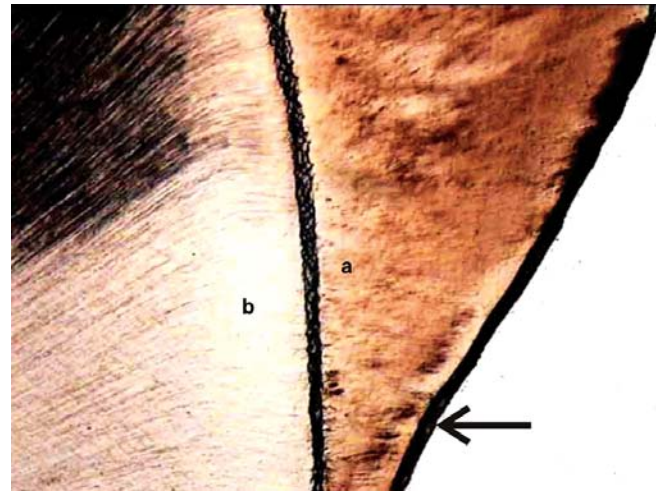


Figure 4.1A: Reducing enamel thickness: Ground section of tooth showing the taper of enamel (a) from the cuspal to cervical region (indicated by arrow). Dentin (b) is separated from enamel by dentino-enamel junction. x 40

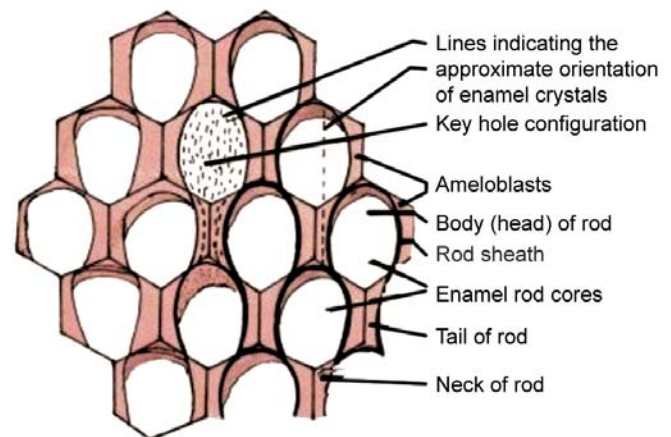


Figure 4.1B: One interpretation of the relationship between the keyhole-shaped enamel rods and the roughly hexagonal ameloblasts. According to this interpretation, each rod is formed by four ameloblasts and each ameloblast contributes to four different rods

In permanent teeth: At the central part of the fossa and pits, the rods turn oblique to nearly horizontal. Near the tip of the cusp and incisal edge, the rods gradually change to increasingly oblique direction. Gradually they become nearly vertical at the tip of the cusp and incisal region. In the cervical region, the rods run slightly apically from dentin surface to outer enamel surface (Fig. 4.5A).

In deciduous teeth: The arrangement of rods in the occlusal two-thirds in deciduous teeth is similar to that in permanent teeth. At the cervical and central part of the fossa and pit, they are nearly horizontal (Fig. 4.5B).

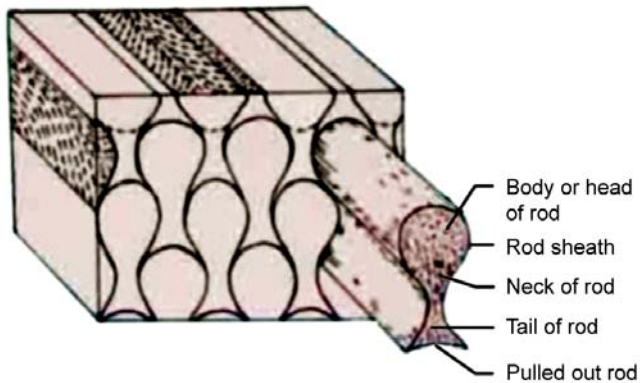


Figure 4.2: One enamel rod is pulled out to show individual rod interdigitation with neighboring rods. Small dashes show the crystal orientation in the enamel rod

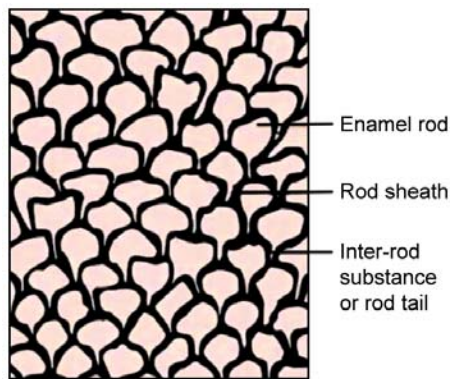


Figure 4.3: Enamel showing the characteristic fish-scale appearance as seen in etched ground section

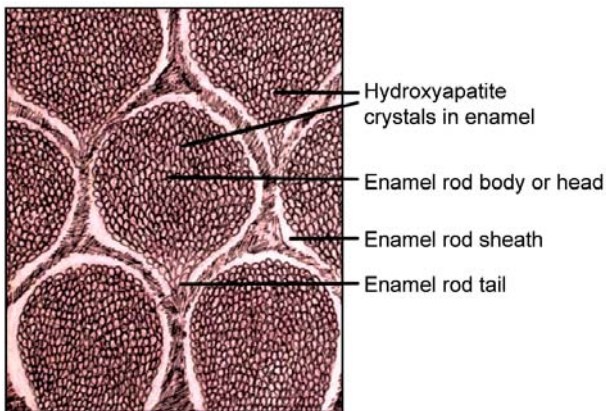


Figure 4.4: Transverse section of enamel rods showing keyhole-shaped rods divided into two parts, head (body) and tail

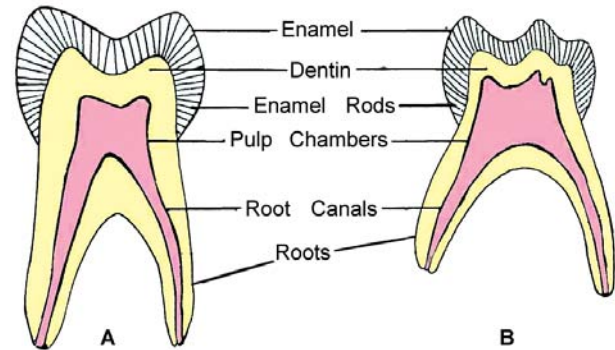


Figure 4.5: Mesiodistal section of; A. Permanent, B. Deciduous maxillary second molars showing direction of enamel rods, pulp chambers and pulp canals

Submicroscopic structures The enamel rods have an average diameter of 4 microns. The diameter of the rod increases from the dentinoenamel junction towards the outer surface of enamel in a ratio of about 1 : 2. The number of enamel rods has been estimated as ranging from 5 million in the lower lateral incisor to 12 million in the upper first molar. The rods are larger at cusp tips and shorter at the cervical region.

The head or body of the enamel rod is the broadest part and is about 5 microns wide, and the elongated thinner portion called as tail is about 1 micron wide. The rod, including both head and tail, is about 9 microns long. The enamel rod is about the same size as a red blood cell. The 'heads' or bodies of the rods are nearer to the occlusal surfaces and 'tails' are directed cervically. (Figs 4.6 A and B to 4.9).

Striations The enamel rod consists of segments. These segments are separated by dark lines. These dark lines give enamel a striated appearance. The rods are segmented because the enamel matrix is formed in a rhythmic manner.

The enamel rods normally have a clear crystalline appearance, permitting light to pass through them. Each rod is filled with crystals, that is, it is made up of numerous apatite crystals. These crystals are irregular in shape. The crystals in head follow the long axis of the rod and those in tail lie in cross axis to the head. In cross-section, the enamel rods appear as round, hexagonal or oval. Recent studies with electron microscope have shown that a more common pattern of enamel rods in cross section looks like a keyhole or paddle-shaped prism (Figs 4.3 and 4.4).



Figure 4.6A: Enamel: Ground section of tooth showing structure of enamel which is made up of long thin rods, which are not straight but pursue a tortuous course x 200



Figure 4.6B: Electron micrograph of cross-section of enamel showing keyhole pattern arrangement of enamel rods or prisms (ER) x 900. RS= Rod sheath RT= Rod tail or inter-rod substance

The crystals are arranged approximately parallel to the long axis of the prisms, although deviations of up to 40 degrees have been observed. Crystals that are more peripherally placed flare laterally about 65 degrees as they approach the periphery and tails of the prisms. The

enamel crystals have an average thickness of about 30 nanometers, an average width of about 90 nanometers and 0.05 to 1 micron in length (Fig. 4.11).

Each apatite crystal is composed of thousands of unit cells that have a highly ordered arrangement of atoms. Each crystal is enveloped in an organic matrix. The basic crystallographic formula for the unit cell crystal of hydroxyapatite is $3\text{Ca}_3(\text{PO}_4)_2 \cdot \text{Ca}(\text{OH})_2$. The formula of calcium deficient polycalcified hydroxyapatite crystal is $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$. In some areas, especially near the cervical region and near the incisal and occlusal region, the rods become more twisted. Such areas are associated with an increased strength of enamel. The rods are

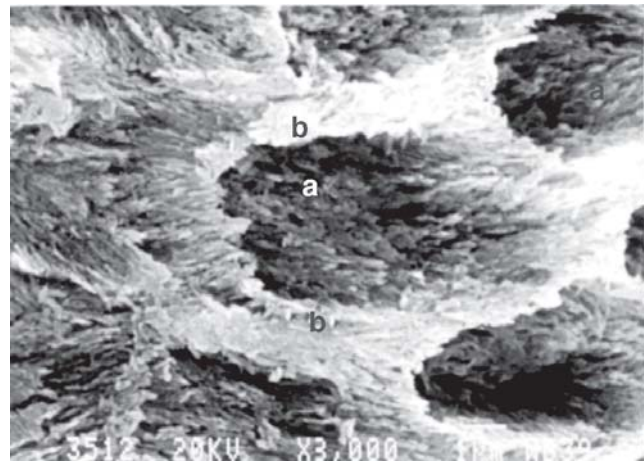


Figure 4.7: Enamel rods as seen under higher magnification showing detailed structure of the enamel rod x 3000 (a) Enamel rod made up of small long crystallites (b) Rod sheath crystals

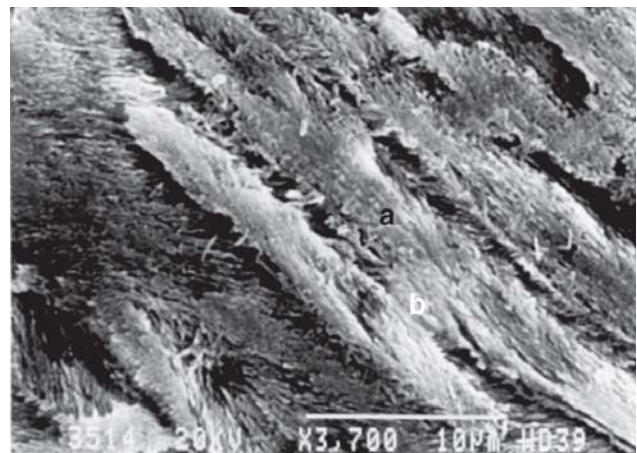


Figure 4.8: Longitudinal section of enamel showing, (a) Enamel rods and (b) crystallites of interprismatic substance. Electron micrograph x 3,700

arranged circumferentially around the long axis of the tooth. In each row, rods run usually perpendicular to the dentin surface with a slight inclination toward the cusp as they pass outward. As the enamel rod rows reach the cusp tip they have a small radius, and the rods run more vertically. As the enamel fractures between adjacent rod rows, the arrangement of the rod rows has a great clinical importance.

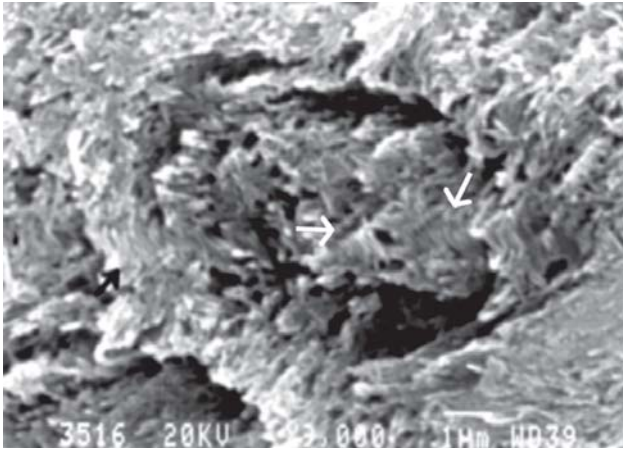


Figure 4.9: Another area of transverse section showing wavy parallel arrangement of crystallites of enamel rods or prisms indicated by arrows. Electron micrograph x 3000

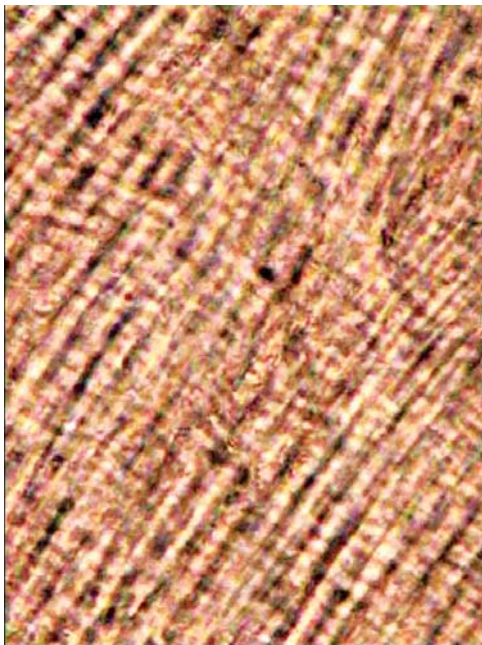


Figure 4.10 A: Enamel: Higher magnification of enamel showing striations x 400

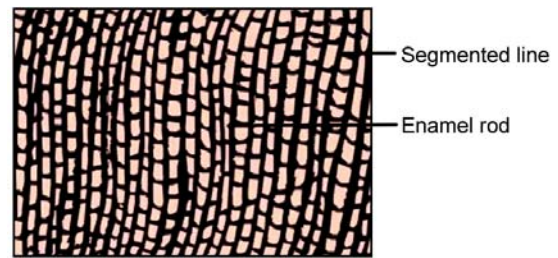


Figure 4.10B: Ground section of enamel. Enamel rod is built up of segments that give it a striated appearance

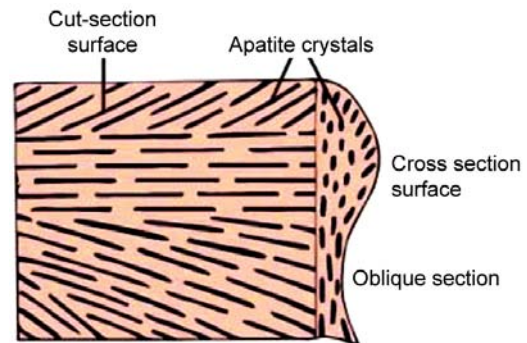


Figure 4.11: Cross-section and cut-section of the mature enamel rod showing orientation of apatite crystals

Rod Sheaths

Under light microscope, a distinct thin layer is seen peripheral to the rods. It has a different refractive index, stains darker and is more acid-resistant than the rod. It is less calcified and contains more organic substance like enamel protein. This layer is known as rod sheath. At the rod sheath the crystals of the rod meet at right angles. The fish-scale appearance of enamel matrix is due to regular arrangement of rod sheath with higher protein content. Some observers believe that rod sheath also contains fine organic fibrils. The rod sheath may be absent in many cases. It happens when there is an increase in crystal size at the border between two adjacent rods till the interspace disappears. The center of the rod is called the core of the rod. When hypomineralized enamel is seen under a high power microscope, it shows wavy bundles (Fig. 4.12). These are darkly stained because of higher protein content. These are the sheath of enamel rod which pick up the stain, as mineral content is dissolved (Fig. 4.12).

Inter-rod Substance (Cement)

Light microscope revealed that the rods are cemented together by inter-rod substance, which has slightly higher

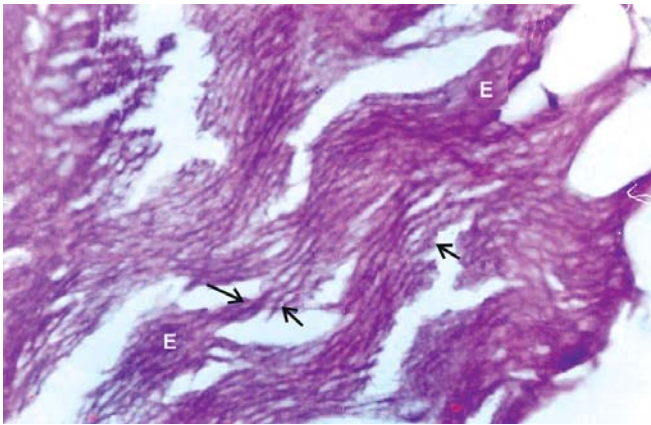


Figure 4.12: Hypomineralized enamel (E) showing wavy disposition of enamel rods. The sheath of enamel rods shown by small arrows takes up eosin stain (pink color) while mineral content has got dissolved during decalcification leaving thin needle like spaces. (H and E x 400)

refractive index than the rods. The crystals are arranged in a different direction in the inter-rod region.

Other Structures

Hunter-Schreger Bands

These are alternating dark and light bands which are best visualized in longitudinal ground section under oblique reflected light. Hunter-Schreger bands are optical phenomena and are considered as produced solely by changes in the rod direction. When a longitudinal ground section of tooth is seen in oblique reflected light, a series of alternating dark and light strips of varying widths (approximately 50 microns on an average) are seen in enamel (Fig. 4.13). This is due to a change in the direction of enamel rods, which are responsible for the appearance of Hunter-Schreger bands or zones of Schreger. They are mostly seen on the inner 25 to 50 percent portion of enamel thickness. The changes in direction of enamel rods minimize cleavage in the apical direction under the influence of occlusal masticatory forces.

Therefore, it may be regarded as a functional adaptation. These bands originate at the dentinoenamel border and pass outward, ending at some distance from the outer enamel surface. They run perpendicular to the Striae of Retzius. The light bands are referred to as diazones and the dark bands are called as parazones. The diazones are areas where rods have been cut in cross-section and

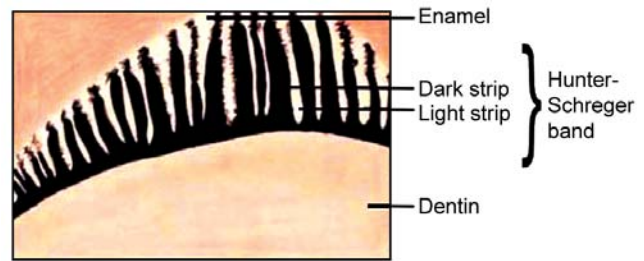


Figure 4.13: Appearance of Hunter-Schreger bands in longitudinal ground section of enamel

parazones are the areas where rods have been cut at longitudinal section. The angle between the diazones and parazones is approximately of 40 degrees. The convexity of curvature of the bands is always directed towards the root.

A number of possible explanations are given for the appearance of these bands. The following are some of them.

1. Most of the workers consider that these bands occur due to change in the direction of rods that is one group of rods extends toward the surface with a mesial drift while the adjacent group might show a distal drift.
2. According to some workers, they are composed of a slightly different content of organic material.
3. They may occur due to differences in permeability.

Incremental Lines of Retzius

The incremental lines of Retzius illustrate the rhythmic successive apposition of layers of enamel during formation of the crown. When a ground section of a tooth is seen under a light microscope, concentric brown lines are seen in the enamel (Figs 4.14 and 4.15). These are called incremental lines of Retzius or Striae of Retzius. In transverse section of a tooth, the incremental lines of Retzius appear as concentric rings (Fig. 4.16). In longitudinal sections, the lines traverse the cusp and incisor area in a symmetrical arc pattern (Figs 4.17A and B) descending obliquely to the dentinoenamel junction. In the cervical part of the crown, they run obliquely. They deviate occlusally from dentinoenamel junction to the surface. When these circles are incomplete at the enamel surface, a series of alternative grooves, which are called the imbrication lines of Pickerill, are formed (Fig. 4.18).

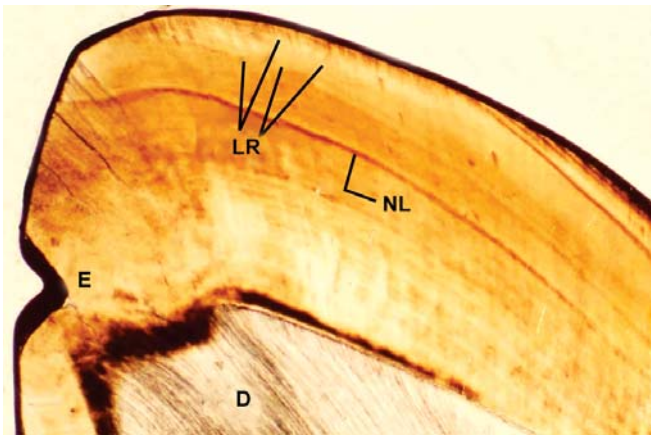


Figure 4.14: Incremental lines of Retzius and neonatal lines: Ground section of tooth: parallel oblique lines representing incremental lines of Retzius (LR) are seen traversing the enamel (E). The thick prominent line represents neonatal line (NL) D= Dentin (Ground section of tooth x 30)

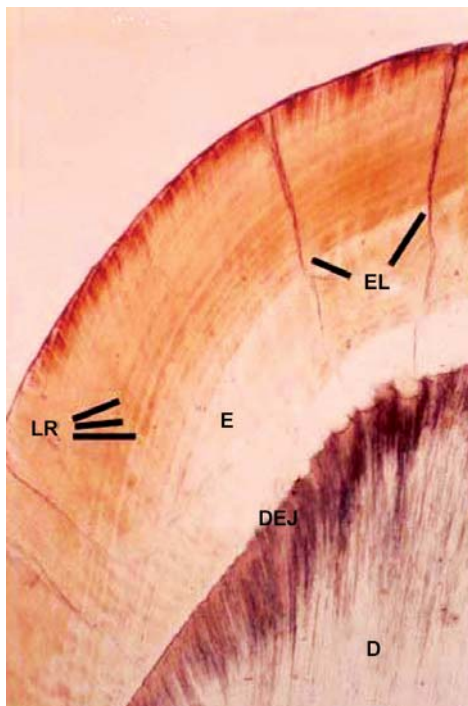
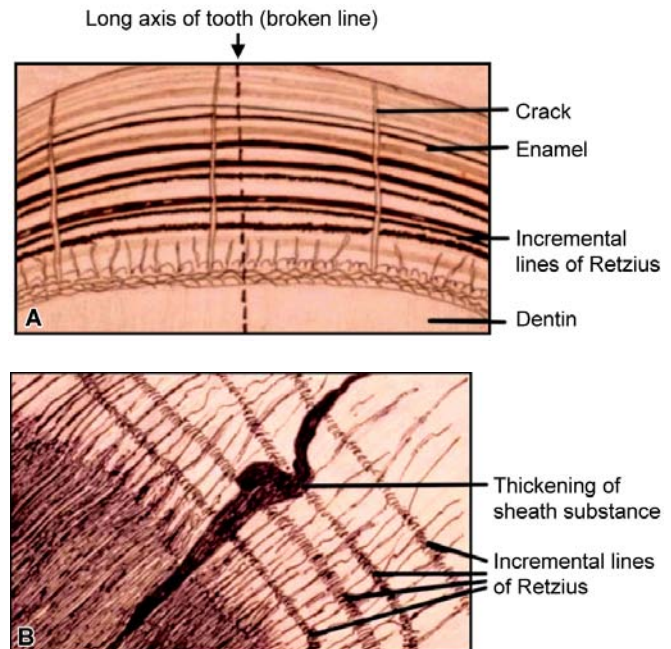


Figure 4.15: Incremental lines of Retzius and enamel lamellae: Ground section of tooth showing incremental lines of Retzius (LR). EL= Enamel lamellae, E= Enamel, DEJ= Dentinoenamel junction, D= Dentin x 100

The elevations between the grooves are called perikymata. They may be compared to the growth rings present in the cross section of a tree. A number of possible explanations have been put forward to explain the formation of these lines.



Figures 4.16 A and B: Incremental lines of Retzius (arranged concentrically). A, In ground section (almost perpendicular to the long axis of tooth shown with broken lines). B, In decalcified section under high magnification

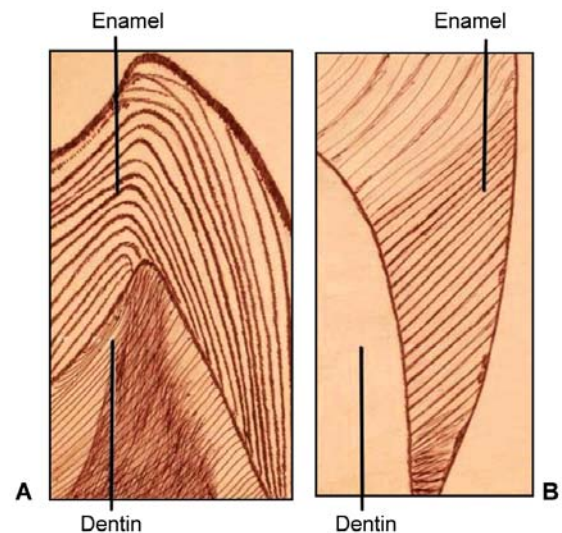


Figure 4.17 A and B: Rhythmic Incremental lines of Retzius in ground section (longitudinal). A, In cuspal region (passing over cusp without reaching enamel surface). B, In cervical region

1. According to some workers these lines represent a hypomineralized or rhythmic formation of the enamel.
2. Periodic bending of the enamel rods, variations in the basic organic structure or in a physiological

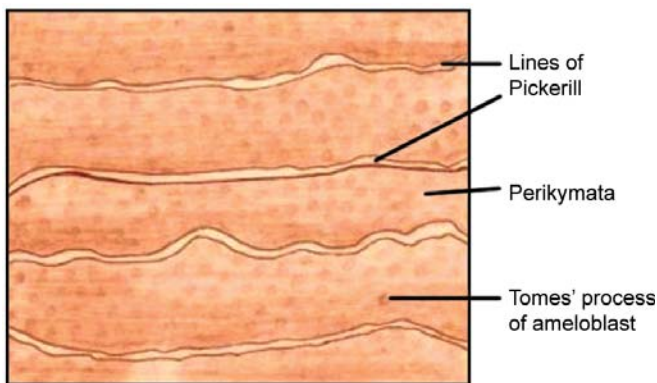


Figure 4.18: Elevations are lines of Pickerill, furrow between them are the perikymata, small pits are impression of Tomes' process of ameloblast (as observed by SEM)

calcification rhythm could be another possible reason for these lines.

3. The occurrence of a few incremental lines (striae) is considered normal, but when they are present in great number or as a broad band it indicates periods of metabolic disturbance or disturbance in amelogenesis.

The incremental lines (striae) of Retzius are more frequently seen in permanent teeth and less frequently in deciduous teeth and prenatal enamel.

Structureless Outer Enamel Layer

There is a structureless outer layer of enamel about 30 microns thick found most commonly towards the cervical area and less often on cusp tips (Fig. 4.19). This structureless layer is called prismless enamel and found in all deciduous teeth and in 70 percent of the permanent teeth. There are no prism outlines visible and all the apatite crystals are parallel to one another and perpendicular to the incremental lines (Striae) of Retzius. It appears that this layer is more heavily mineralized than the bulk of enamel beneath it.

Other microscopic details that have been observed on outer surfaces of enamel of newly erupted teeth are the perikymata, rod ends and cracks (lamellae).

Perikymata

Perikymata are wave-like, transverse grooves. They are shallow furrows and most probably the external manifestation of incremental lines of Retzius. They are continuous around a tooth and usually lie parallel to each other and to the cementoenamel junction. Usually there are 30 perikymata per millimeter in cementoenamel junction

and 10 perikymata near the occlusal or incisal region of a tooth. Except in the cervical region, their course is usually regular. Perikymata are absent in the prenatal occlusal parts of the deciduous teeth. They are present in the postnatal cervical parts of the deciduous teeth. In prenatal period, there is undisturbed supply of nutrients even at the cost of mother's health, hence they are absent in the prenatal period.

Enamel Caps—Enamel caps are the small elevations on the surface of enamel. These caps are 10 to 15 micron in diameter and develop due to the enamel deposition on nonmineralizable debris. Larger caps are known as enamel brochs.

Surface pits—surface pits represent the ends of the ameloblasts and size of surface pit is 1 to 1.5 micron in diameter.

Enamel Rod Ends

The enamel rod ends are concave and vary in depth and shape. They may contribute to the adherence of plaque material with a resultant caries attack, especially in young people. The rod ends are deepest at the incisal and occlusal surfaces and shallowest at the cervical areas (Fig. 4.20).

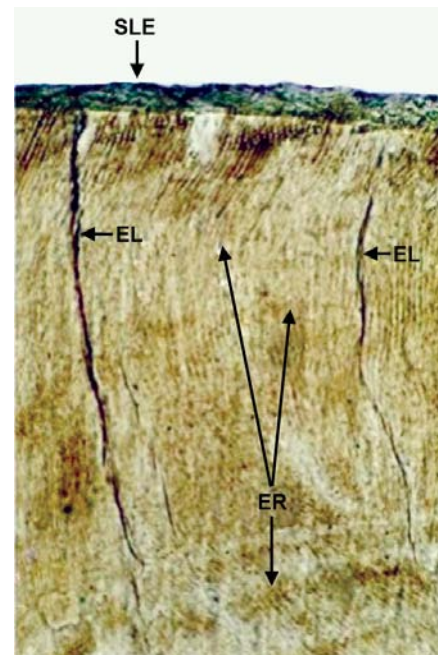


Figure 4.19: Enamel: Structureless outer surface layer of enamel (SLE) is seen. Enamel rods (ER) and enamel lamellae (EL) are also visible. Ground section (x 200)

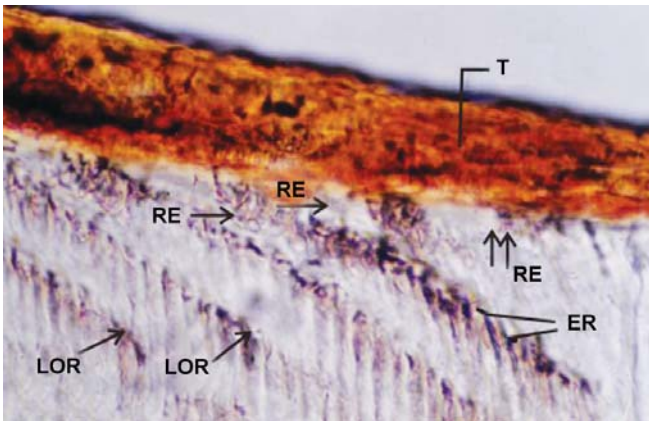


Figure 4.20: Enamel rod ends (RE) seen in high magnification in ground section appear as curved rods. Layer of tartar (T) can be seen on enamel surface. LOR= Lines of Retzius, ER= Enamel rods (x 400)

Cracks

Originally the term “crack” was used to describe the very narrow, fissure-like structures that are present on almost all surfaces. They are actually the outer edges of enamel lamellae. They originate from dentinoenamel junction and run at right angles to it. Their length is variable but most of them are less than a millimeter in length. However, with age the dimpled surface anatomy of the enamel may gradually wear and become smooth (Fig. 4. 21). However only few of them reach the occlusal or incisal surface.

Neonatal Line or Ring

In deciduous teeth, the enamel develops partly before and partly after birth. The line or boundary between the two portions of enamel in deciduous teeth is known as neonatal line or neonatal ring (Fig. 4.22). It appears due to the abrupt change in the environment and nutrition of the newborn (infants). It is an accentuated incremental line of Retzius. Usually the prenatal enamel is better developed as there is timely and uninterrupted supply of all nutrients, even at the cost of mother’s health, than postnatal enamel.

The fetus develops in a well-protected environment with sufficient supply of all the essential nutrients from mother’s blood. Hence, neonatal lines are absent in the occlusal and incisal parts of deciduous teeth, while they are present in postnatal cervical areas.

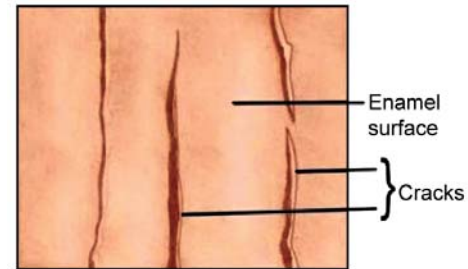


Figure 4.21: Progressive loss of surface structure in a 50 years old patient. Enamel surface appears completely smooth by naked eye. Under magnification, appearance of “Crack” is a feature of advancing age

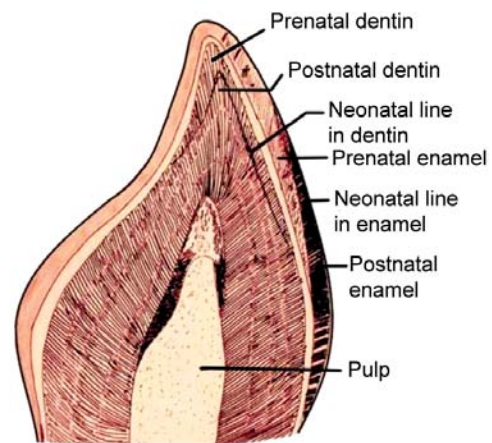


Figure 4.22: Longitudinal ground section of deciduous canine showing neonatal line in enamel and dentin along with prenatal and postnatal enamel and prenatal and postnatal dentin

Nasmyth’s Membrane (Primary Enamel Cuticle)

Nasmyth’s Membrane is a nonmineralized, electron dense membrane usually found between the epithelium of dentogingival junction and the enamel surface. It is formed by an accumulation of basal lamina material produced by the junctional epithelium of the dentogingival junction. Enamel is incapable of repairing itself once it is destroyed because the ameloblast cell degenerates following the formation of the enamel rod. The final act of the ameloblast cell is secretion of a layer covering the end of the enamel rod. This layer is referred to as the Nasmyth’s membrane (named after its first reporter) or the primary enamel cuticle. This delicate membrane covers the entire enamel of newly erupted tooth and is worn away by mastication and cleaning.

After laying down of the enamel rods, ameloblasts become indistinguishable from cells of stratum intermedium with which they are in close contact. The two cell layers eventually merge with the overlying oral epithelium to form reduced enamel epithelium. It is believed that this reduced enamel epithelium plays some role in passive eruption of teeth and overlies the primary cuticle. Histochemical studies have shown that the reduced enamel epithelium is proteinaceous structure along with a variable amount of carbohydrate. The secondary cuticle is known as dental cuticle. It covers enamel and a part of cementum. This is about 10 microns in thickness.

Pellicle (Salivary Pellicle)

After tooth is cleaned, salivary proteins and glycoproteins having strong affinity for enamel get adsorbed to the enamel surface very quickly and form a very thin layer called the salivary pellicle. It is less than one micron thick. In the beginning it is bacteria-free and is a dark and amorphous layer. Erupted enamel is usually finally covered by pellicle, which appears as precipitated salivary proteins (Fig. 4.23).

This pellicle is removed by mastication or vigorous brushing and is again formed by itself within hours. After 24 to 48 hours of pellicle formation, colonies of microorganisms develop over it. Then it is called bacterial plaque (Fig. 4.24).

Enamel Lamellae

Enamel lamellae are very thin, leaf-like structures, sometimes visible to naked eye. They extend from the enamel surface towards the dentinoenamel junction, rarely extending into dentin. The enamel lamellae contain mostly organic material and represent improperly mineralized enamel. Lamellae may develop in planes of tension (Figs 4.19 and 4.25 to 4.27).

Enamel lamellae can be differentiated into three types:

Type A lamellae composed of poorly calcified rod segment.

Type B lamellae composed of degenerated cells.

Type C lamellae arising in erupted teeth where the cracks are filled with organic matter and debris from saliva.

Type A is restricted to enamel and type B and C may reach the dentin. If crack in enamel is filled with

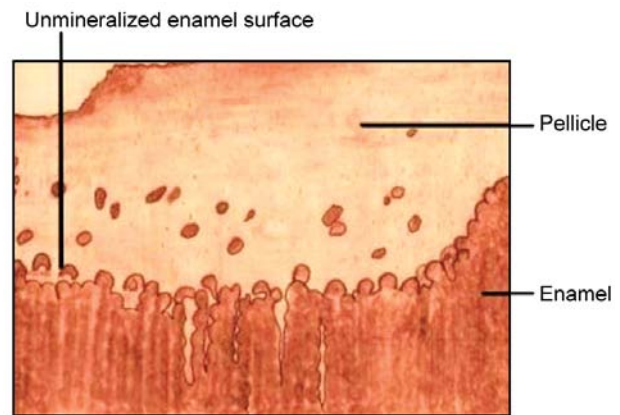


Figure 4.23: Enamel surface is covered by pellicle (as seen under electron microscope)

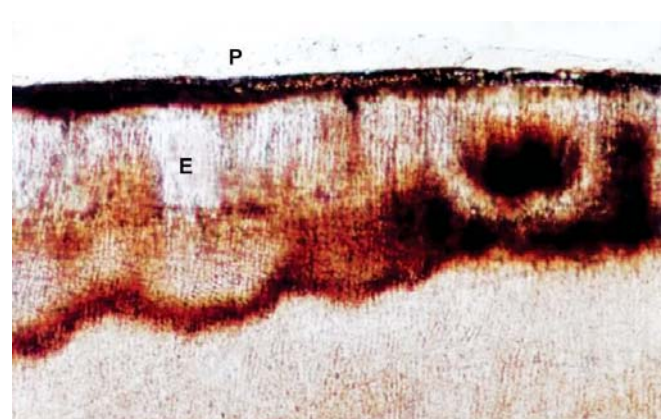


Figure 4.24: Plaque: (P) formation can be seen above the surface of enamel (E) as a thin layer of refractive material (Ground section. x 400)

connective tissue, cementum may be formed in the crack and lamellae may consist of cementum. The enamel lamellae may develop in planes of tension and are more frequently seen in cervical areas. It has been observed that enamel lamellae may be sites of weakness in a tooth. They may form a road for entry of bacteria that may initiate caries.

Enamel lamellae can be distinguished from enamel cracks by careful decalcification of ground section. The cracks disappear whereas enamel lamellae persist.

Enamel Tufts

Enamel tufts are hypocalcified enamel rods and interprismatic substance that originates at the dentinoenamel junction and extends into enamel for about

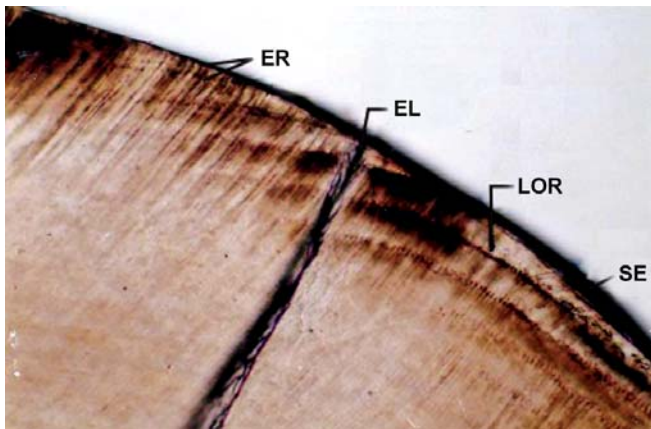


Figure 4.25: Structures of enamel: Ground section of the tooth: Enamel lamella (EL) are considered as cracks or artifacts in enamel. Incremental lines of Retzius (LOR), enamel rods (ER) and structureless surface layer are clearly visible. Surface enamel (SE) x 150

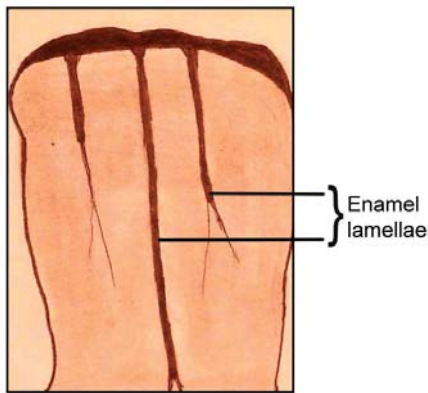


Figure 4.26: Decalcified ground section of incisor with leaf-like enamel lamellae

one-third to one-fifth of its total thickness. They appear branched and contain greater concentrations of enamel protein than the rest of the enamel. The enamel protein content of tuft was 13.17 kd protein rather than amelogenin. Similar to enamel lamellae they extend into the enamel in the direction of the long axis of crown and may play a role in the spread of dental caries. They are known as enamel tufts because they look like tufts of grass projecting into enamel. Enamel tuft is a narrow ribbon-like structure, inner end of which occasionally may very slightly project into dentin (Fig 4.27 and 4.28 A and B).

Developmentally, they are formed due to the abrupt change in the direction of rod, which leads to different ratio of inter-rod and rod enamel, creating less mineralized

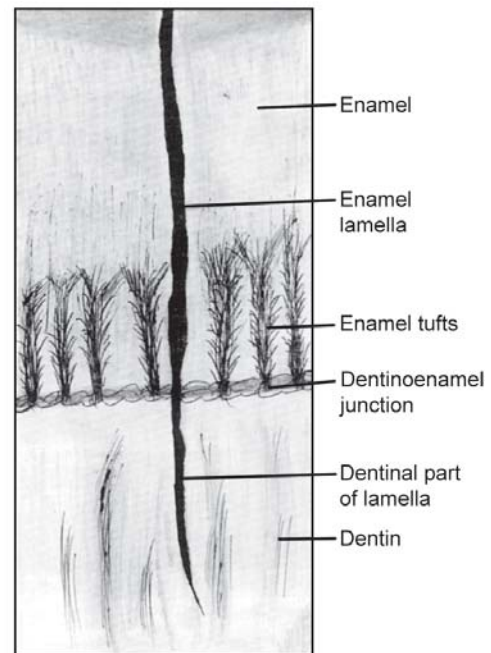


Figure 4.27: Enamel lamella extending from enamel surface into dentin and enamel tufts seen in a transverse ground section

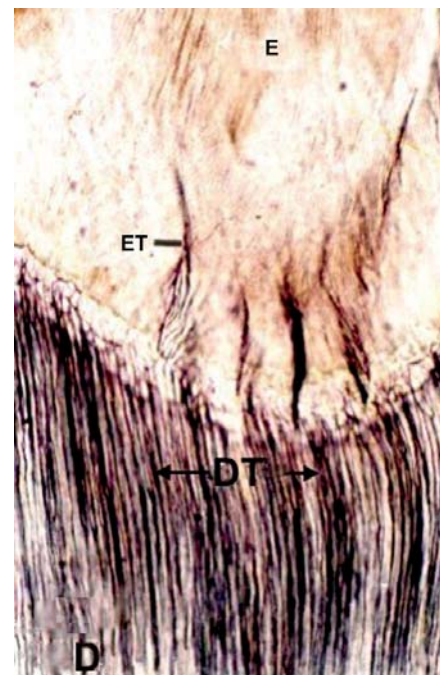


Figure 4.28 A: Dentin enamel junction: Ground section of tooth: Ribbon like wavy bands of enamel rods appearing like tufts of grass called enamel tufts (ET) are seen arising from dentinoenamel junction. Dental tubules (DT) are also clearly visible x 150. E = Enamel, D = Dentin

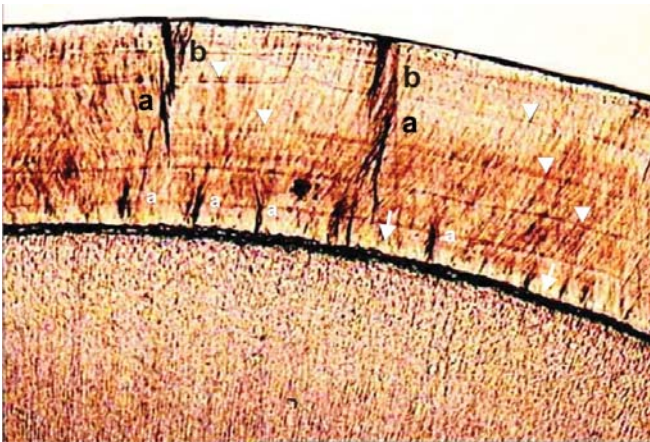


Figure 4.28 B: Enamel tufts: Ground section (horizontal) showing grass like tufts (a) of enamel rods arising from dentinoenamel junction (indicated by arrows) and extending towards surface of enamel. Two enamel lamellae (b) are also present. Horizontal lines of Retzius are indicated by arrow heads x 40

and weakened planes. They are best seen in transverse ground section of enamel.

Dentinoenamel Junction

The dentinoenamel junction is established as soon as the two hard tissues enamel and dentin begin to form. The dentinoenamel junction is a scalloped interface between the enamel and dentin. Dentin has pitted surface, which supports the enamel (Figs 4.29 to 4.31). Small curved projections of enamel fit into small concavities of the dentin. This relationship assures the firm hold and increased adherence of the enamel cap on the dentin. The surface of the dentin at the dentinoenamel junction is pitted and convexities of the scallops are directed towards the dentin. The dentinoenamel junction is a hypermineralized zone that is about 30 microns thick. It is most prominent before completion of mineralization. In ground section, this junction is clearly seen.

Even before mineralization of dentin and enamel, the pitted dentinoenamel junction is preformed.

It is evident in the arrangement of the ameloblasts and the basement membrane separating it from dental papilla.

Enamel Spindles

Odontoblastic processes sometimes cross the dentinoenamel junction and get entrapped in the enamel matrix. Since mostly they are thickened at their end they have been termed as enamel spindles. They appear to originate

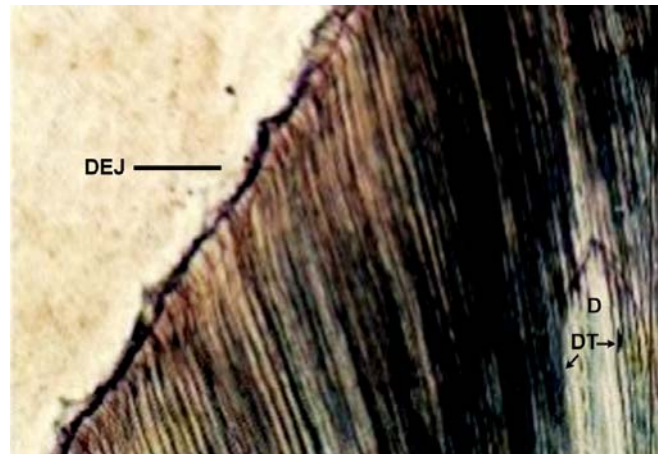


Figure 4.29: Ground section of tooth showing enamel (E), dentin (D) and dentinoenamel junction (DEJ). DEJ is a scalloped line with convexity towards dentin and concavity toward enamel. Enamel projections fit snugly into shallow depressions of dentin. ER= Enamel Rods DT= Dentinal tubules (x 150)

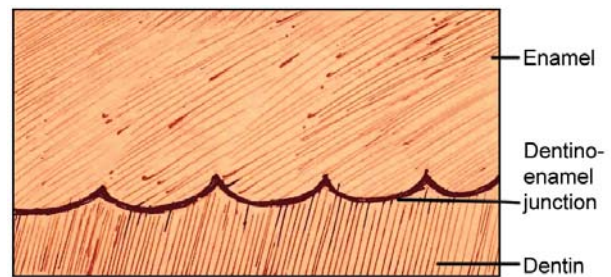


Figure 4.30: Scalloped dentinoenamel junction (DEJ) in ground section

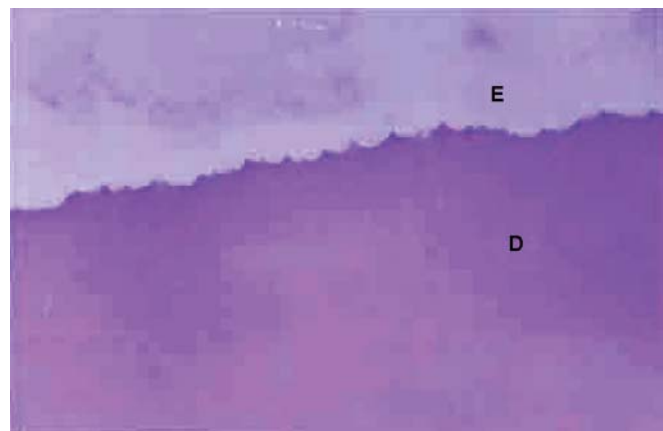


Figure 4.31: Decalcified section of tooth showing dentin (D), scalloped dentinoenamel junction with concavity towards enamel (E) dissolved during decalcification and concavity towards dentin (D) (H & E x 100)

from processes of odontoblasts that extend into the enamel epithelium before mineralization starts. They may serve as pain receptors, thereby explaining the enamel sensitivity experienced by some patients during cavity preparation. The direction of the odontoblast processes and spindles in the enamel corresponds to the original direction of the ameloblasts and is at right angles to the surface of the dentinoenamel junction (Fig. 4.32). In dried ground sections, the organic material of the spindles disintegrates and the space is filled by air, which appears dark in transmitted light (Fig. 4.33).

Gnarled Enamel

The enamel rods at the cuspal and incisal region appear intertwined, twisted and inter-twisted, and are more irregular (Figs 4.34 to 4.38). Such kind of optical appearance of enamel is called as gnarled enamel. They are more so at the cuspal region than incisal region. It extends throughout the thickness of the enamel at the cusp tips and incisal edges. The enamel rods on the occlusal surface of the premolars and molars converge in their outward course (Figs 4.36 to 4.38).

Gnarled enamel aids in resisting the high masticatory loads without fracture that the cusps have to bear.

Enamel Droplets or Enamel Pearls

Occasionally, the cells of the epithelial root sheath remain adherent to the dentin surface, they may differentiate into functioning ameloblasts and form small round islands of enamel. Such droplets of enamel are called enamel pearls. They may be found near or in the bifurcation or trifurcation of the roots of permanent molars.

Enamel pearls very rarely contain a small layer of dentin and very, very rarely a small strand of pulp tissue extending from the pulp chamber or root canal of the tooth.

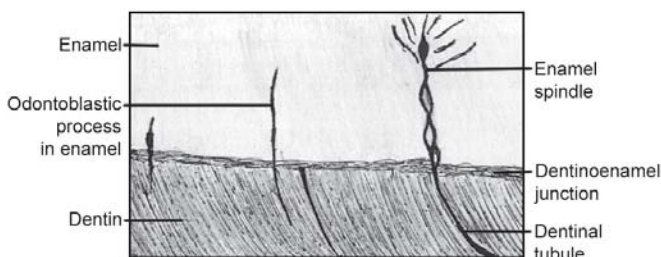


Figure 4.32: Odontoblastic processes extending into enamel as enamel spindles, as seen in ground section

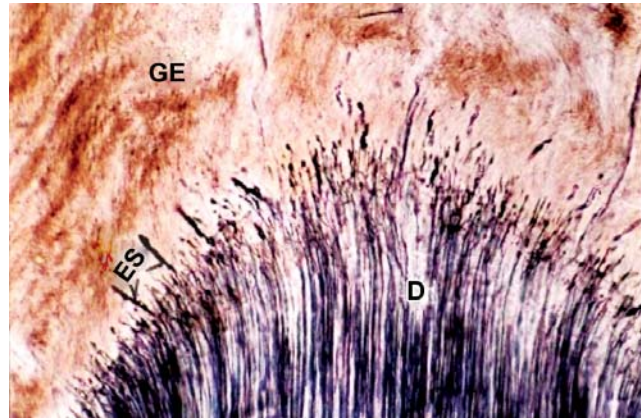


Figure 4.33: Enamel spindles (ES) are club shaped extensions of odontoblastic processes into the enamel. Enamel in this case is gnarled, and consists of complexly interwoven enamel rods (x 150). D= Dentin, GE= Gnarled enamel. Ground section of tooth

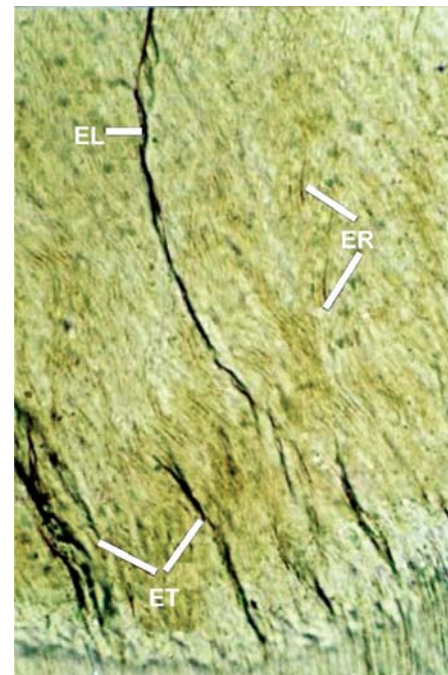


Figure 4.34A: Gnarled enamel: Ground section of enamel near the apical region of incisor showing complexly arranged wavy bundles of enamel rods (ER) in gnarled enamel. ET= Enamel tuft, EL=Enamel lamella (x200)

AGE CHANGES

Enamel does not contain nerves and is incapable of repair and replacement. The surfaces of unerupted and recently erupted teeth are covered completely with pronounced rod ends and perikymata. At the points of highest contours of the surfaces, these structures soon disappear.

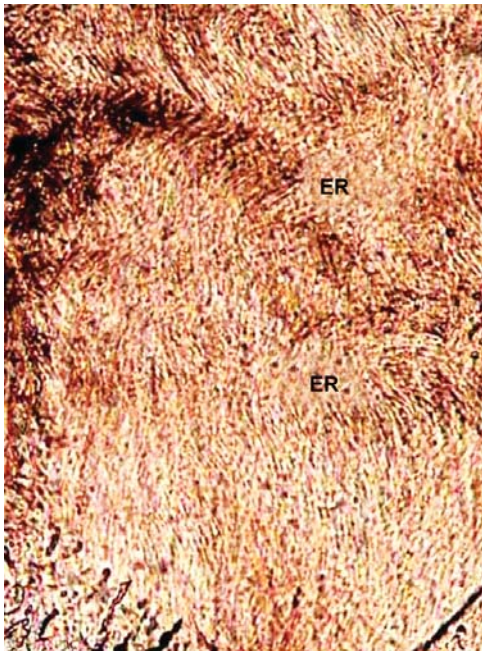


Figure 4.34B: Gnarled enamel: Complexly arranged wavy bundles of enamel rods (ER) in gnarled enamel. Ground section x 40

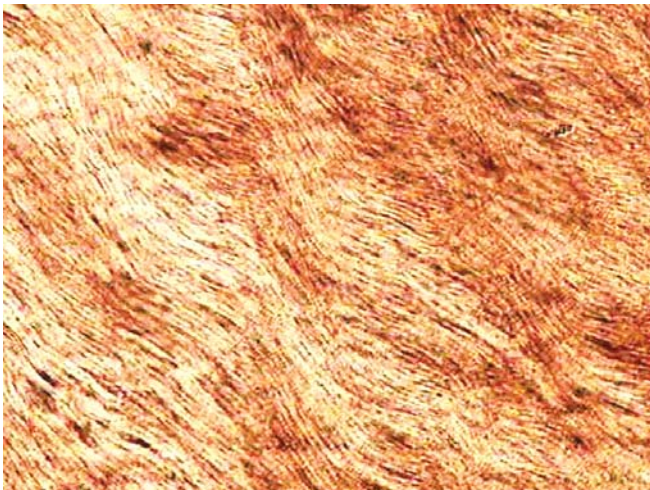


Figure 4.35: Gnarled enamel: Higher magnification showing Complex wavy bundles of enamel rods. Ground section x 100

Gradually there is generalized loss of the rod ends and flattening of the perikymata. Ultimately, the perikymata disappears completely.

Gradually, with age the enamel is worn away in the regions where masticatory forces are applied, which is called as 'attrition'. The most important age change in enamel is attrition or wear of occlusal surfaces and

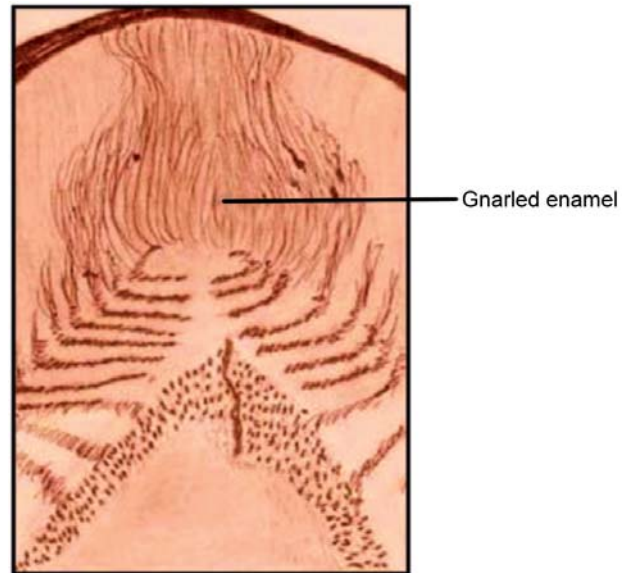


Figure 4.36: Gnarled enamel. Ground section of enamel showing spiral changes of prism direction giving the appearance known as gnarled enamel x 40

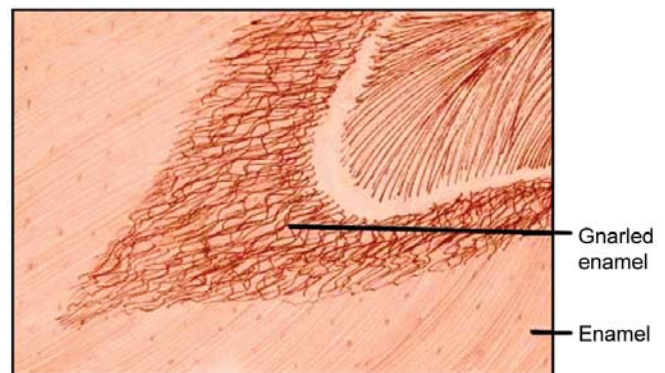


Figure 4.37: Gnarled enamel x 40

proximal contact points as a result of mastication. With advancing age, enamel becomes darker in color and its resistance to caries may be increased. This is due to addition of organic material to enamel from the environment, increased pigmentation of organic part and increased thickness of dentin. Localized increase of certain elements such as nitrogen and fluorine is found in the superficial layer of enamel of older teeth. Gradually, enamel becomes less permeable with advancing age. Facial and lingual surfaces lose their structures more than proximal surfaces, as on these surfaces food rubs during mastication. Enamel found in young individuals works

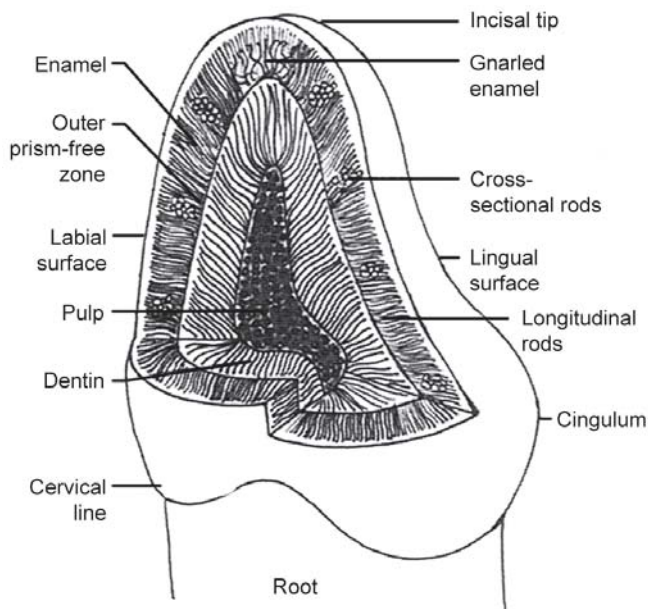


Figure 4.38: Enamel rod orientation in both longitudinal and cross-section of the crown of lower canine. Gnarled enamel (having intertwined rods) is present at the incisal tip

like a semi-permeable membrane. It permits the slow passage of water from surrounding and substances having smaller molecular size through pores in between the crystals. Due to aging, the pores reduce in size and number as the crystals acquire more ions and increase in size. Water content of enamel also decreases with age.

DEVELOPMENT

Enamel is an ectodermally originated tissue covering the anatomical crown of a tooth. It is formed by the dental organ. Dental organ is derived from a localized proliferation of the oral epithelium. The dental (enamel) organ originates from the stratified epithelium of the primitive oral cavity.

Epithelial Enamel Organ

Epithelial enamel organ is also called epithelial dental organ. After growth of papilla and the enamel organ, the developing tooth reaches the morphodifferentiation and histodifferentiation stage which is also called as the bell stage. At bell stage, the cells of inner enamel epithelium take the shape of the crown of the tooth they have to form. After originating from the stratified epithelium, the enamel organ consists of four different layers.

1. Outer enamel epithelium
2. Stellate reticulum
3. Stratum intermedium
4. Inner enamel epithelium (which become the ameloblasts that form the enamel).

The various layers of epithelial cells are named according to their morphology, location or function.

The meeting line between the inner enamel epithelium and the connective tissue of the dental papilla is the final dentinoenamel junction. At the basal opening of enamel organ where outer enamel epithelium and inner enamel epithelium meet, the cervical loop is formed. From the loop towards the future crown of the tooth, the inner and outer enamel epithelia are separated from each other by a large mass of cells that are differentiated into two separate layers. The inner layers, which consist of two or three rows of flat polyhedral cells is closer to the inner enamel epithelium. It is called the stratum intermedium. The center layer consisting of more loosely arranged cells is called as the stellate reticulum (see Figs 3.12 to 3.18 and 3.21 to 3.32).

Outer Enamel Epithelium

Before mineralization starts, the outer enamel epithelium is made up of a single layer of cuboidal cells. This layer is separated from the surrounding connective tissue of the dental sac by a delicate basement membrane (see Fig. 3.25). Till mineralization of enamel, this regular arrangement of the outer enamel epithelium is maintained only in cervical part of the enamel organ. In the region of highest convexity of the organ, the cells of the outer enamel epithelium become irregular in shape nearly similar to the cells of the outer portion of the stellate reticulum. Before enamel and dentin formation, the capillaries around the epithelial enamel organ proliferate and grow toward the enamel organ (see Figs 3.19 to 3.23). Just before dentin and enamel formation starts the blood capillaries may even protrude into the stellate reticulum. This increased vascularity provides rich metabolism at the time when rich supply of substances from the blood stream is required in the inner epithelium (see Figs 3.19 to 3.25).

During the formation of enamel, cells of the outer enamel epithelium undergo specialized changes for the faster transportation of the materials from surrounding connective tissues to the inner enamel epithelium. These changes are the development of villi and cytoplasmic vesicles and the large number of mitochondria. The walls

of the blood capillaries in contact with outer enamel epithelium become very thin for faster percolation, transfusion and transportation of the materials from the blood capillaries to the region of inner enamel epithelium, where enamel and dentin formation is taking place.

Stellate Reticulum

The stellate reticulum forms the middle part of the enamel organ. In this region, the cells are star-shaped with long processes in all directions. The processes of cells are attached to the processes of the adjacent cells (see Figs 3.9 to 3.16). These cells are separated from each other by the large intercellular spaces. These spaces are filled with intercellular substance. These processes of the cells are also connected with the cells of outer enamel epithelium and the stratum intermedium by desmosomes. The desmosomes are circular, dense bodies that are formed at the site of attachment between certain epithelial cells.

Due to this special structure of the stellate reticulum, it has the properties of resistance and elasticity. Therefore it acts as a buffer against physical forces. In this way, it prevents the developing dentinoenamel junction from distortion and avoids gross morphologic changes in the crown. The stellate reticulum allows selective flow of nutritional elements from the outer blood vessels to the specialized formative cells, ameloblasts and odontoblasts (see Figs 3.19 to 3.26 and 3.33). When first few layers of dentin are laid down, the thickness of stellate reticulum is reduced to further facilitate the supply of nutritional elements to the formative cells. Gradually the original source of nutritional supply to the inner enamel epithelium from the dental papilla is stopped.

Stratum Intermedium

Between the stellate reticulum and the inner enamel epithelium lie the cells of the stratum intermedium. These cells are flat to cuboidal in shape and are arranged in one to four layers (see Figs 3.19 to 3.26 and 3.33). These cells are connected with neighbouring cells among themselves and also with the cells of stellate reticulum and the inner enamel epithelium through desmosomes. The tonofibrils are present in cytoplasm. They are oriented parallel to the surface of the developing enamel. Even after the cells of the inner enamel epithelium stop dividing, the cells of stratum intermedium show mitotic division

The function of the cells of the stratum intermedium is not very clear but probably they help in enamel formation by one of the following processes.

- a. They control the fluid content of the ameloblasts, by helping in fluid diffusion, into and out of the ameloblasts.
- b. By contributing required formative elements or enzymes.

Inner Enamel Epithelium (Ameloblastic layer)

Basal cell layer of the oral epithelium gives rise to cells of the inner enamel epithelium. These cells assume columnar shape and differentiate into ameloblasts. Hence, it is also known as ameloblastic layer. The ameloblasts produce enamel matrix, which is the first step of enamel formation. The border between the inner enamel epithelium and dental papilla will be the dentinoenamel junction. The shape of the inner enamel epithelium determines the final occlusal or incisal portion of the crown. Differentiation into ameloblasts and production of enamel matrix first takes place in the regions of cusps and incisal edges and then proceeds towards the cervical loop.

Cervical Loop

At the free, basal and broader end of the enamel organ, the outer and inner enamel epithelial layers are continuous and reflected onto one another as the cervical loop (Fig. 4.39). The cuboidal cells gradually become lengthier at this region of transition of outer enamel epithelium and the inner enamel epithelium. After completion of the crown, the cells of this portion produce Hertwig's epithelial root sheath, which forms the root of the tooth.

LIFE CYCLE OF AMELOBLASTS

Depending on their functions, lifespan of the cells of the inner enamel epithelium (ameloblasts) can be divided into six stages – (a) Morphogenic, (b) organizing and differentiating (c) formative (d) maturative or mineralizing (e) protective (f) desmolytic.

The development of a tooth is fast and most advanced at the cuspal and incisal regions, and slow and least

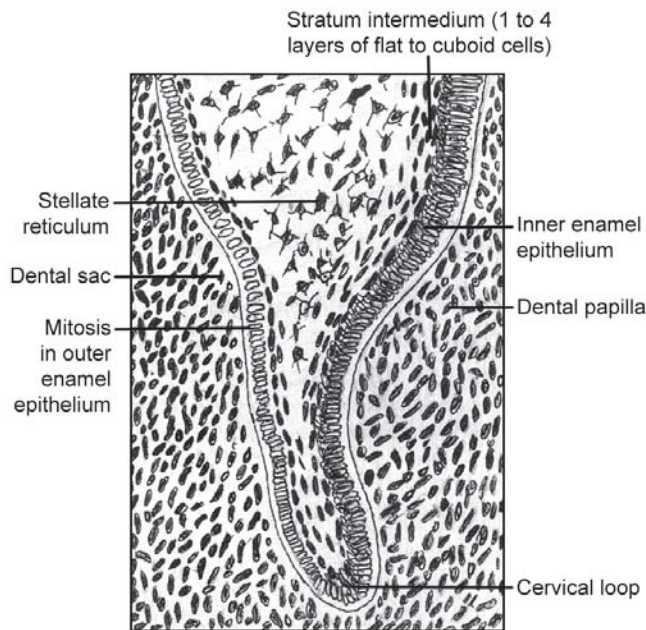


Figure 4.39: Structures of the cervical loop region (seen under higher magnification). Transition of the outer into the inner enamel epithelium

advanced at the cervical loop region. Therefore, if not all, but most of the stages are seen in different regions of a tooth. In developing adjacent teeth of any dentition, all the stages can be definitely seen (see Figs 3.15 and 3.18).

Morphogenic Stage

During morphogenic stage the cells of the dental organ along with the dental papilla react by differential growth to produce the shape of the crown (Figs 4.40 and 4.41). During this stage, the ameloblasts are short and columnar, with large oval nuclei that nearly fill the cell body. The centrioles and the Golgi apparatus are located near the basal or proximal ends of the ameloblasts, and the mitochondria are evenly scattered throughout the cytoplasm. During differentiation of ameloblasts, terminal bars appear along with the migration of the mitochondria toward the basal end of the cell. The terminal bar represents areas of close contact between the cells. The connective tissue of the dental papilla is separated from the inner enamel epithelium by a delicate basal lamina. The pulpal layer adjacent to basal lamina is a cell free zone. It is a light zone containing argyrophilic fibers. The ameloblast is attached to another ameloblast by desmosomes present at both the proximal and distal ends of the cell.

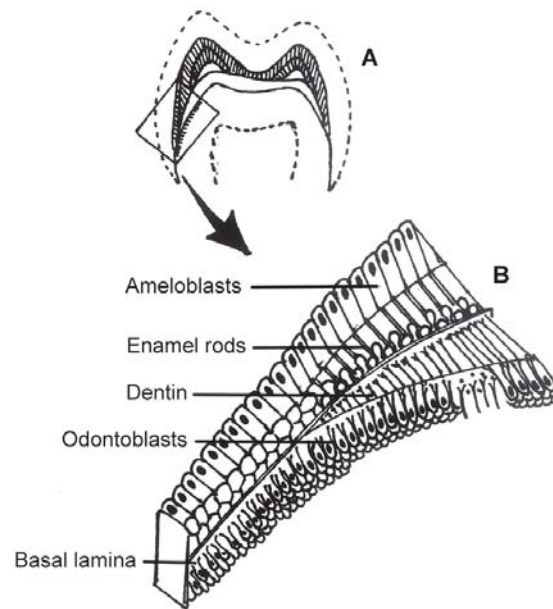


Figure 4.40 A: Early dentinogenesis and amelogenesis (B). Higher magnification of inset in (A) showing differentiation of odontoblasts and ameloblasts

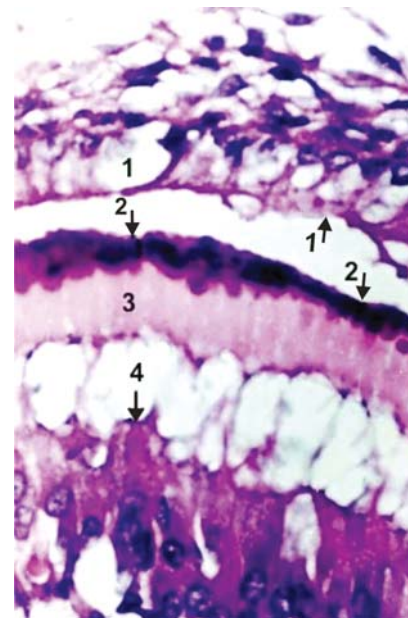


Figure 4.41: Tooth development: Ameloblastic layer (1) and odontoblastic layer (4) show high secretory activity and cells are low columnar to tall columnar in shape. Dense enamel (2) can be seen overlaid on dentin (3). Empty dentinal tubules can be appreciated in the newly formed dentin. The scalloped nature of dentinoenamel junction is very clear with convexity of enamel substance and concavity of the dentinal matrix into which enamel matrix is indenting (H & E x 400)

Organizing and Differentiating Stage

In the organizing and differentiating stage, cells of the inner epithelium interact with the adjacent connective tissue cells of the dental papilla, which differentiate into odontoblasts. In this stage, change in the appearance of the cells of the inner enamel epithelium takes place. They become longer and the nucleus-free zones at the distal ends of the cells become almost as long as the proximal parts containing nuclei. During last phase of the organizing stage, the formation of the dentin by the odontoblasts begins.

By staining methods it is found out that fine acidophilic granules are present in the proximal part of the cell. Electron microscope has shown that the granules are mitochondria, and these get concentrated in this part of the cell. The cell-free zone, which was present between the inner enamel epithelium and the dental papilla, disappears. The zone disappears because of the elongation of the epithelial cells towards the papilla (Figs 4.42 and 4.43).

The cells of the inner enamel epithelium differentiate into ameloblasts. The ameloblasts elongate and their nuclei shift to the proximal side towards stratum intermedium (Fig. 4.44). The Golgi complex increases in volume and occupies central core of the cell. The amount of the rough endoplasmic reticulum increases. At this stage, the ameloblast becomes a highly polarized cell.

Adjacent ameloblasts are attached by junctional complexes. These junctional complexes are present in the proximal and distal ends of the cell. These encircle the cell at their extremities. There is functional difference between the proximal and distal junctional complexes.

The basal lamina supporting the ameloblasts disintegrates after the deposition of a layer of predentin and during differentiation of the ameloblasts. Reduced enamel (dental) epithelium consists of an inner layer of ameloblasts and outer layers formed from the remainder of the dental organ (Figs 4.44 to 4.49).

The first appearance of dentin appears to be a critical phase in the life-cycle of the inner enamel epithelium (ameloblast). With the formation of first layer of dentin, the cells of the inner enamel epithelium (ameloblasts) are cut off from their original source of nourishment, that is, capillaries of dental papilla. After that the cells of inner enamel epithelium (ameloblasts) get their nourishment from the surrounding capillaries. These capillaries may also penetrate the outer enamel epithelium to reach the

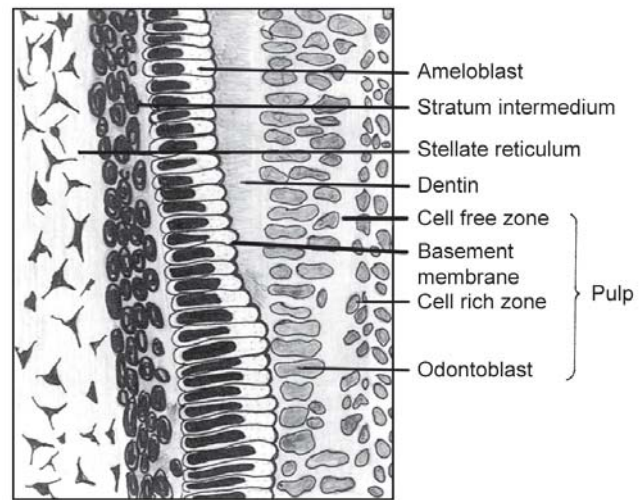


Figure 4.42: The inner enamel epithelium is separated from connective tissue of the dental papilla by basement membrane. The clear zone contains fine argyrophilic fibers (as seen under electron microscope)

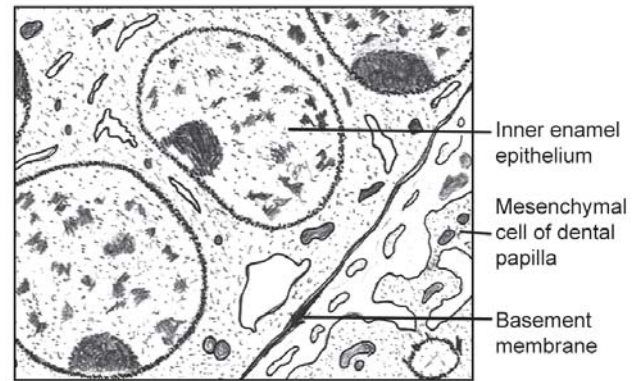


Figure 4.43: Inner enamel epithelium is separated from the connective tissue of the dental papilla by typical basement membrane (Details as observed by electron microscope)

cells of inner enamel epithelium (ameloblasts). With change of the nutritional source the capillaries in the dental sac proliferate and stellate reticulum reduces in size and gradually disappears. By these changes, the distance between the capillaries and the stratum intermedium and the ameloblast cells is reduced (refer to Figs 3.18 to 3.28).

Formative Stage

After the first layer of dentin has been formed, the ameloblasts enter their formative stage. It appears that the presence of dentin is necessary for the beginning of

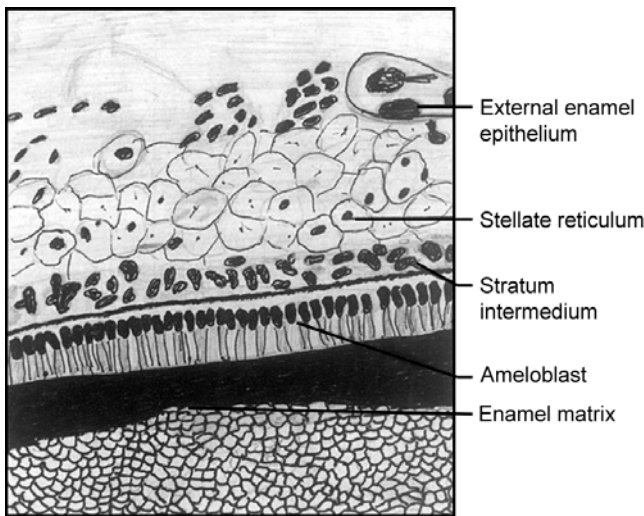


Figure 4.44: Secretory ameloblasts seen under the light microscope. These cells are tall and columnar having nuclei near the proximal ends

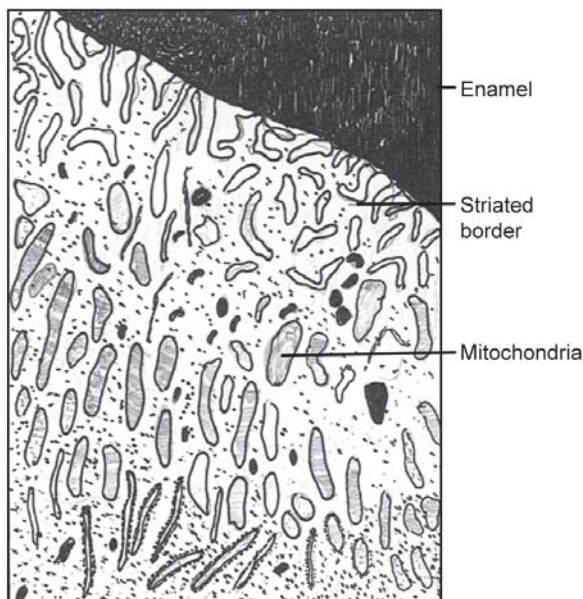


Figure 4.45: Distal (secretory) end of the maturative ameloblast. The accumulation of mitochondria and complex folding of the plasma membrane is seen during secretory phase of the ameloblast (as seen under electron microscope)

enamel matrix formation just as it was necessary for the epithelial cells to come in close proximity with the connective tissue of the pulp during differentiation of the odontoblast and the beginning of dentin formation. The important fundamental law of organogenesis and histodifferentiation is the mutual interaction between one group of cells with another.

Ameloblasts retain almost the same length and arrangement during formation of enamel matrix. The first apparent change is the development of blunt cell processes on the surfaces of ameloblasts. These processes penetrate the dental lamina and enter the predentin.

Maturative or Mineralizing Stage

After most of the thickness of the enamel matrix has been formed in the occlusal or incisal area, enamel maturation (complete mineralization) occurs. The ameloblasts are reduced in size and are closely attached to enamel matrix during enamel maturation. The cells of the stratum intermedium lose their cuboidal shape and assume a spindle shape. Their regular arrangement is also disturbed. Ameloblasts also play an important role in maturation. They display microvilli at their distal end and vacuoles in cytoplasm during maturation. These structures indicate an absorptive function of these cells. The growth of crystals continues to a point of compaction and very little space is left between them.

Most of the organelles associated with synthesis are enclosed in autophagic vacuoles and these are digested by lysosomal enzymes. Now there occurs the shifting of many remaining organelles to the distal part of the cell, and a complex folding of the distal plasma membrane takes place to form a striated border. This striated border greatly increases the surface area of the extremity of the ameloblast. It indicates that the rapid transport of material is taking place across the plasma membrane (Fig. 4.42).

Protective Stage

When the enamel has completely developed and fully mineralized, the ameloblasts lose their striated borders and cease to be arranged in a well-defined layer and get mixed up. These can no longer be differentiated from the cells of the stratum intermedium and outer enamel epithelium. These cells secrete a material that forms a layer of stratified epithelial covering of the enamel, which is called reduced enamel epithelium. Until the tooth erupts, the function of reduced enamel epithelium is the protection of the mature enamel by separating it from the connective tissue. Due to a breach in reduced enamel epithelium, if connective tissue contacts the enamel, anomalies like cementum over the enamel may develop. During this stage, the epithelial enamel organ may retract from the cervical edge of the enamel. This may lead to deposition of afibrillar cementum on the enamel surface (Figs 4.43 to 4.49).

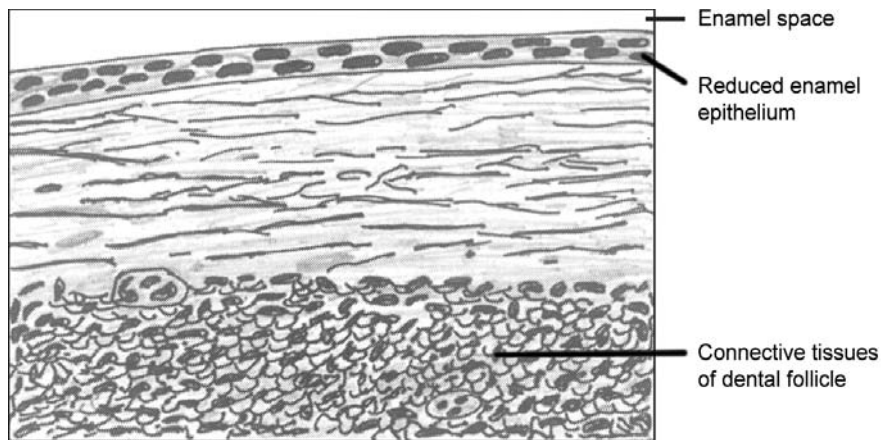


Figure 4.46: Details observed in electron photomicrograph of reduced enamel epithelium (layers of flattened cells) cover surface of unerupted tooth and play a role in formation of the junctional epithelium

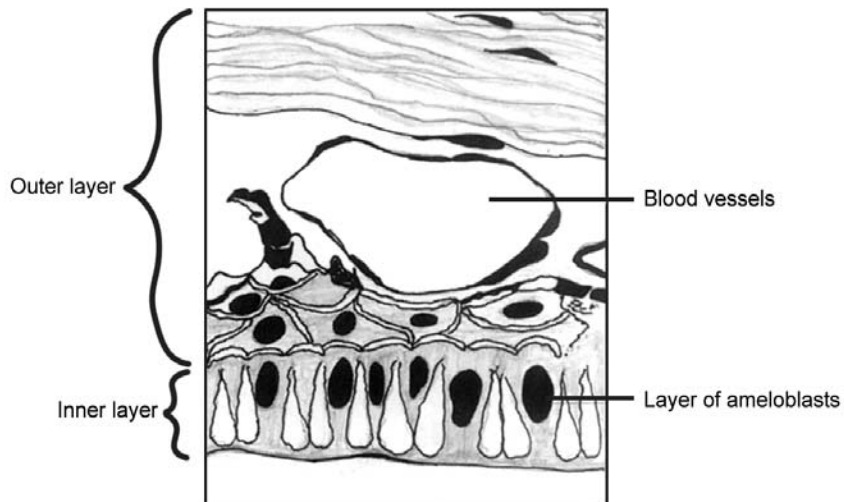


Figure 4.47: The reduced enamel (dental) epithelium showing inner layer of ameloblasts and outer layer showing remaining enamel organ

Desmolytic Stage

The reduced enamel epithelium, after proliferation, induces atrophy of the connective tissue. Which separates it from the oral epithelium. The epithelial cells elaborate enzymes that are able to destroy connective tissue fibers between reduced enamel epithelium and oral epithelium by desmolysis and help in eruption. Premature degeneration of the reduced enamel epithelium may prevent the eruption of a tooth by non-desmolysis of connective tissue resulting into soft tissue impaction.

AMELOGENESIS (ENAMEL FORMATION)

The formation of any hard tissue essentially involves the laying down of an organic matrix and mineralization of this matrix. Enamel is different from other hard tissues, which are all derived from connective tissue. Enamel is ectodermal in origin; it has a unique organic matrix and a different pattern of mineralization. Amelogenesis (enamel formation) is a complex process. Two processes are involved in the formation of enamel: A. Organic matrix formation, B. Mineralization and maturation of enamel

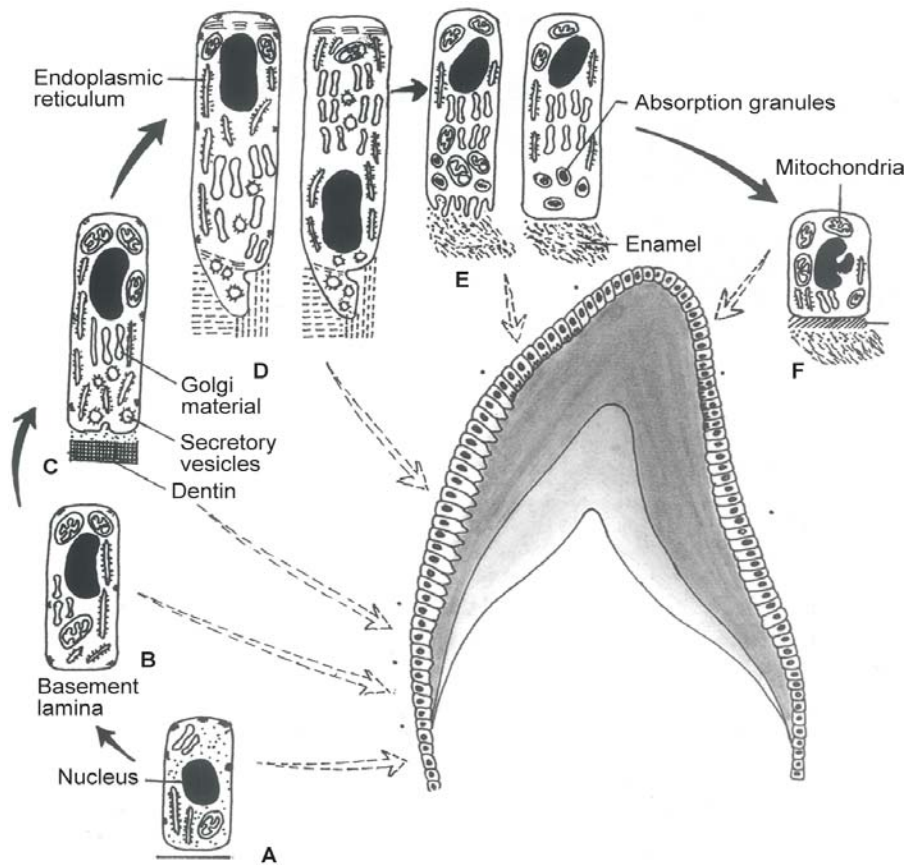


Figure 4.48: Cytologic changes seen at different stages in the life-cycle of the ameloblast in various regions of the mandibular canine

- A. The morphogenic stage –at cervical region. The cell is relatively undifferentiated.
- B. The organizing stage- it is the next stage which is just incisal to the cervical region. The cell is elongated the nucleus moves to the part of the cell which is farthest from dentin. The cell induces the differentiation of odontoblast.
- C. and D. The secretory stages- it comes after organizing stage at the middle third of the border, initial matrix secretion begins later on. This thin layer will be continuous with inter rod enamel. The cell has to migrate to provide room for Tomes' process.
- E. The maturative stage- it comes after secretory stage in the incisal third region labially. It shows the complex folding of plasma membrane to form striated border.
- F. The protective stage- it is the last stage seen at the incisal third region on lingual side. It shows the absence of the striated border and the cells become short.

matrix. The initiation of mineralization is started before completion of matrix formation. Usually, mineralization starts at cuspal and incisal region while matrix is being laid down at cervical region of the tooth.

Organic Matrix Formation

The ameloblasts start their secretory activity after a thin layer of dentin has been laid down. As enamel deposition proceeds, a thin continuous layer of enamel is formed along the dentin. This is called the dentinoenamel membrane. This separates the distal ends of the enamel rods and dentin. Major proteins of enamel matrix are

amelotin, amelogenin, ameloblastin enamel in and tuftelin. Amelotin was secreted by mature ameloblast and help in enamel formation. Amelogenins form small nanospheres, between which enamel crystals are formed and play important role in maintaining the spaces between the crystals. Deficiency of amelogenin result in hypoplastic teeth. Amelogenins also help in the formation of cementum. Ameloblastin and enamelin help in crystal growth and nucleation. Deficiency of ameloblastin results in lack of structural layer of enamel formation. Tuftelin is present at dentinoenamel junction and help in cell signaling.

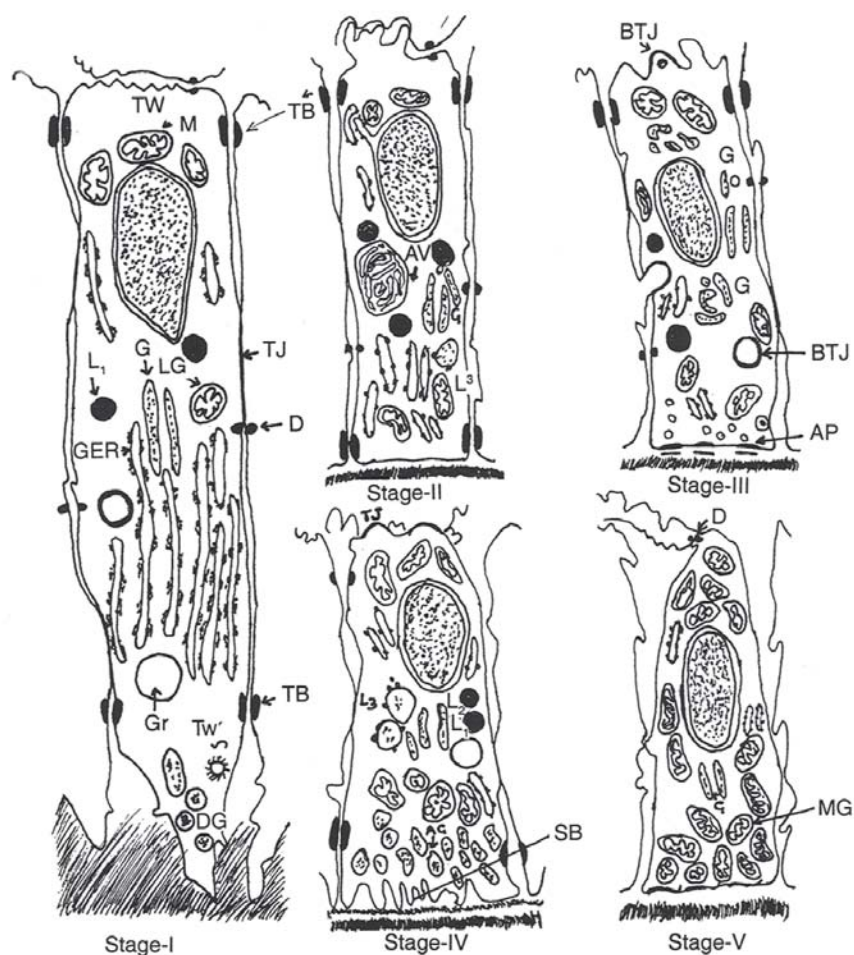


Figure 4.49: Drawings from electron micrographs of enamel formation. Individual ameloblast from five stages formative to maturation. Organelles AG= Absorption granules, AP= Apical contact specialization (Hemidesmosomes), AV= Autophagic vacuole (Lysosomes), BTJ= Bulb type of contact, D = Desmosomes, DG = Dense granules, G = Golgi apparatus, GER = Granular endoplasmic reticulum Gr = Pale granules, L1 L2 L3 = Lysosomes. LG = Lipid granules, M = Mitochondria, MG = Mitochondrial granules, SB = Striated border, TB= Terminal bar. TJ= Tight junction, TW = Terminal web. (From Reith E J: J, *Ultrastruct Res* 30: 111,1970)

Development of Tomes' Process

The surface of the ameloblast facing the developing enamel is rough. The interdigitation between ameloblasts and enamel rods exists because the long axes of the ameloblasts are not parallel to the long axes of the rods. The projections of the ameloblasts into enamel matrix are called Tomes' processes. Tomes' processes are partly delineated by incomplete septa. These also contain secretion granules, endoplasmic reticulum and mitochondria (Figs 4.50 and 4.51).

Once the Tomes' process is established, secretion of enamel protein by secretory granules occurs through narrow channels. These two channels are located at two different regions in the Tomes' process. These are at the

distal end and at the proximal part near junction of adjacent process (Figs 4.52 and 4.53).

The proximal secretory sites completely encircle the Tomes' process. The secretion at the proximal site precedes that at the distal extremity. Because of this, walled pits are formed, which are occupied by distal end of the process. These pits are filled by secretion from the distal end of the Tome's process. Now the rod-like structure of enamel rod is formed.

Secretory Ameloblast

At least two ameloblasts are involved in the synthesis of each enamel rod and contain typical secretion granules as well as rough endoplasmic reticulum and

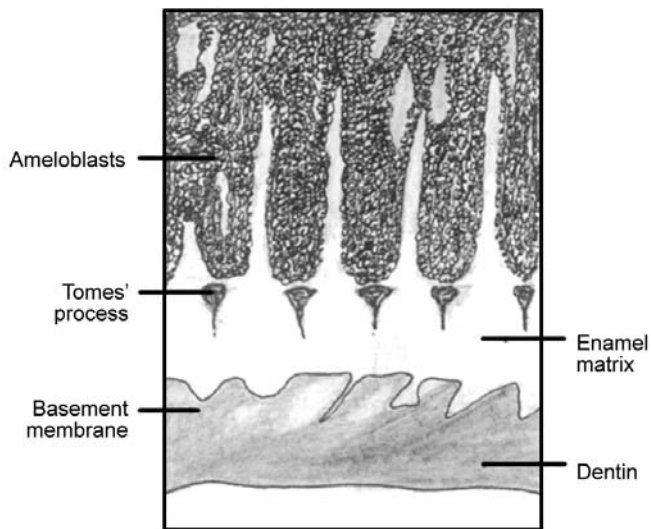


Figure 4.50: Tomes' process showing "picket fence" arrangement. Rods are at an angle to ameloblasts and Tomes' process

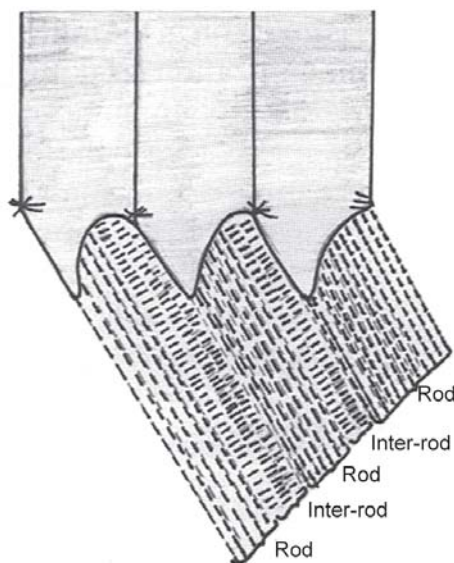


Figure 4.51: The relationship between enamel rods and Tomes' process formation. The formation of interprismatic and prismatic enamel from ameloblasts are seen. The long axes of the ameloblasts are not parallel to the long axes of the rods. The projection of the ameloblasts into enamel matrix have been termed Tomes' process

mitochondria. The bulk of the head of each rod is formed by one ameloblast, whereas three other ameloblasts contribute to the total formation of each rod. The maximum portion of the head' of each rod is formed by one ameloblast. The other three ameloblasts contribute to the tail of each rod. Therefore, each rod is formed by four different ameloblasts and each ameloblast contributes in the formation of four rods.

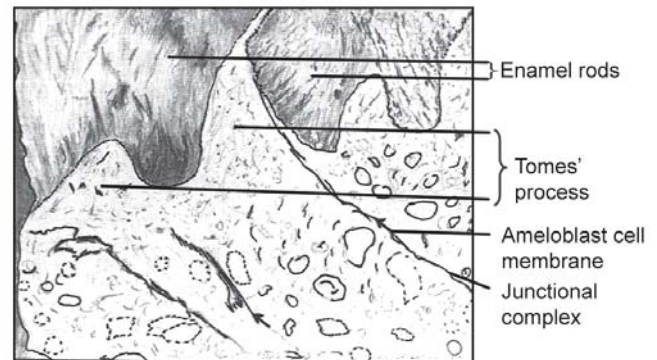


Figure 4.52: Ends of ameloblast cells and adjacent enamel rods are nearly perpendicular, which indicates approximate position of Tomes' process (As observed by electron microscope)

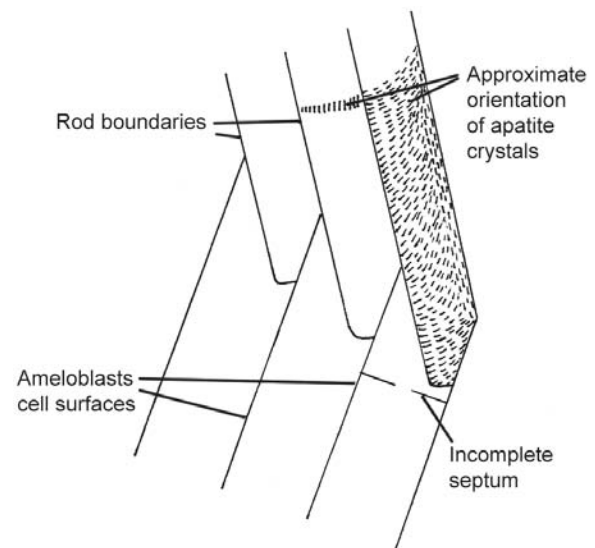


Figure 4.53: Drawing showing that long axes of the ameloblasts are not parallel to long axes of the rods and each enamel rod is formed by at least two ameloblasts

Distal Terminal Bars

When the Tomes' process begin to form, terminal bars appear at the distal ends of the ameloblast. They separate the Tomes' process from the ameloblast cell proper. Structurally, distal terminal bars are localized condensations of cytoplasmic substance, which is closely associated with the thick cell membranes.

Maturing enamel covered with ameloblasts Maturing enamel is covered with smaller sized ameloblasts than the ameloblasts over incompletely formed enamel. These smaller ameloblasts are packed with mitochondria.

Transition stage – In this stage, changes occur in the ameloblasts which are as follows

1. Reduced height of ameloblast.
2. Secretion of enamel stops completely.
3. Removal of amelogenin starts.
4. Apoptosis of 50 percent ameloblasts
5. Autophagocytosis of protein synthesis organelles.
6. Deposition of basal lamina by ameloblasts.
7. Attachment of ameloblast to the basal lamina by hemidesmosomes.

Modulation – During maturative stage, in the apical cytoplasm developing ameloblasts change alternatively in ruffled borders and smooth borders ameloblasts in a cyclic manner in cervicoincisal direction. This change is known as modulation and occur in every 5 to 7 hours. During maturative phase, size of ribbon shaped crystals are increased from 1.5 to 25 micron and after tooth eruption crystal size increases due to ionic exchange with saliva.

Difference between Enamel and Other Hard Tissues

Formation of any hard tissues of the body essentially involves the laying down of organic matrix into which subsequently mineral salts are deposited. Enamel is ectodermal in origin while other hard tissues are derived from connective tissue. Enamel has a unique organic matrix and distinctive pattern of mineralization. Enamel formation is a complex process, which comprises two main stages (a) secretion and (b) mineralization and maturation. During enamel mineralization, organic components and water are lost while in other hard tissues during mineralization, organic components and water are not lost. More than 90 percent of initially secreted protein is lost during enamel maturation. The remaining protein surrounds the individual crystals.

Mineralization and Maturation of the Organic Enamel Matrix

Mineralization of the organic matrix takes place in two stages. The time interval between the two stages is very small. In the first stage, immediately partial mineralization (25% to 75% of eventual total mineral content) occurs in

the organic matrix segments and the interprismatic substance as they are laid down. First, mineral is deposited in the form of crystalline apatite. The first stage is immediately followed by the second. In the second or maturation stage, the gradual completion of mineralization takes place. The process of mineralization starts from the cusps and incisal edges of the crown and progresses cervically. At each rod level mineralization and maturation begin at the dentinal end. In this way there is an integration of two processes. The rod maturation takes place from dentin side and the sequence of rod maturation is from cusp and incisal edge of the crown towards the cervical line.

Maturation starts before the matrix has reached its full thickness (Fig. 4.53). While the maturation of inner, first formed enamel is going on in the inner first formed matrix, the initial mineralization is taking place in the recently formed outer matrix. In maturation, growth of the enamel crystals is seen. Due to the loss of organic material and water, the organic matrix gradually becomes thin to make room for the growing crystals.

Organic and inorganic substances can be separated by either decalcification or incineration. In the process of decalcification the organic constituents can be retained (Fig. 4.54). The enamel, being over 90% mineral, is lost after decalcification (Fig. 4.55).

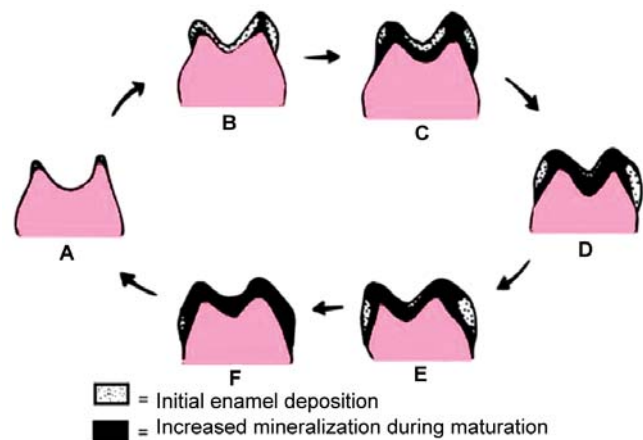


Figure 4.54: A molar tooth showing pattern of mineralization during maturation. Maturation commences before full thickness of enamel is formed in any area. Mineralization during maturation shows a different pattern than initial deposition

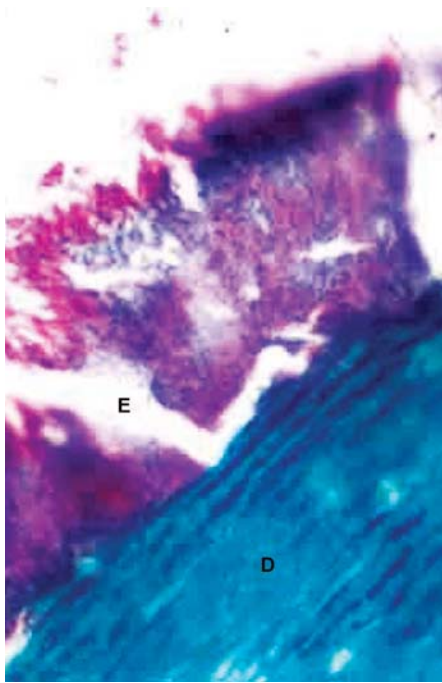


Figure 4.55: Masson's trichrome stain: Absence of collagen fibers can be seen in enamel (E). Organic matrix is indicated by the red stain picked up by hypomineralized enamel while dentin (D) stains green due to collagen matrix. (x 400)

CLINICAL CONSIDERATIONS

Caries on Enamel

The following are the structural predisposing factors of caries on the enamel surface.

- Deep enamel pits and fissures:** They provide potential space for the retention of food and bacteria and produce caries. Because of thin enamel at the floor of pits and fissures, caries reaches the dentin very early (Fig. 4.56). It spreads along dentino-enamel junction undermining the enamel. This results in a big carious lesion without giving any signs and symptoms because of the minute entrance of the cavity. For their detection, careful clinical and radiographic examination is required.
- Dental Lamellae:** They are also caries prone as they contain much organic material. (Fig. 4.57).
- Plaque deposits:** Generally plaque deposits at the gingival third of the teeth. It has a predilection for surface cracks, pits and fissures of occlusal surfaces, overhanging margins. These are mainly the areas that are protected from the normal mechanical cleaning action of the tongue, the cheeks and the lips (Fig. 4.53).

- Calculus:** It is an adherent calcified or calcifying mass that forms on surface of natural teeth and also on dental prostheses. Generally it is mineralized bacterial plaque. Prevention of calculus formation is dependent upon

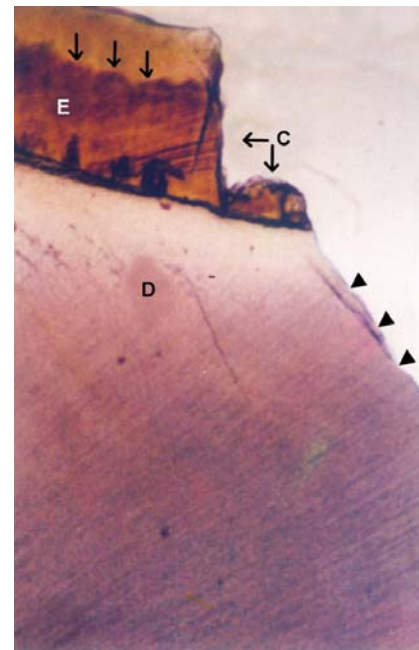


Figure 4.56: Caries of enamel: Ground section showing margins of the cavity (C) caused by caries of enamel (E). Margins of the cavity are irregular. Caries is seen extending to dentin (D) also indicated by arrowhead. The enamel adjacent to the cavity shows deep brownish discoloration indicated by arrows x 40

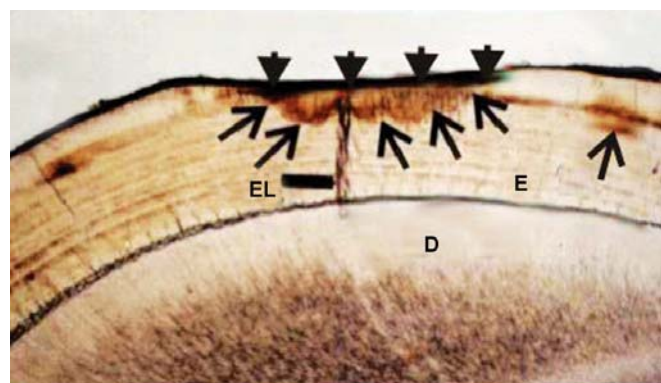


Figure 4.57: Smooth surface caries of enamel. The surface layer of the enamel (E) indicated by the arrow heads is intact. Area beneath the surface indicated by arrows shows brown pigmentation of enamel due to early caries. Pigmentation occurs due to breakdown products or due to food etc. Lateral spread of caries is also seen and indicated by arrows. EL= Enamel Lamella, D= Dentin. (Ground Section x 50)

removal by the patient of the fresh, uncalcified deposits with toothbrush and dental floss (Figs 4.20 and 4.58)

Supragingival plaque strongly influences the growth, accumulation and pathogenic potential of subgingival plaque. These microbial aggregations on tooth surface should be prevented from depositing so that it may be compatible with gingival health.

Cervical region of enamel—If cervical region of enamel is rough, or decalcified then food debris gets accumulated on this area and results in gingival inflammation. To avoid this condition, cervical region should be kept smooth and polished by proper dental management.

Cavity Preparation

During cavity preparation, the course of unsupported enamel rods is important. Unsupported enamel rods should not be left at the cavity margins because under masticatory forces they break and produce leaky margins, that would ultimately lead to failure of the restoration (Fig. 4.59). Direction of rods at cervical regions is different in deciduous and permanent teeth. This is the reason for a different type of cavity preparation at cervical region of deciduous and permanent dentition as the directions of enamel rods are different in this region.

Pits and Fissures

During development, faulty coalescence of the lobes occurs resulting in very deep pits and fissures (Figs 4.60 A and B). In very deep pits and fissures, food and bacteria may get trapped, that may predispose the tooth to dental caries.

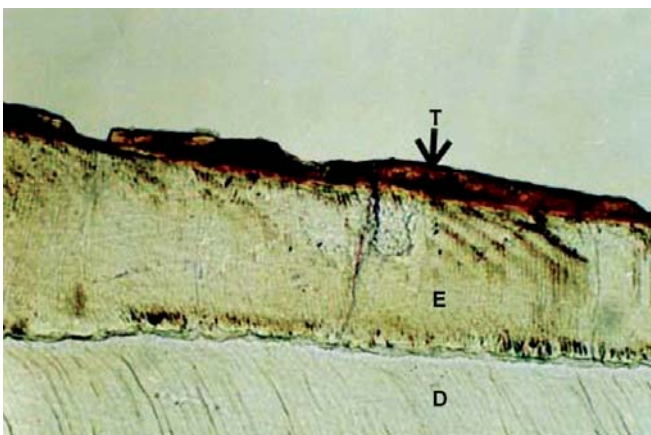


Figure 4.58: Tartar: Ground section of tooth showing thin layer of tartar (T) deposited on the surface of enamel (E), D= Dentin (x 100)

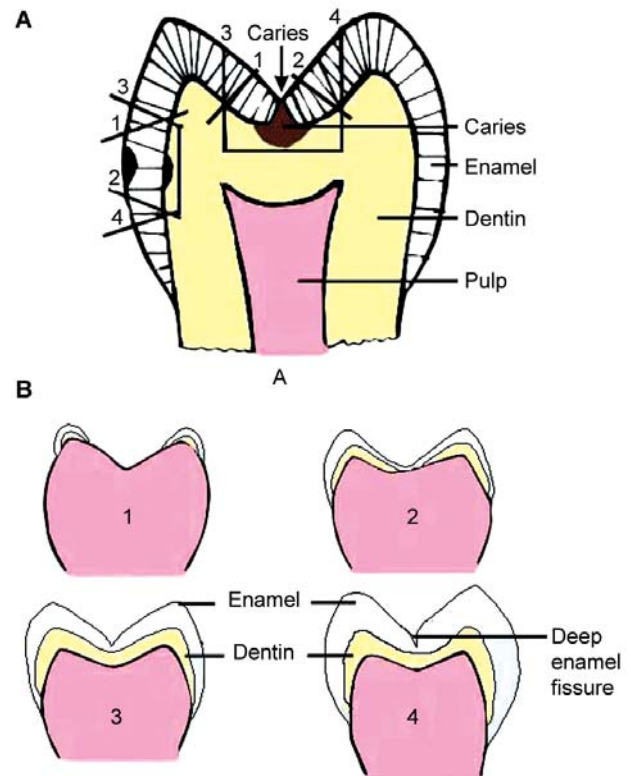


Figure 4.59: A. Course of enamel rods in relation to cavity preparation in molar. 1 and 2 showing incorrect preparation of cavity margins, 3 and 4 showing correct preparation. B. Development of deep enamel fissure. Thin enamel layer forming floor of fissure

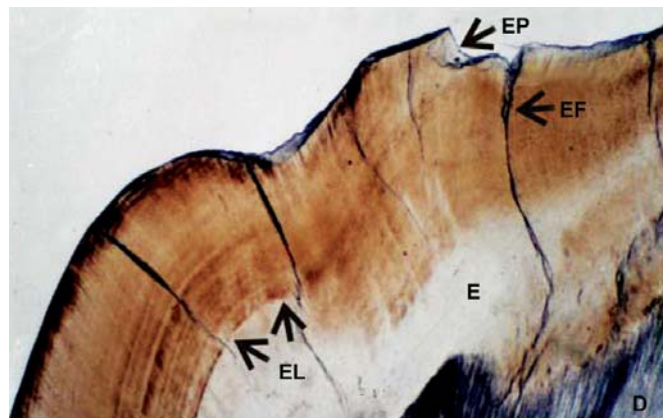


Figure 4.60A: Enamel pits and fissures: Ground section of tooth showing irregular depression in the surface of enamel (E) called enamel pit (EP) and a crack, enamel fissure (EF) leading downward from the enamel pit. These pits and fissures act as entry point of bacteria and predispose the tooth to caries. (EL) Enamel Lamellae, (D) Dentin (x 100)



Figure 4.60B: Enamel pit: Ground section of tooth showing a depression in the cuspal surface of crown (indicated by arrow). This is called enamel pit x 40

When the defect involves one-fourth or less of the thickness of the enamel, then it should be corrected by enameloplasty. When it involves more than one-fourth of the thickness of the enamel, fillings should be done.

Dens Invaginatus

Dens invaginatus is a developmental variation. It is recognized as an invagination of enamel into dentin. (Figs 4.61 and 4.62).

To prevent caries, pulp infection and premature loss of the tooth, this condition should be recognized early. The tooth should be prophylactically restored. This defect can be recognized radiographically before eruption of the tooth.

Shade Selection

Enamel appears temporarily whiter within minutes when dried and when the tooth is isolated from the moist environment by rubber dam or absorbents. Thus the shade must be selected before isolation of the tooth.

Acid Etching

Phosphoric acid etching is done on enamel surface for the micromechanical 'bonding' of composite restorative materials or pit and fissure sealants with enamel. For this,

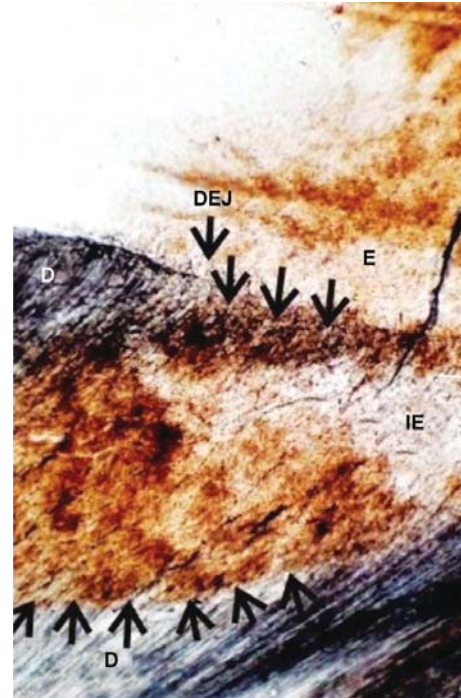


Figure 4.61: Dens invaginatus: Ground section of tooth showing inclusion of enamel (invagination) into dentin. The normal enamel (E) is seen above the dentinoenamel junction (DEJ). Invaginated enamel (IE) is located below the DEJ and is surrounded by dentin (D) (x 100)

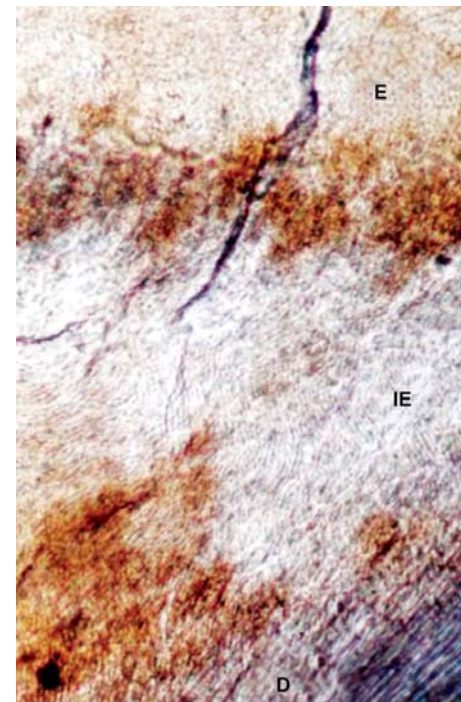


Figure 4.62: Dens invaginatus: High magnification showing wavy bundles of enamel rods of invaginated enamel (IE) located within dentin (D). Ground section (x 400)

30 to 40 percent of phosphoric acid is applied for 60 seconds. It etches ends of the enamel rods, provides increase in surface areas by roughness and undercuts, providing more adherence of sealant to enamel rod ends. The rod sheath contains more organic material than the rod itself and offers greater resistance to demineralization than the rod. This etching produces an irregular and pitted surface with numerous microscopic undercuts by an uneven dissociation of heads and tails of enamel rods. Therefore, the purposeful and controlled acid etching of the enamel rods is done to provide microretention for composite or sealant (Fig. 4.63).

Fluoridation

Systemic and topical fluoride treatment causes formation of hydroxyfluorapatite, which is more resistant to acid attacks and thus more resistant to dental caries. If fluoride is present during amelogenesis, all the enamel crystals become more resistant to acid dissolution. The concentration of fluoride must be carefully and precisely controlled because of the sensitivity of secretory ameloblasts to the fluoride ion, giving rise to discolored mottled enamel. Due to the semipermeable nature of enamel, topical fluoride, fluoridated toothpastes and fluoridated drinking water provide a higher concentration of fluoridation in the surface enamel of erupted teeth.

Bleaching

Effects of carbamide peroxide type of whitening agents on enamel surfaces have been studied. It was found that

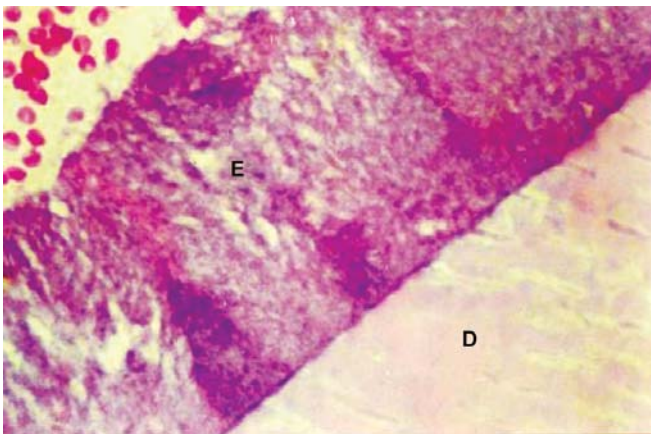


Figure 4.63: Partly decalcified section of tooth: Enamel (E) shows organic matrix enclosing empty spaces representing dissolved prisms during the process of decalcification. D= Dentin (H and E x 200)

they cause very minute pitting on the surface of the enamel and form an initial carious type of lesion.

Lateral Spread of Caries

When caries reaches the dentinoenamel junction, the hypocalcified tufts allow lateral spread along this junction.

Remineralization

Etched enamel can be remineralized by sodium and stannous fluoride solutions. Fluoride ion penetrates the porous etched enamel surface. In low concentrations, fluoride stimulates remineralization.

Will Natural Enamel Restorations be Possible Soon?

It is hoped that very soon current restorative materials will be replaced with dental restorations which will be identical and similar to natural tooth enamel. The tiny spheres that regulate the formation and organization of tooth enamel by controlling the crystalline growth of the substance have been identified. These spheres are called nanospheres. Their diameter is 18 to 20 nanometers. They are formed by a naturally occurring family of proteins which are specific to tooth. These proteins are called amelogenins. Although we cannot make enamel but it is very clear how nature does it.

Formation of Enamel

Amelogenins self-assemble in the shape of enamel and form the extracellular matrix. Then within this, the formation of inorganic enamel crystals starts. In the beginning, the crystals grow only at their end surfaces. The nanospheres act as spacers and build a scaffold on which finally the mature enamel is formed. Nanospheres have the capacity to regulate the growth of crystals. The enzymes break down the amelogenins. Thereafter, the crystals start to grow on all of their surfaces. By this growth on all the surfaces, the crystals become thick and clump together. In this way mature enamel is formed. The mean rate of enamel formation is 3.5 microns per day. The rate of enamel formation increases from cervical region to incisal or occlusal region and from inner to outer surface of enamel.

By taking a gene for an amelogenin from a mouse in 1994, an identical recombinant amelogenin was produced.

For this a bacterial reproductive process was used. This recombinant amelogenin can self-assemble to make nanosphere structures. These structures are identical to those that are observed in the enamel of humans and other animals. Nowadays, the crystals are given within synthetic matrices that are made from recombinant amelogenins.

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Dentin

- Introduction
- Physical and chemical properties
- Structure
 - Odontoblastic processes
 - Dentinal tubules
 - Peritubular dentin (intratubular dentin)
 - Intertubular dentin
 - Predentin
- Development of dentin
 - Differentiation of odontoblasts
- Dentinogenesis
- Types of dentin
- Structural lines
- Innervation of dentin
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- Clinical considerations

INTRODUCTION

Dentin is a hard tissue, which is also mildly elastic in nature. It is also called substantia eburnea. It forms the bulk and provides general form and shape to the tooth. It is covered by the enamel in the crown and by the cementum in root. A thin layer of dentin is formed before the enamel and cementum are formed. Therefore dentin determines the shape of the crown and also the number and size of the roots. Internally, dentin forms the wall of the pulp cavity. Both the dentin and the pulp are embryologically derived from dental papilla. Dentin is characterized as a hard tissue with tubules throughout its thickness. The dentinal tubules contain cytoplasmic processes given off by specialized cells, called odontoblasts. Dentin is considered as a vital tissue. It reacts to stimuli because it contains living protoplasm in its cytoplasmic processes. Dentin resembles bone both physically and chemically, but differs from bone in the fact that it contains only protoplasmic processes of the odontoblast cells, while in the bone, osteocytes are enclosed within the bony matrix.

PHYSICAL AND CHEMICAL PROPERTIES

The color of dentin varies from light yellow in deciduous teeth to pale yellow in permanent dentition, becoming darker with age. The lower content of mineral salts in dentin renders it more radiolucent than enamel. It is less hard than enamel but more hard than bone or cementum. Dentin hardness varies between crown and root dentin in a tooth and between different types of teeth. Dentin hardness is more in the central part than near the pulp or at its periphery. Dentin is harder in permanent teeth than in deciduous teeth. Dentin is mildly elastic and subject to slight deformation, hence supports the brittle enamel under masticatory forces. The modulus of elasticity of dentin is 1.67×10^6 PSI. Dentin has a density of 22.1 gram/milliliter and its Knoop hardness number is 68. It is semi-transparent and highly permeable due to the presence of dentinal tubules. Permeability decreases with advancing age. On the basis of a study, Shamala Chandra, Satish K Khera, Keith V Krell concluded the following:

- The permeability of young dentin along the proximal surfaces gradually increases occluso-gingivally. The cervical third is the most permeable and the occlusal third is the least permeable.

- There is no statistically significant difference seen in the permeability on the mesial and distal surfaces of the same teeth.
- In the scanning electron micrographs, it appears that there were more tubular openings in the cervical third of proximal sections as compared to the occlusal and the middle thirds.

Dentin consists of 65 percent inorganic matter and 35 percent organic matter and water. Tencate has reported that mature dentin contains approximately 70 percent inorganic material, 20 percent organic material and 10 percent water by weight. By volume, mature dentin contains approximately 45 percent inorganic material, 33 percent organic material and 22 percent water. The inorganic component is made up of hydroxyapatite and several trace elements. Hydroxy-apatite has formula $3\text{Ca}_3(\text{PO}_4)_2 \cdot \text{Ca}(\text{OH})_2$ (present in enamel, dentin) and formula of poorly crystallized calcium deficient hydroxyapatite is $\text{Ca}_{10}(\text{OH})_2 (\text{PO}_4)_6 \cdot \text{Ca}_{10}(\text{PO}_4)_6 (\text{OH})_2$ which is present in cementum and bone. The hydroxyapatite crystals of dentin are rich in carbon, poor in calcium and smaller in size in comparison to the crystals of enamel. The organic component is made up of predominantly collagenous fibrils (Type I) and ground substance of mucopolysaccharides, glycosaminoglycans, proteoglycans, and phosphoproteins with small amounts of citrate, chondroitin sulfate, insoluble protein and lipids. Hydroxyapatite crystal of dentin is much smaller than enamel. Proteins that are only present in dentin are dentin sialoprotein and dentin phosphoprotein.

STRUCTURE

The dentin is composed of the following:

Odontoblasts, bodies of which are arranged in a layer on the pulpal surface of dentin and their cytoplasmic processes are seen inside the dentinal tubules, which extends into the dentin.

- Numerous tubules called the dentinal tubules, which contain protoplasmic processes of odontoblast (Tomes' fiber). These tubules extend from the pulp towards the periphery and join the enamel at the dentinoenamel junction in the crown. They join the cementum at the dentino-cemental junction in the root.
- As dentin calcifies, the hydroxyapatite crystals mask the individual collagen fibers.

Odontoblastic Processes

The cytoplasmic extensions of the odontoblast are called odontoblastic processes. The cells are present in the peripheral pulp at the pulp- predentin border. Their processes extend into the dentinal tubules. They narrow as they go further into the dentin nearer to the enamel (one micron in diameter), but are largest in diameter near the pulp (3 to 4 microns). The cell bodies of the odontoblast are about 7 microns in diameter and about 40 microns in length. Therefore, the odontoblastic processes become narrow to about half the size of the cell as they enter the dentinal tubules. Some processes extend throughout the thickness of dentin. In other areas of dentin where the tubules are obliterated by mineral deposits, a shortened process may be present. The odontoblastic process is made up of microtubules of 20 micron in diameter and smaller filaments about 5 to 8 micron in diameter. Sometimes mitochondria, microvesicles and bodies resembling lysosomes are present.

These processes divide near the dentinoenamel junction and may extend into the enamel and then are called as enamel spindles. Sometimes side branches appear along the length of the processes, which extend into the adjacent tubules (Fig. 5.1).

Dentinal Tubules

The dentinal tubules are sigmoid ('S') shaped curved structures which run perpendicularly from the pulp toward the periphery. Near the root tip, incisal edges and cusps, the dentinal tubules are almost straight. (Figs 5.2 A and B, and 5.3). The first convexity of the doubly curved dentinal tubules is directed towards the apex of the tooth. Because of their peculiar curved shape, the dentinal tubules are longer than the thickness of dentin. The thickness of dentin is between 3 to 10 mm. The diameter of the dentinal tubules is larger at the pulpal side (1.5 to 3 microns) than at the dentino-enamel junction (one-

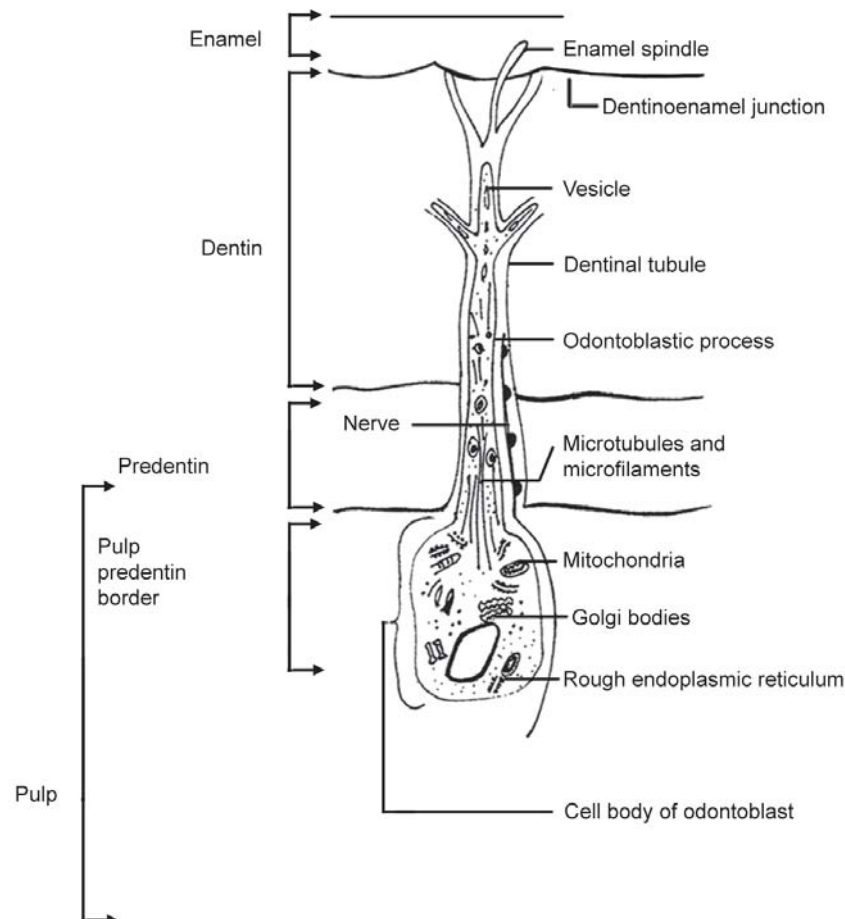


Figure 5.1: The odontoblast and its process in the dentinal tubule

micron). The number of tubules per unit area at the pulpal end is nearly four to five times more than at the outer surface of the dentin. The ratio between the outer and the inner surfaces of dentin on an average is 5:1. Therefore, the tubules are farther apart in the outer layers and closer together near the pulp (Fig. 5.4).

In other words the tubules are widely separated at the outer surface of the dentin whereas they are crowded together near the pulpal cavity. The root dentin has lesser tubules compared to the crown. The dentinal tubules show more branching at their terminal parts (Figs 5.5 and 5.6). The lateral branches are called as canaliculi or microtubules. These microtubules originate at right angles to the main tubules every one to two microns

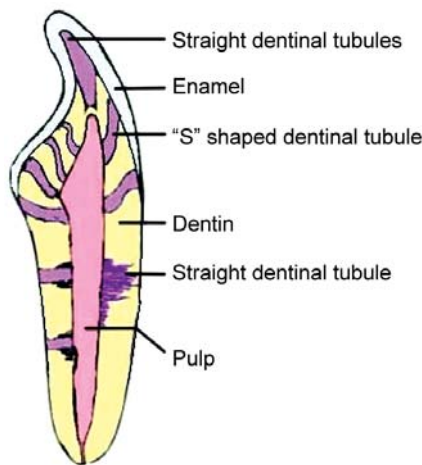


Figure 5.2A: Dentinal tubule in crown show "S" shaped course but at incisal tip and in the root of tooth course of the dentinal tubule is straight as seen in the ground section of incisor

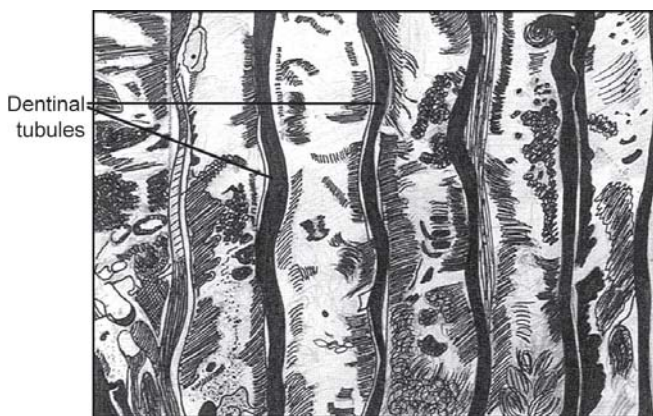


Figure 5.2B: Longitudinal section of dentinal tubules (as seen under scanning electron microscope)

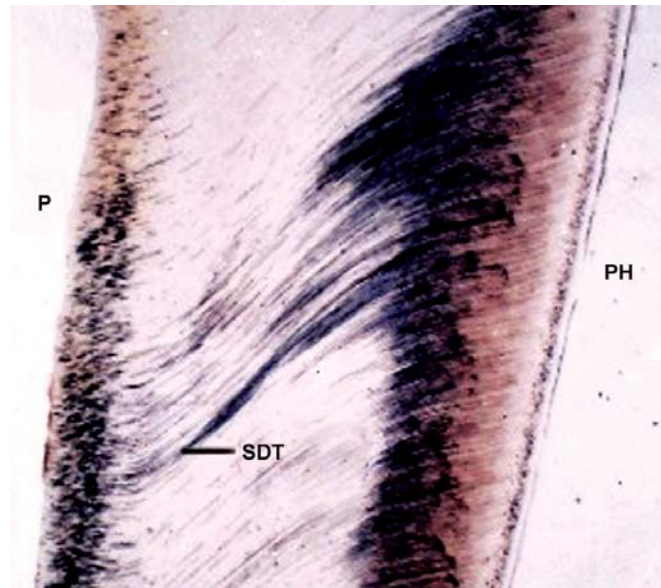


Figure 5.3: S-shaped dentinal tubules (SDT) running from pulp (P) to the periphery (PH). The first curve of S is towards the apex of the root. Ground section (x 100)

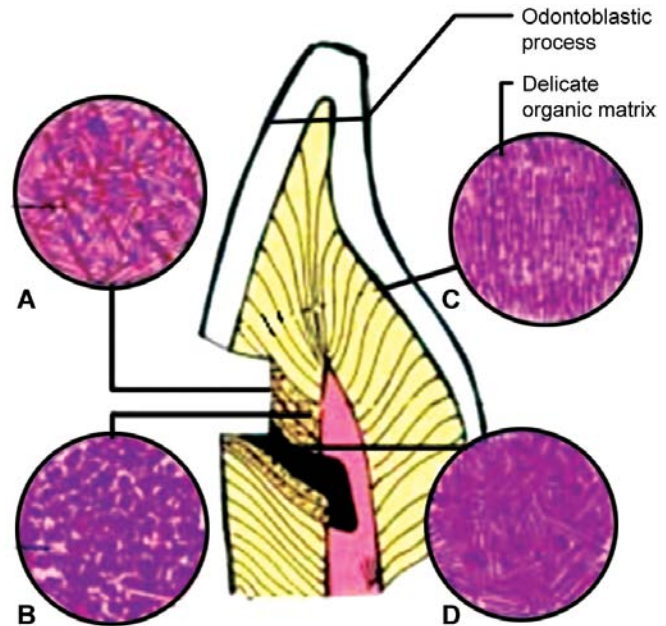


Figure 5.4: The size, curvature and distance between tubules in outer, mid and inner surface of dentin. The tubules are approximately one micron in diameter at their outer end (A) 1.5 to 2 micron midway through dentin (B) and 1.5 to 3 micron near the pulpal end (C) Microbial penetration follows line of least resistance to reach the pulp (D). Approximately 30,000 dentinal tubules are exposed by cutting 1 mm² dentin surface. Cut dentinal tubules in the floor and walls are often different

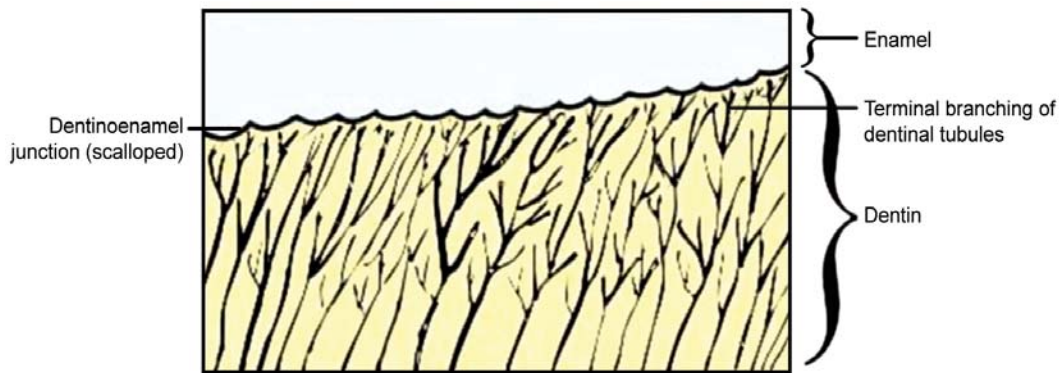


Figure 5.5: Terminal branching of dental tubules near the dentinoenamel junction. The branching of dental tubule in root dentin is more profuse than in coronal dentin (Ground, longitudinal section)

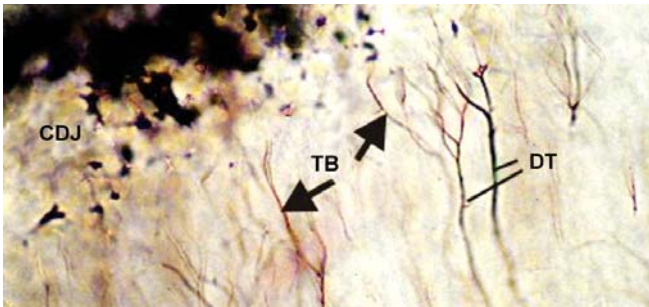


Figure 5.6: Terminal branching (TB) of dental tubules (DT) near cementodentinal junction (CDJ). Ground section (x 400)

along its length. These microtubules are one micron or less in diameter and enter the intertubular dentin. Some of them enter the adjacent or distal tubules. The primary coronal dentinal tubules have a different structure compared to root and reparative dentin. The dentinal tubules also show branching and loop formation near the cementum. This is seen as a granular layer in ground sections. The dentinal tubules contain the odontoblastic processes and a thin organic sheath lining the dentinal tubules. This layer is called lamina limitans. The number of dentinal tubules decreases with increasing age. This result may account for the reduced sensitivity. A few dentinal tubules extend through the dentinoenamel junction into the enamel for several millimeters. These are termed as enamel spindles.

Peritubular Dentin (Intratubular Dentin)

A cross-section of the dentinal tubules when viewed in a ground section shows the dentin that immediately surrounds the dentinal tubules and is termed peritubular dentin or intratubular dentin. The intratubular/

peritubular dentin has more mineral content and less collagen fibers than the intertubular dentin. Radiographic studies of cut sections have shown the increased mineral density in the peritubular dentin (Figs 5.7 to 5.9). The peritubular dentin forms the walls of the tubules in all the dentin. The continuous deposition of peritubular dentin causes reduction in the size of tubular lumen. Several investigators believe that the calcified tubule wall has an inner organic lining termed the lamina limitans. This is a thin organic membrane, which is high in glycosaminoglycan (GAG). The intratubular dentin is formed after the intertubular dentin formation occurs (Figs 5.10 and 5.11).

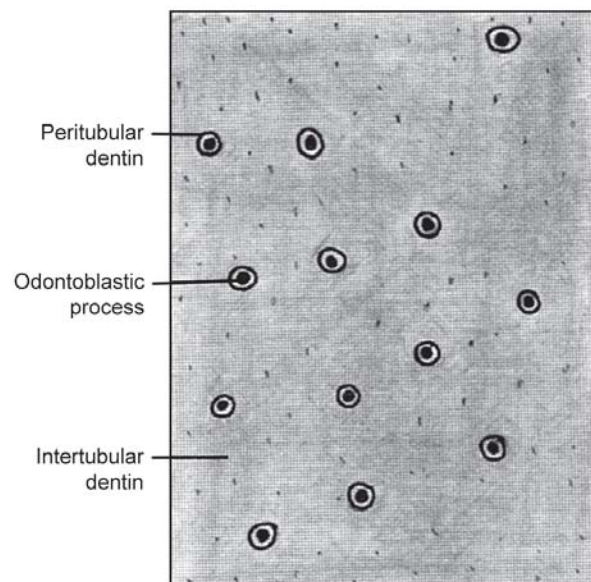


Figure 5.7: Increased mineral density in the peritubular dentin shown with soft roentgen ray

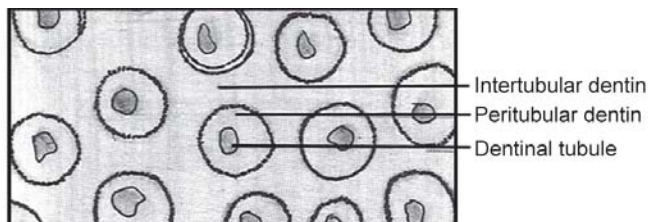


Figure 5.8: Dentinal tubules appear as circular regions and peritubular dentin surrounds the dentinal tubules and intertubular dentin is in between dentinal tubules (as seen in transverse ground section)

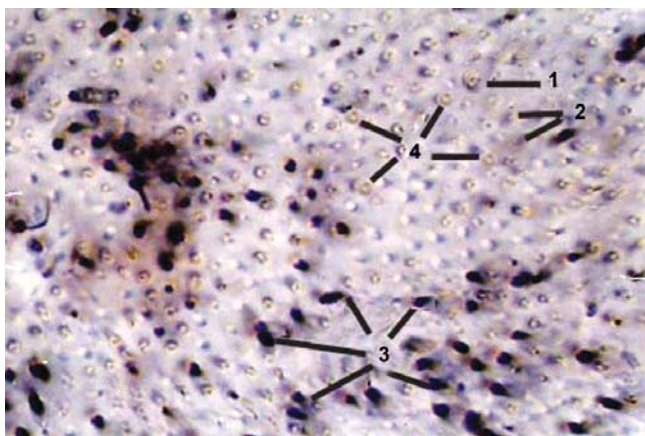


Figure 5.9: Cross-section of dentinal tubules (Ground section): The hollow dentinal tubules are surrounded by a mantle of peritubular dentin (1) and separated by more abundant intertubular dentin (2) Empty tubules filled with air (dead tracts) appear black in transmitted light (3) Odontoblastic process (4). (x 150)

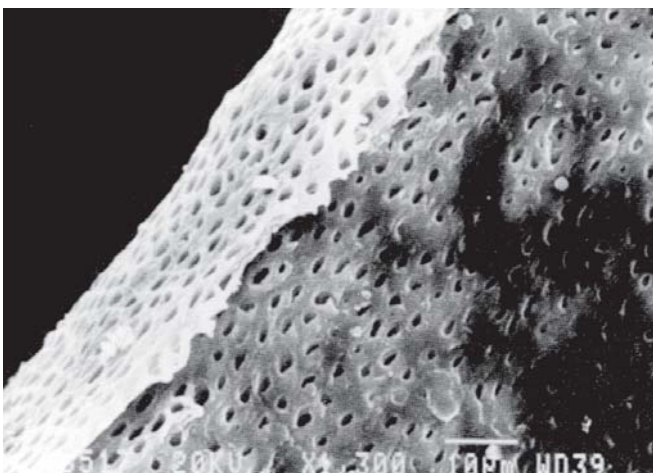


Figure 5.10: Transverse section of dentin from area near the root canal showing dentinal tubules seen as punched out holes in the dentin matrix. Electron micrograph. (x 3000)

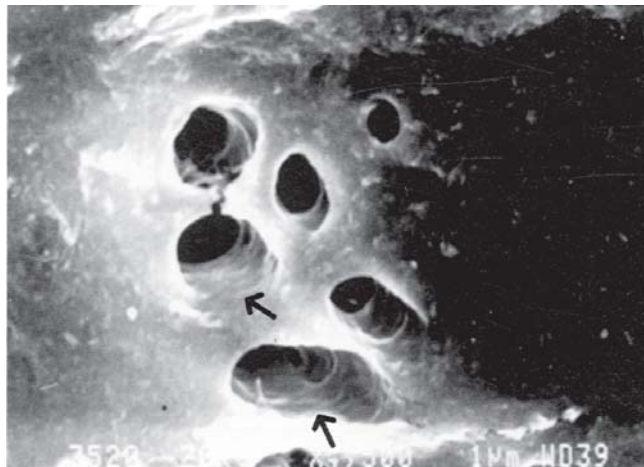


Figure 5.11: Slightly oblique cross-section of dentinal tubules showing walls of dentinal tubules very clearly indicated by arrows. Electron micrograph (x 4,500)

The formation of intratubular dentin is a slow continuing process. It can be accelerated by external stimuli. Intratubular dentin is approximately two times as thick in outer dentin than in inner dentin. Because of its growth, the diameter of the dentinal tubule is constricted near the dentinoenamel junction (Figs 5.12 and 5.13). Periodontoblastic space is present between the odontoblastic process and peritubular dentin and it contains dentinal fluid.

Intertubular Dentin

The main mass of dentin is composed of intertubular dentin that is present between the dentinal tubules or peritubular dentin. In demineralized dentin, loss of peritubular dentin (Figs 5.14 and 5.15) occurs, so the tubule diameter will appear similar in the inner and outer dentin visualized with a light microscope. Although it is highly mineralized, this matrix consists of tightly interwoven network of type I collagen. The ground substance consists of glycoproteins, proteoglycans and plasma proteins. Although it is highly mineralized, this matrix, like bone and cementum, is retained after decalcification whereas peritubular dentin is not (Fig. 5.16). About one half of its volume is organic matrix, mainly collagen fibrils, which are randomly arranged around the dentinal tubules. The fibrils are 0.2 to 0.5 microns in diameter and show cross banding at 64 microns intervals, which is typical for collagen. Hydroxyapatite crystals, which are approximately 0.1 microns in length, are formed along

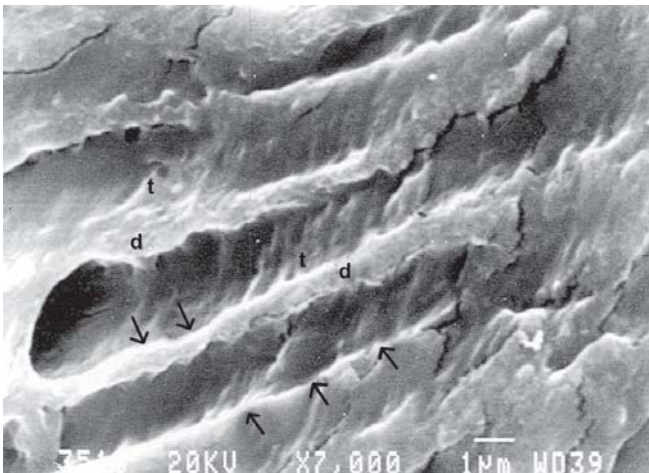


Figure 5.12: Longitudinal section of dentin showing cut open dentinal tubules (t). Arrows indicate dense mantle of peritubular dentin. (d) indicates intertubular dentin. Electron micrograph (x 7000)

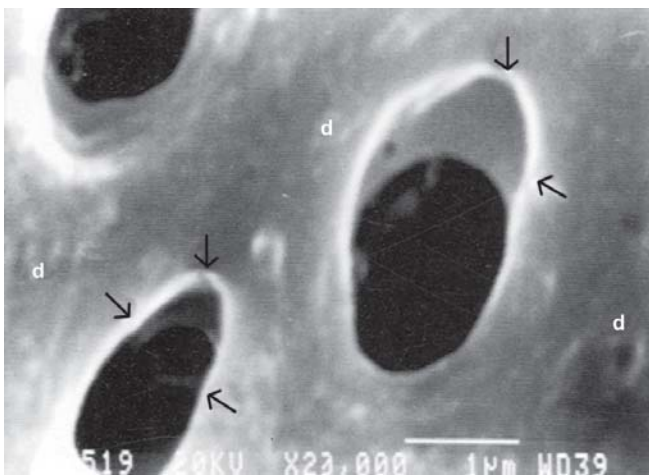


Figure 5.13: Electron micrograph of cross-section of dentinal tubules showing thick mantle of peritubular dentin indicated by arrows and intertubular dentin (d). (x 23,000)

the collagen fibers with their long axes arranged parallel to the collagen fibers.

Predentin

Predentin is the innermost portion of dentin that is not mineralized. The predentin is located adjacent to the pulpal tissue and is 2 to 6 microns thick. Its thickness depends upon the activity of the odontoblast. The predentin represents the first formed dentin (Figs 5.17 and 5.18). This predentin layer, secreted by the odontoblasts, is a protein carbohydrate complex consisting of proteglycans,

phosphoproteins, plasma proteins, glycoproteins and collagen fibrils. Calcium and phosphorus salts are deposited into this matrix (Figs 5.19 and 5.20). As the collagen fibers undergo mineralization at the predentin-dentin front, the predentin then becomes dentin and thus a new layer of predentin forms around the pulpal tissue. The periodontoblastic space (the space around the odontoblastic process), and the space peripheral to the odontoblastic processes are filled with extracellular fluid. This fluid has an important role in sensory transmission. Unmyelinated vesiculated nerve endings for sensory perception are also found at the pulpal end of the periodontoblastic space of the dentinal tubules (Figs 5.17 to 5.21).

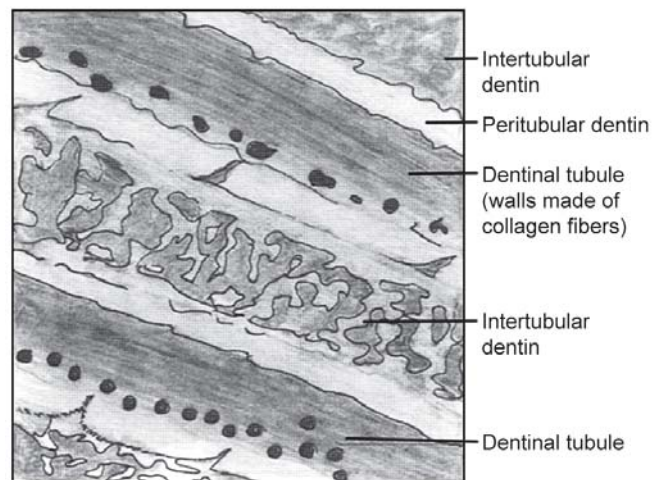


Figure 5.14: Peritubular dentin around the dentinal tubules, in between them is the intertubular dentin (as seen in scanning electron microscope)

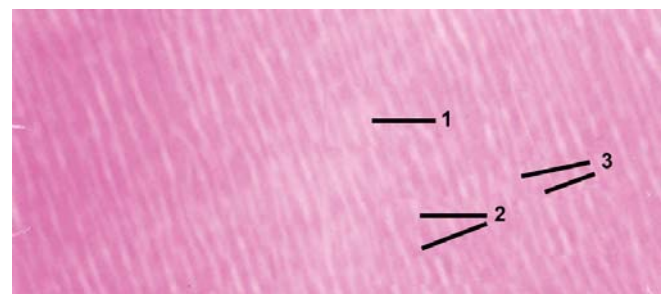


Figure 5.15: The hollow dentinal tubules (1) surrounded by a thin rim of denser peritubular dentin (2) and separated by more abundant intertubular dentin (3). Longitudinal section of dentinal tubules (H and E stained section of decalcified tooth x 150)

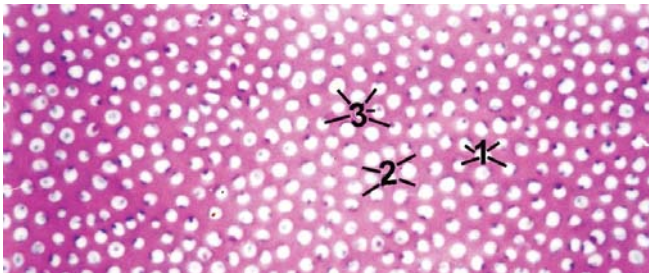


Figure 5.16: Cross section of dentinal tubules: The hollow dentinal tubules (1) show the presence of central odontoblastic processes (3) (violet dots). Inter tubular dentin (2) is smooth pink in color. Peritubular dentin is lost (H and E section of decalcified tooth x 150)

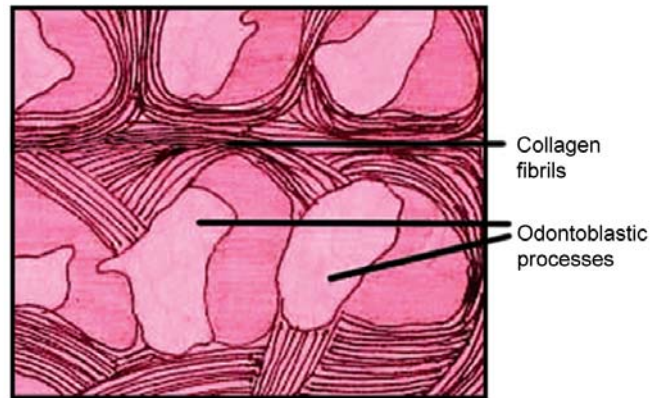


Figure 5.19: Surface of pre-dentin in circumpulpal dentin showing the collagen fibrils forming a network arranged perpendicular to the odontoblastic processes

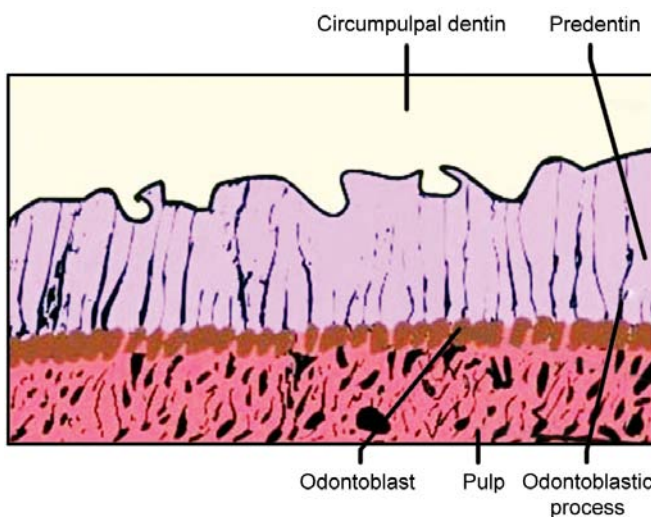


Figure 5.17: The pre-dentin present between the odontoblast and circumpulpal dentin. (Decalcified section of pulp and dentin)

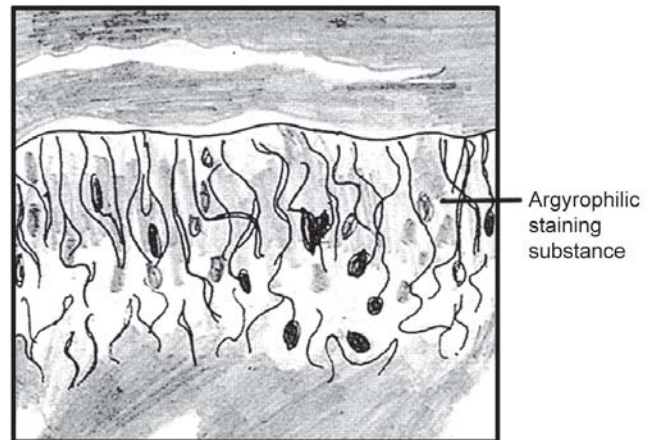


Figure 5.20: In pre-dentin argyrophilic nature of the ground substances among the odontoblasts appear like bundles of collagen fibers. The collagen formed by the odontoblasts is apical to the cell body (seen in silver stained section by light microscope)

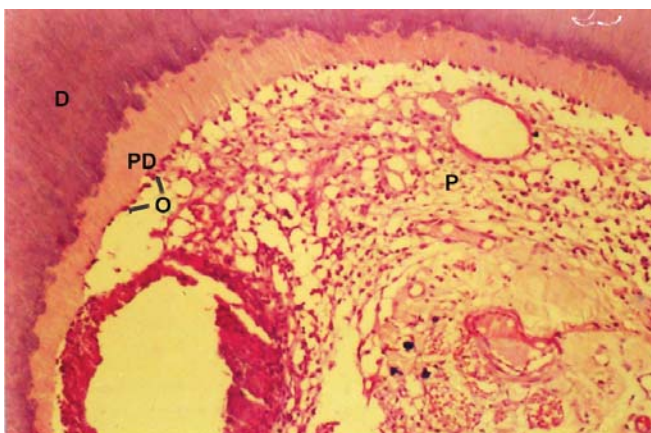


Figure 5.18: A layer of lightly stained pink unmineralized pre-dentin (PD) is seen adjacent to the pulp (P) Dentin (D) is mineralized and bluish. O= Odontoblasts. (Decalcified tooth H and E x 100)

DEVELOPMENT OF DENTIN

Dentin formation occurs by the cells, known as odontoblasts or dentinoblasts. These odontoblasts differentiate from the ectomesenchymal cells of the dental papilla after the organizing influence of the inner enamel epithelium. Thus, the dental papilla is the formative organ of the dentin.

Dentin Formation Pattern

Dentin formation begins at the late bell stage of development of tooth at the tip of the cusp. This occurs in the papillary tissue adjacent to the tip of the folded inner

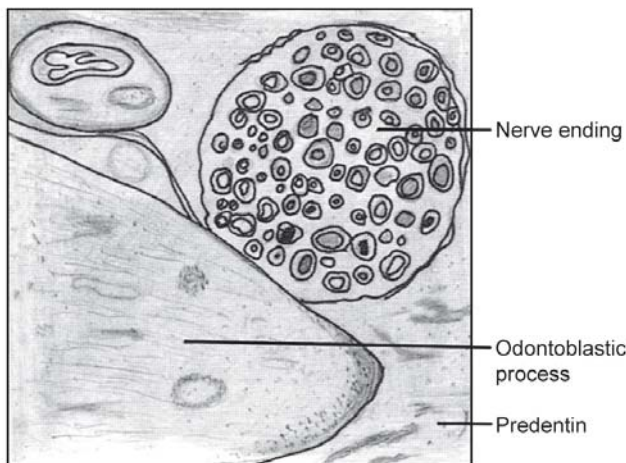


Figure 5.21: Vesiculated nerve ending in zone adjacent to odontoblastic process in predentin (as seen under electron microscope)

enamel epithelium. This site indicates where cuspal development will begin. From here the dentin formation spreads to cusp slope. Dentin formed constitutes the dentin of the crown of the tooth.

The root dentin forms later. This requires proliferation of epithelium cells from cervical loop of the enamel organ around the growing dental papilla to initiate the differentiation of the root odontoblasts.

Differentiation of Odontoblasts

Odontoblasts differentiate from undifferentiated ectomesenchymal cells. Laminin alpha 2 is important for odontoblastic differentiation and regulate the expressions of dentin matrix proteins. MAPIB gene is implicated for odontoblast differentiation. Before the beginning of dentinogenesis, the cells of the inner enamel epithelium are short columnar in shape. These divide rapidly and accommodate growth of the tooth germ. The cells of inner enamel epithelium are supported by basement membrane, which separates the epithelium from the dental papilla. At this time, the cells of the papilla are separated from inner enamel epithelium by an acellular zone (Fig. 5.22).

Cell division stops in the cells of the inner enamel epithelium. Their shape changes from short columnar to tall columnar. The cell nuclei migrate away from dental papilla. With these changes, changes also occur in adjacent dental papilla. The ectomesenchymal cell near the acellular zone enlarge to become first preodontoblasts

and then odontoblasts. These odontoblasts increase in volume and contain increasing amounts of rough endoplasmic reticulum and Golgi complexes.

The acellular zone between dental papilla and the inner enamel epithelium gradually obliterates. The odontoblasts differentiate and occupy this zone. Epithelial cells are required to initiate the differentiation of odontoblasts.

After the differentiation, three functional states of the odontoblasts can be seen. These are the secretory odontoblast, transitional odontoblast and the resting odontoblast.

Secretory odontoblast: Under light microscope, secretory odontoblast is seen as a large plump cell with an open-faced nucleus and basophilic cytoplasm. The nucleus is situated basally. Along the axis of cell are present the large cisternae of rough endoplasmic reticulum and the Golgi apparatus comprises several stacks of saccules. Secretory odontoblasts exhibit alkaline phosphatase activity along their plasma membrane.

Transitional odontoblast: Transitional odontoblast stage can be seen in electron microscope. The cell becomes narrower, its nucleus is displaced from its basal location. There is condensation of chromatin. The amount of endoplasmic reticulum reduces and is located around the nucleus. The autophagic vacuoles present help in reorganization of the cytoplasm of the odontoblast.

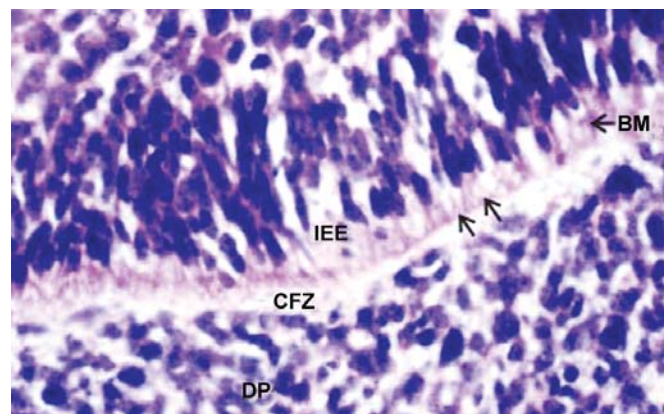


Figure 5.22: The cells of inner enamel epithelium (IEE) are supported by basement membrane (BM) indicated by arrows. There is a cell free zone (CFZ) between inner enamel epithelium and dental papilla (DP) from which odontoblasts will eventually differentiate (H and E x 400)

Resting odontoblast: Resting odontoblasts are small-flattened cells. The nucleus is closed. The cell is with less cytoplasm and no Golgi complex. The nucleus is situated apically and forms an infranuclear region. The supranuclear region is devoid of organelles. Secretory granules are absent. Resting odontoblasts are capable to changing into active secretory odontoblasts.

DENTINOGENESIS

Collagen Matrix Formation

After the differentiation of the odontoblasts, the collagen matrix is formed first and then it is mineralized along the pulpal border. Dentin is the first formed dental hard tissue. Ameloblasts within the enamel organ differentiate first and have an inductive effect on the odontoblasts that secrete collagen (Fig. 5.23).

During differentiation of the odontoblasts, their shape changes from ovoid to columnar and their nuclei become basal in orientation. The apical end of the cells gives rise to one or more processes in contact with basal lamina. The odontoblast gradually increases in length to 40 microns but remains constant in width (7 microns). The rough endoplasmic reticulum and the Golgi apparatus show the presence of proline which slowly migrates into

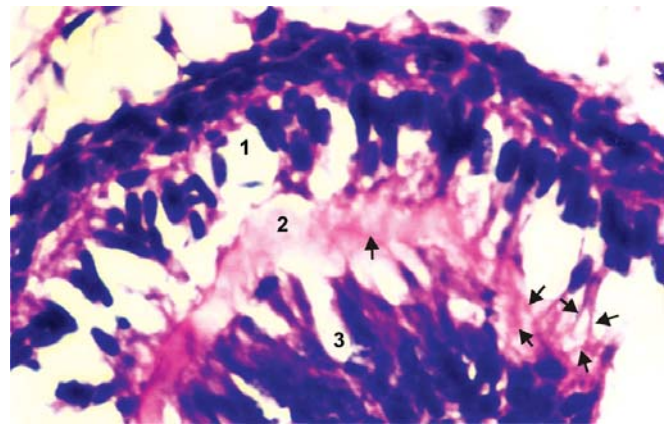


Figure 5.23: Earliest dentinogenesis or formation of dentin at the tip of incisor. The ameloblasts are tall, columnar (1) and nuclei are basal in location. The pre-dentin laid down is more fibrous in nature. The fine fibers in pre-dentin are called Korff's fibers indicated by arrows (2). The cell free zone has been obliterated. (3) Odontoblastic layer- crowded giving multilayered appearance. (H & E x 400)

the cell process in the form of dense granules. These granules are then emptied into the extracellular collagenous matrix of the pre-dentin. The odontoblast recedes leaving behind a single extension. The large number of initial processes join to form one process which becomes enclosed in a tubule (Figs 5.24 to 5.26).

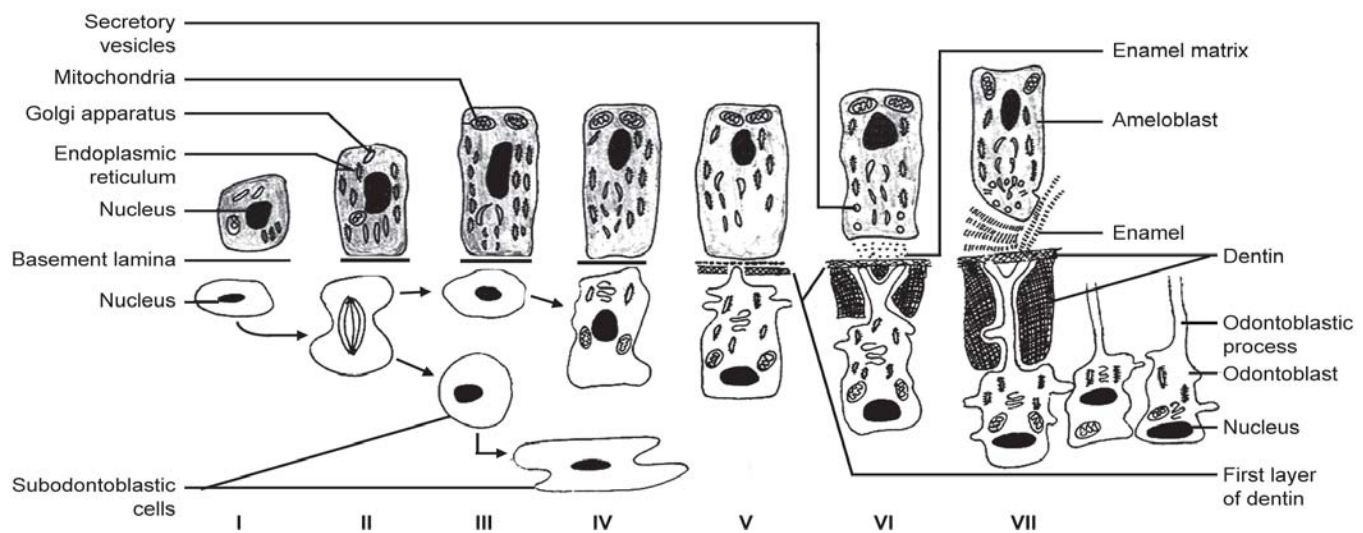


Figure 5.24: Life cycle of an odontoblast and its relation with ameloblasts (shaded). The odontoblasts are formed after ameloblasts (I to IV). The preodontoblast cells become columnar and large supranuclear Golgi material and rough endoplasmic reticulum develop in them (IV). The nucleus moves to the basal third of the cell and a series of processes develop (V). One process becomes more prominent and secretes matrix (VI). The cell body retreats centripetally leaving a long process within a dentinal tubule (VI). When the first layer of dentin is deposited, the ameloblasts get activated for further deposition of enamel (VII)

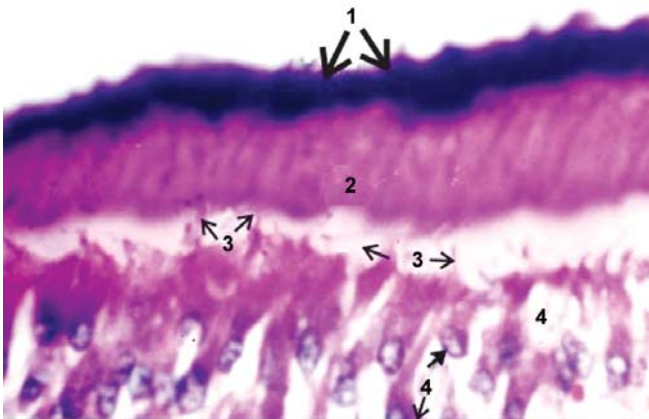


Figure 5.25: Dentin development: Thin odontoblastic processes (3) arising from the apical surface of odontoblasts (4) are seen at places entering the dentin matrix (2). These processes reside in the dentinal tubules. (1) Enamel (H & E x 400)

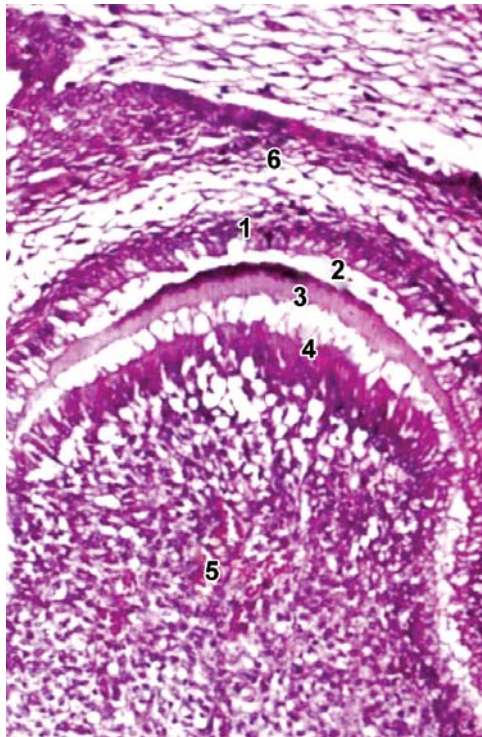


Figure 5.26: Tooth development: Dentinogenesis starts first at the center of the central incisors

1. Ameloblastic layer is tall columnar and highly secretory in nature.
2. Laid down enamel (Dark purple homogeneous thin layer below the letter '2').
3. Dentin is formed before enamel. Dentin layer is thicker and lighter in color and enamel is thinner and darker in color.
4. Odontoblastic layer is multilayered and show high secretory activity.
5. Dental papilla is highly cellular.
6. Collapsed stellate reticulum.

With the progression of matrix formation, the odontoblastic process increases in length along with the dentinal tubules. Till crown formation is completed and the teeth erupt and move into occlusion, about 4 micron thickness of dentin is formed daily. Afterward, it decreases to about one micron per day, slowing down further after completion of root development. But reparative dentin formation might continue whenever and wherever required at the rate of 4 microns per day for several months. The initial dentin formed along the cusp tips is called Korff's fibers (Figs 5.23 and 5.27). Von Korff's fibers are the collagen fibers of larger diameter and contain type III collagen.

All predentin is formed in the apical end of the cells and along the forming tubule walls. Both the collagen and the other components of the extracellular matrix are secreted by odontoblasts (Figs 5.28 A and B). Masson's Trichrome stain is used to demonstrate collagen matrix of the dentin. The collagen fibers of the dentin stain green



Figure 5.27: The silver-stained Korff's fibers of early forming dentin (as seen under light microscope)

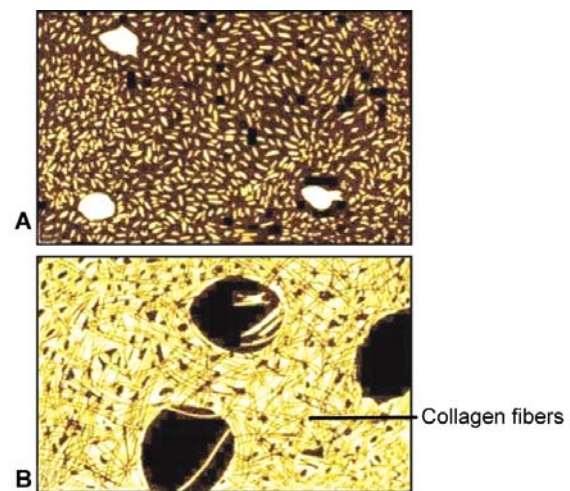


Figure 5.28: (A) Cross-section of undecalcified peripheral dentin, crisscross arrangement of collagen fibers; (B) Random arrangement of calcifying collagen fibers around the dentinal tubules

while the odontoblasts take red hue (Fig 5.29). Radicular dentin is less mineralized and slowly formed in comparison to coronal dentin.

Mineralization

Pattern of mineralization depends on the rate of dentin formation. Mineralization occurs by globular, or calcospheric calcification and it involves the deposition of the crystals in certain areas. Initially mineralization starts by deposition of very fine plates of hydroxyapatite on the surface of collagen fibrils and the ground substance (Fig 5.26). Later crystals are deposited within the fibrils themselves. The arrangement of these crystals is in an orderly manner, with their long axes paralleling the fibril long axes and in rows conforming to the 64 nanometers (nm) or 640 Å striation pattern. Crystal deposition starts radially from common centers in a so-called spherulite form. These are the first sites of calcification (Figs 5.30 and 5.31).

Important factors in mineralization are as follows:

1. Odontoblast secrete dentin phosphoprotein (DPP) which is important for mineralization and also control the growth of apatite crystals

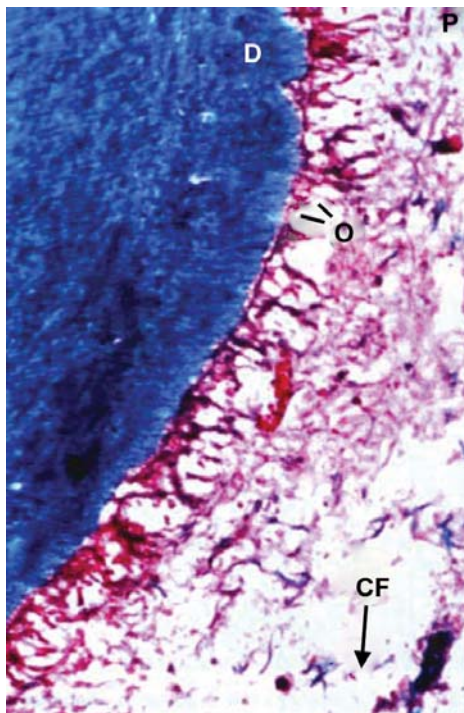


Figure 5.29: Masson's trichrome stain showing green coloration of collagen matrix of dentin (D). The odontoblasts (O) take reddish hue. Fine collagen fibrils (CF) present in the pulp (P) also stain green (x 100)

2. Osteonectin was secreted by odontoblasts, this protein promote the binding of apatite crystals to collagen but inhibit the growth of apatite crystals.
3. Osteopontin also promote the mineralization.
4. Gamma carboxyglutamic acid (Gla) protein attract the calcium.

PHEX gene is implicated for dentin mineralization. Dentinogenesis, first results in the production of an organic matrix. This organic matrix is calcified with apatite crystallites, through which run cytoplasmic extensions of the odontoblasts which occupy dentinal tubules. Dentin deposition at the coronal portion of the tooth occurs in an incremental manner.

The general calcification process is gradual but the peritubular dentin becomes highly mineralized at a very early stage. The ultimate crystal size formed is very small, about 3 nanometers (30 Å) in thickness and 100 nanometers (1000 Å) in length. The apatite crystals of dentin resemble those found in bone and cementum but three hundred times smaller than those found in enamel. The pulp predentin forming front may show globular dentin calcospherite mineralization (Figs. 5.32 and 5.33).

TYPES OF DENTIN

Primary Dentin

This type of dentin is formed prior to the complete eruption of the teeth and root completion. It forms the major bulk of the dentin. It is composed of mantle dentin and circumpulpal dentin. Primary dentin fulfills the initial formative function of the pulp.

Mantle Dentin

Mantle dentin is the first formed dentin in the crown close to the dentinoenamel junction. It is bounded by a zone of interglobular dentin and dentinoenamel junction. This zone has fibrils perpendicular to the dentino-enamel junction. The collagen fibrils are larger than those present in the circumpulpal dentin. It is thus the outer or most peripheral part of the primary dentin. It is less mineralized and is approximately 150 nanometer (15 microns) wide. The organic matrix of mantle dentin consists of ground substance that lacks loosely packed collagen fibrils.

Circumpulpal Dentin

The primary dentin that surrounds the pulp is called the circumpulpal dentin. It forms the bulk of the tooth. It contains slightly more mineral than the mantle dentin

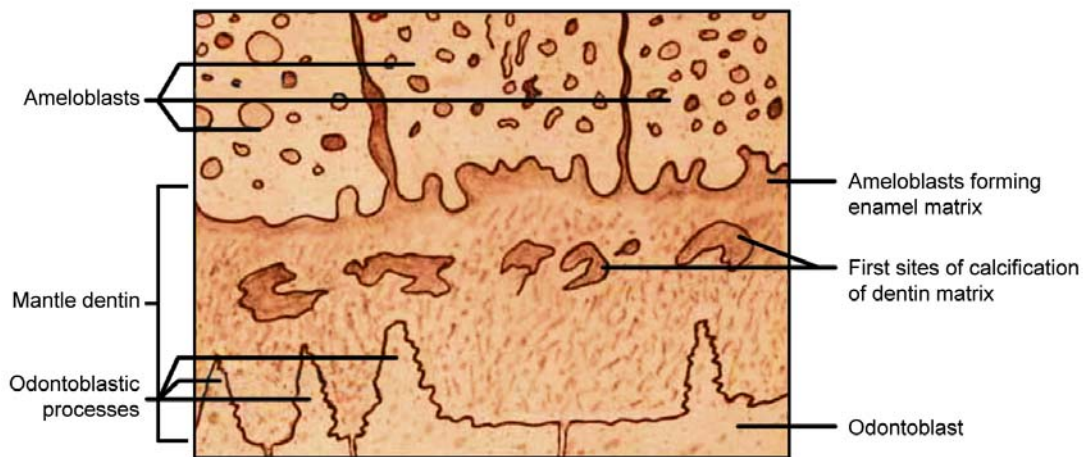


Figure 5.30: First formed enamel matrix at dentinoenamel junction, below it are first sites of calcification of dentin matrix. Predentin zone, with odontoblastic processes extending from the odontoblasts, is visible at the bottom of the field (as seen under electron microscope)

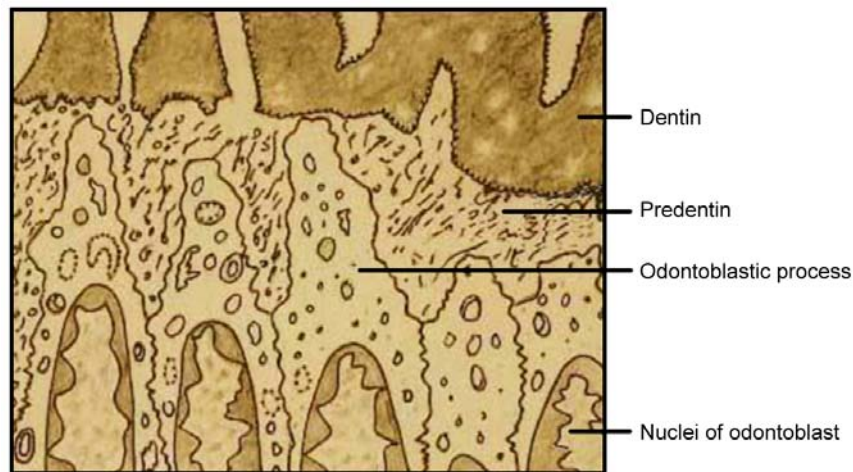


Figure 5.31: Predentin and dentin in a developing tooth is calcified and predentin below is composed of collagen fibers. Odontoblastic process can also be seen (as seen under electron microscope)

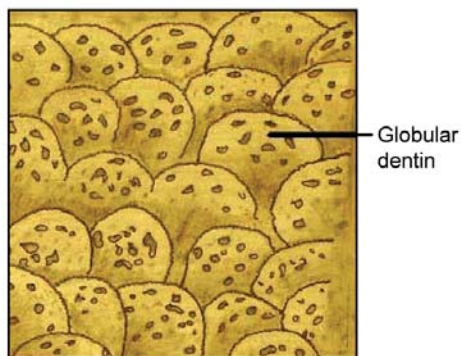


Figure 5.32: Globular dentin (calcospherite mineralization) formation at predentin (scanning electron micrograph)

(Fig. 5.34). It contains collagen fibers that are smaller in diameter (0.05 microns) and more closely packed compared to the mantle dentin. It represents all of the dentin formed before root completion.

Secondary Dentin

The secondary dentin is formed after root completion and complete eruption of teeth. Rarely secondary dentin is also seen in unerupted teeth. This is seen as a regular, narrow band of dentin around the pulp cavity. The secondary dentin contains fewer dentinal tubules than the primary dentin. A bend is seen in the dentinal tubules at the junction of the primary and secondary dentin (Figs 5.35 and 5.36).

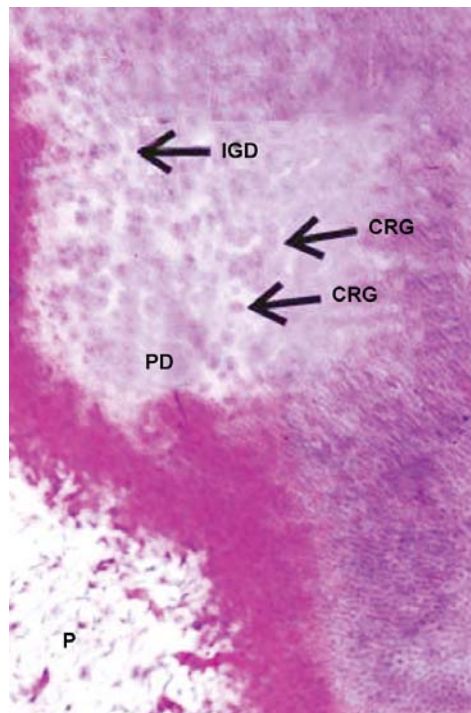


Figure 5.33: The predentin (PD) and dentin near the pulp (P) shows calcified round globules (CRG) or round globular calcospherites of calcium. Predentin and dentin showing calcified round globules (CRG) seen in mineral laying down in some cases. When those globules fail to fuse to form homogenous calcified dentin the intervening hypomineralized dentin is called Interglobular Dentin (IGD) (H & E x 100)

The secondary dentin grows slowly and reduces the size of the pulp with advancing age. Its formation is not uniform and occurs in a greater amount on the wall and pulpal floor. This deposition of secondary dentin protects the pulp. Secondary dentin shows an incremental pattern and a tubular structure. This tubular structure is continuous with the primary dentin.

Secondary dentin is deposited at the periphery of pulp. There is greater deposition of the secondary dentin on the roof and the floor of the pulp chamber. Because of this there is an asymmetrical reduction in size and shape of the pulp chamber and the pulp horn. This is clinically important. The changes in the pulp space, which is known as the pulp recession can be detected radiographically. This is helpful in determining the form of cavity preparation in certain restorative procedures. Secondary dentin scleroses more readily than the primary dentin. This reduces the overall permeability of the dentin and protects the pulp.

Reactionary or Regenerated Dentin

If odontoblasts are survived after exposure to the operative procedures, abrasion, erosion or caries and produce dentin then this dentin is known as reactionary or regenerated dentin.

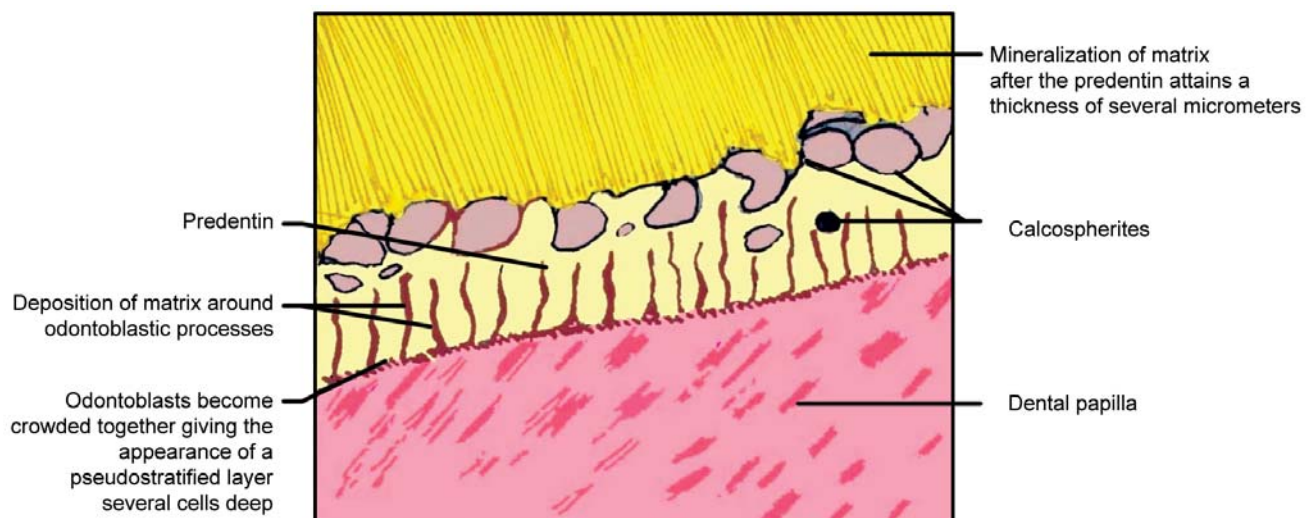


Figure 5.34: Formation of circumpulpal dentin through deposition and subsequent mineralization of dentin matrix (Predentin)

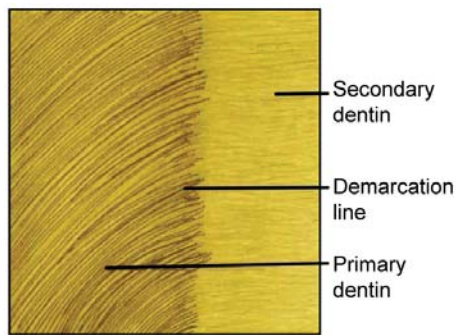


Figure 5.35: Dentinal tubules show sharp bend when they enter from primary dentin into secondary dentin (As seen in ground section).

Reparative / Tertiary / Reactive / Response Dentin

The dentin formed in response to caries, abrasion, erosion, attrition, or operative procedures in which most of the odontoblasts are cut or injured or die is known as reparative, tertiary, reactive or response dentin (Figs 5.37A and B and 5.38). Either of these processes causes the odontoblasts to be cut or injured. This results in the degeneration of a large number of odontoblasts. A few, however, survive and keep forming reactionary dentin. Those that are killed are replaced by the movement of undifferentiated cells from the cell rich zone or from the

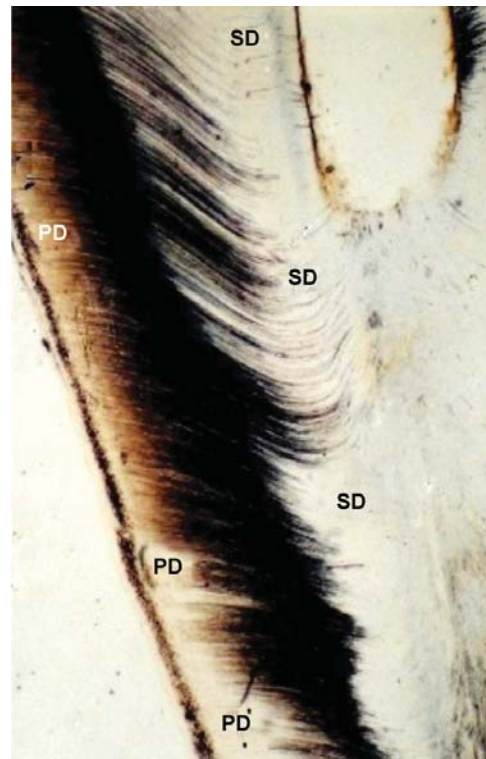


Figure 5.36: Dentinal tubules showing bending as they enter from primary dentin (PD) into secondary dentin (SD). Ground section x 100.

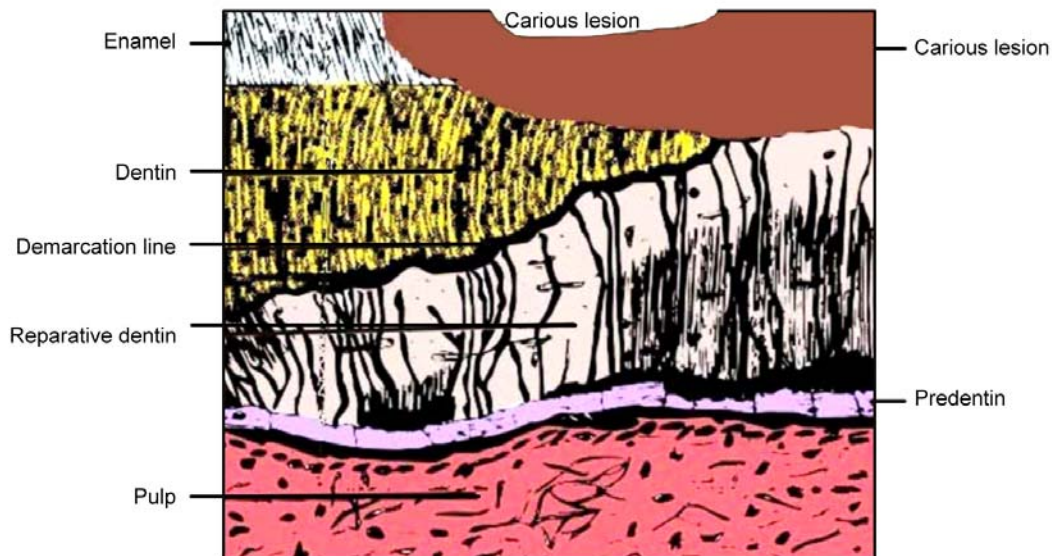


Figure 5.37: Reparative dentin is stimulated by penetration of toxic products as well as chemical substances of carious lesion into dentin. Dentinal tubules are more twisted and less in number than normal dentin (as seen in decalcified-section).

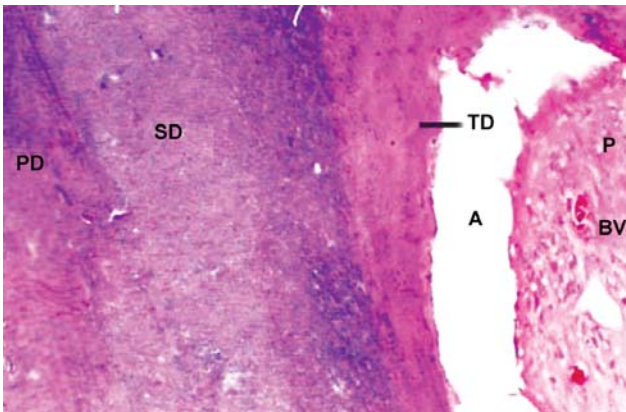


Figure 5.38: Reparative or Tertiary Dentin: Thick layer of Tertiary Dentin (TD) is seen bordering the pulp (P), PD= Primary dentin, Secondary dentin (SD), Blood vessel (BV), Artifact (A) (H&E x 100)

undifferentiated perivascular cells present deeper inside the pulp. The newly differentiated odontoblasts deposit the reparative dentin. (Figs 5.39 to 5.41) This process is started by the pulp and it helps seal off the area of injury causing resolution of inflammation and removal of dead cells. Reparative dentin has lesser and more twisted tubules than normal dentin. Dentin forming cells are often included in the rapidly produced intercellular substance often called osteodentin. In some cases, a combination of osteodentin and tubular dentin is also seen (Fig. 5.42).

The quality and the quantity or the degree of tertiary dentin produced is related to the intensity and the duration of the stimulus. In most of the cases, there is no continuity between dentinal tubules of tertiary dentin and

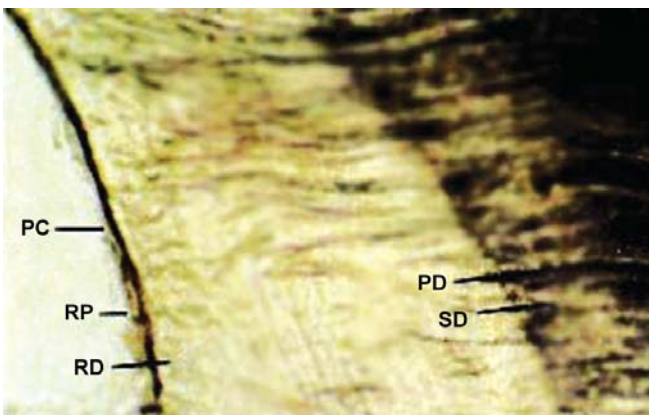


Figure 5.39: Tertiary/Reparative Dentin: Ground Section of Tooth: Thick layer of reparative dentin (RD) shows very few haphazardly arranged twisted tubules and abundant matrix. Secondary (SD) and primary dentin (PD) can be seen at the peripheral aspect x 150. PC= Pulp cavity lined by, RP= Remnant of pulp

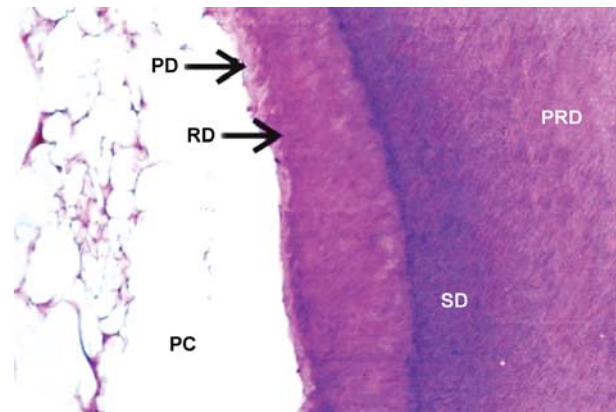


Figure 5.40: Reparative or Tertiary Dentin (RD) is freshly laid down, SD= Secondary dentin , PRD = Primary dentin. A thin layer of PD= unmineralized predentin can also be seen, PC = Pulp cavity (H & E x 100).

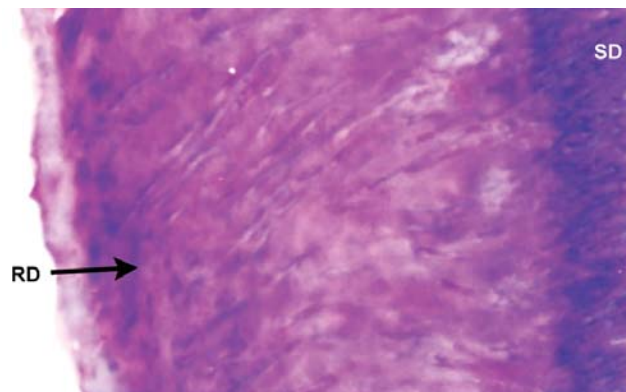


Figure 5.41: Tertiary or reparative dentin showing sparse dentinal tubules, only some of the tubules are continuous with the tubules of secondary dentin (SD). (H & E stained section of decalcified tooth)

overlying primary or secondary dentin. This minimizes dentin permeability at the site of deposition thus giving protection to the underlying dental pulp. Dentin phosphophoryn is not present in tertiary dentin. Tertiary dentin is further subclassified as (a) reactionary dentin which is deposited by preexisting odontoblasts which have surrounded the injury (b) reparative dentin which is deposited by newly differentiated odontoblasts like cells.

Sclerotic or Transparent Dentin

The presence of caries, attrition, abrasion, erosion or external stimuli may cause increase in dentin deposition as well as an increase in the mineralization of old existing dentin.

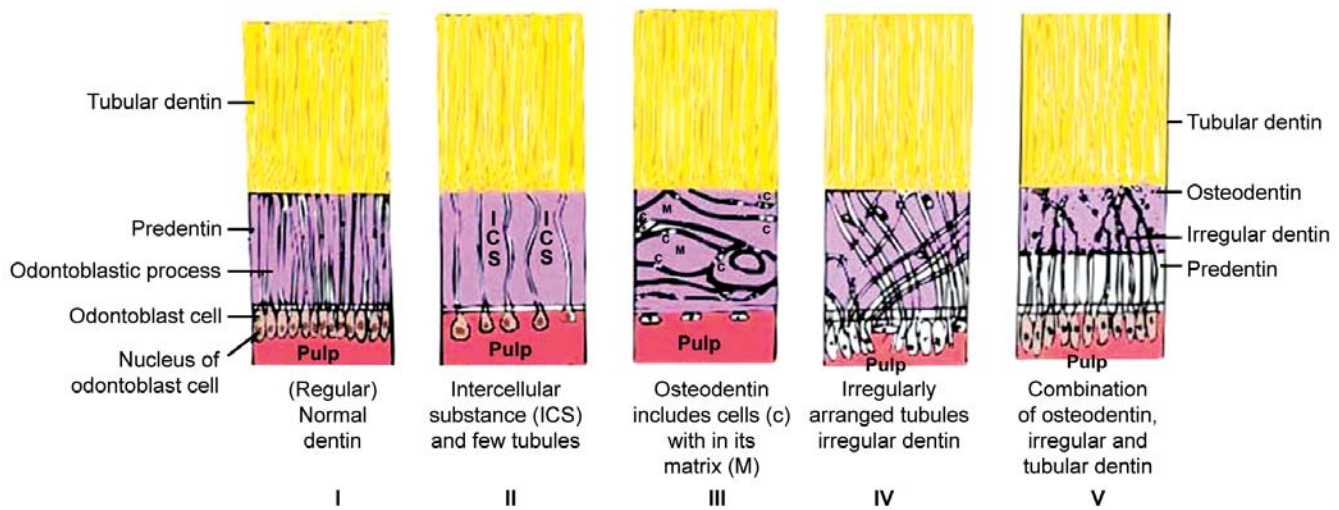


Figure 5.42: Normal (Regular) dentin (I) and different types of reparative Dentin (II to V), (II) proper reparative dentin. Dentin (II), (III) osteodentin, (IV) irregular and (V) their combination of osteodentin, irregular and tubular dentin

The excessive formation of collagen fibers and apatite crystals in the dentinal tubules can lead to complete obliteration of dentinal tubules that may be considered a defensive reaction of the dentin. Initially the apatite crystals are only sporadic but gradually a fine meshwork of crystals is formed. The refractive indices of dentin in which the tubules are occluded are equalized, and such areas become translucent or transparent (Figs 5.43 to 5.49). Gradually, the tubule lumen is obliterated with mineral, which appears quite like the peritubular dentin. The amount of sclerotic dentin increases with age. It can also be present under slowly progressing caries. It is mostly present in the apical region of root below the attrited cusps in the crown and abrasion in the midway of crown. Sclerosis may help to prolong pulp vitality and reduce

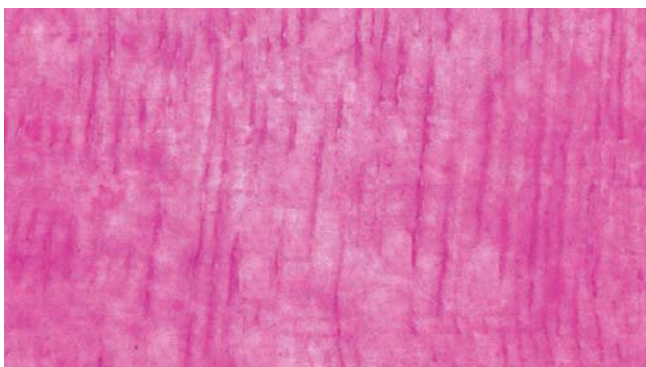


Figure 5.43: Section of Sclerotic Dentin- Broken sclerosed dentinal tubules are visible x 40

dentin permeability. It appears dark in reflected light and transparent in transmitted light. Rarely sclerosed dentin is discolored when it very near to the carious lesion (Fig. 5.46).

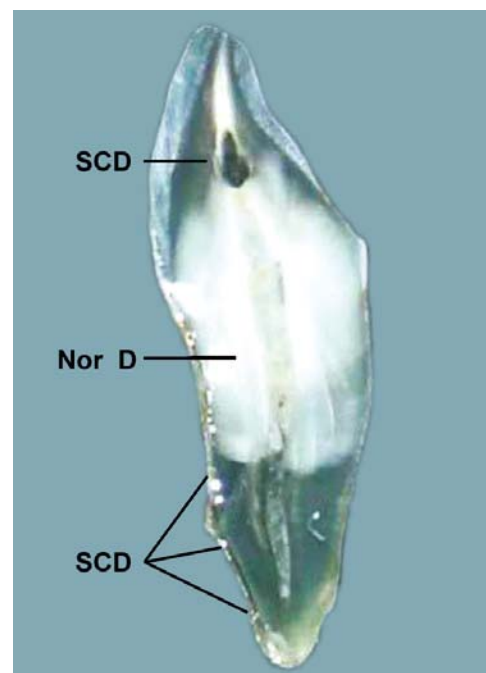


Figure 5.44: Sclerotic Dentin: The dentin in the incisal half of crown and apical half of the root is transparent showing Sclerotic dentin (SCD) where dentinal tubules are fully calcified. Normal dentin = Nor D

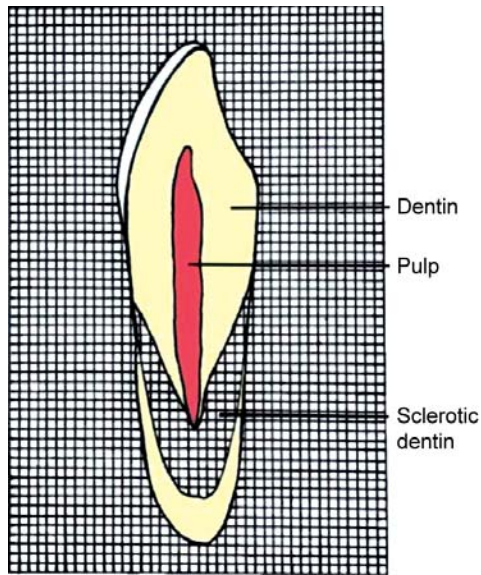


Figure 5.45: Ground section of a tooth. The pattern on which the section is placed can be seen through the translucent sclerotic dentin

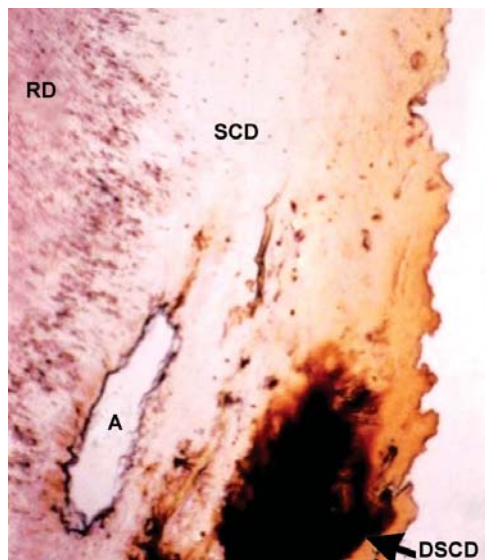


Figure 5.46: The dentin shows rough ragged margin due to caries. The underlying dentin is homogenous, devoid of dentinal tubules due to sclerosis of dentin. The dentinal tubules have been completely obliterated by collagen and mineral. SCD= Sclerotic dentin, RD= Reparative Dentin. (Ground section x 20). DSCD= Discolored sclerotic dentin due to caries) A= Artifact

Sclerotic dentin is generally observed in the teeth of elderly people. Mineral density is greater in this area. Fracture toughness of sclerotic dentin was less than normal dentin but crystals are smaller and more harder in comparison to normal dentin.

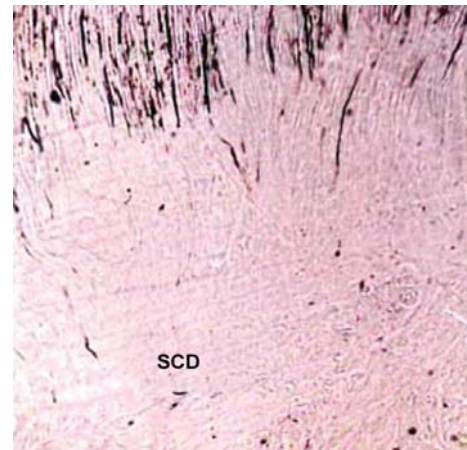


Figure 5.47: Sclerotic dentin (SCD). Under higher magnification the dentinal tubules are not visible as they are obliterated x 40

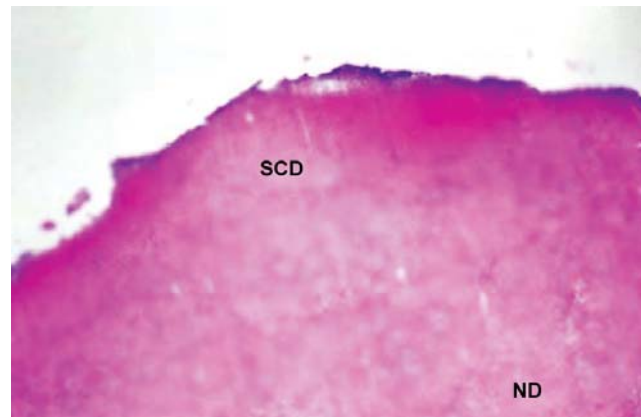


Figure 5.48: Decalcified section of caries dentin. The margin is irregular and frayed. Under the cavity it is light to deep pink area which shows absence of dentinal tubules due to Sclerosis SCD= Sclerotic Dentin, ND= Normal Dentin (H & E x 40)

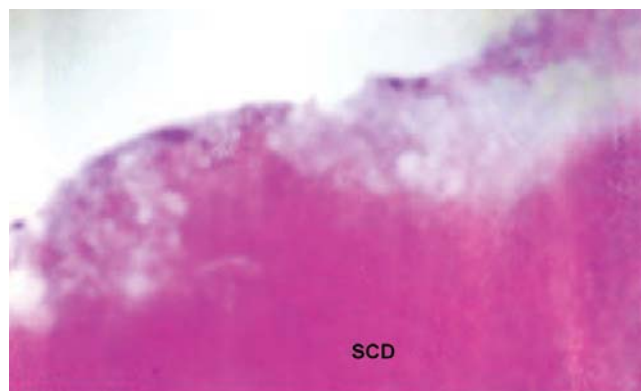


Figure 5.49: Higher magnification of Figure 5.48 shows pink homogenous matrix of sclerosed dentin (SCD). Dentinal tubules are absent. (H & E x 200)

Interglobular Dentin /Spaces

Interglobular dentin are the areas of unmineralized or hypomineralized dentin which persist within the normally mineralized dentin. Mineralization of dentin takes place by precipitation of inorganic calcospherite granules, which enlarge and coalesce to form a homogenous layer of calcified dentin. Sometimes mineralization of dentin begins in small globular areas that fail to fuse into a homogenous mass. This results in formation of irregular areas of hypocalcified matrix, called inter globular dentin (Figs 5.50 to 5.54). They are seen along the incremental lines, especially in the crown of teeth in the circumpulpal dentin just below the mantle dentin. The uninterrupted passage of dentinal tubules through the interglobular dentin indicates a defect in mineralization and not in matrix formation (Figs 5.51, 5.52 and 5.54). In dry ground sections, a small amount of interglobular dentin may be lost and a space results which appears black in transmitted light hence it is also called interglobular spaces (Figs 5.52 and 5.53).

It is frequently seen in teeth in which there has been a deficiency of vitamin D or exposure to high level of fluoride during dentinogenesis.

STRUCTURAL LINES

Two types of structural lines can be found in dentin.

- The structural lines related to the formation of the dentin are called incremental lines or imbrication lines.
- The other types of lines are related to the curvature of the dentinal tubules.

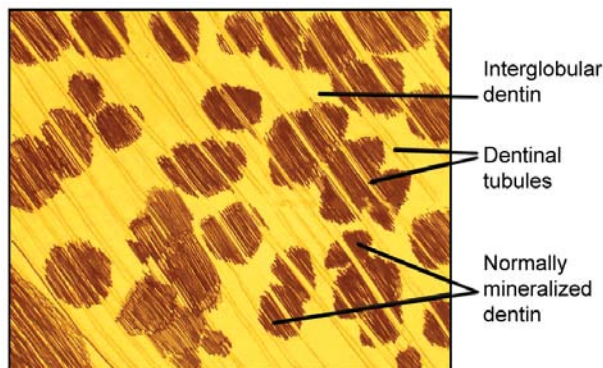


Figure 5.50: Hypomineralized or unmineralized interglobular dentin within normally mineralized dentin. The dentinal tubules pass uninterruptedly through interglobular dentin (as seen in decalcified section)

Incremental Lines of von Ebner (Contour Lines of Owen)

Dentin formation is a rhythmic process. There are alternate phases of activity and inactivity. These phases are represented in fully formed dentin as incremental lines. Thus the incremental lines of von Ebner or imbrication lines reflect the daily rhythmic pattern of dentin matrix deposition in the formative process. They are seen as fine lines at right angles to the dentinal tubules and are called imbrication lines or incremental lines of von Ebner

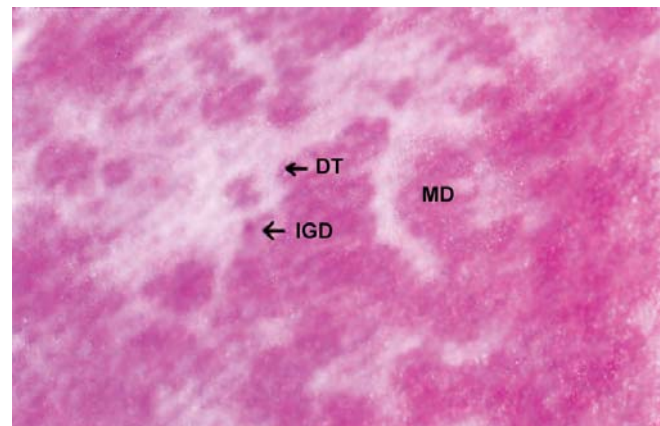


Figure 5.51: Interglobular Dentin: Dentinal tubules (DT) pass through lighter hypomineralized areas [Interglobular dentin (IGD)] uninterruptedly indicating that there is no defect in tubule formation in areas of interglobular dentin. MD= Mineralized Dentin (globules). (H & E x 400)

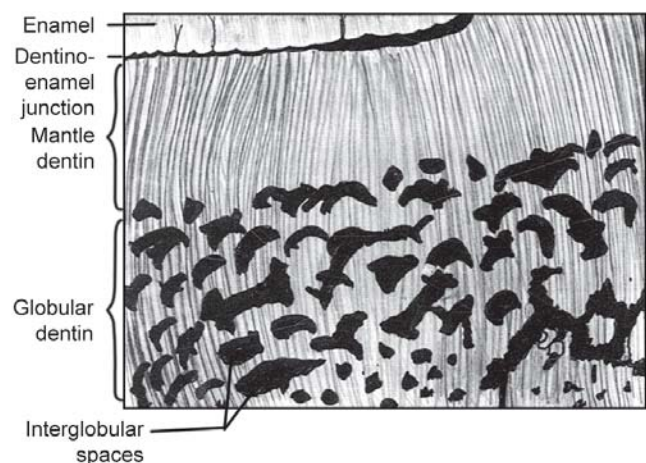


Figure 5.52: Interglobular dentin appears at a short distance from the dentinoenamel junction. In dry ground section interglobular spaces appear black in transmitted light

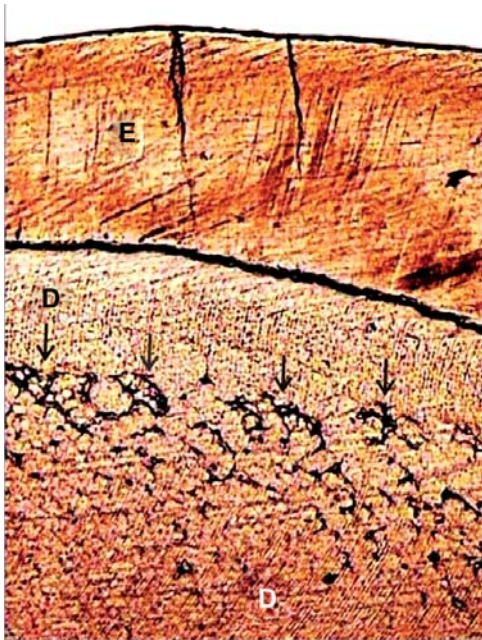


Figure 5.53: Interglobular dentin: Ground section of tooth showing irregular dark areas in dentin (D). These areas indicated by arrows represent interglobular dentin. (E). Enamel x 40

(Figs 5.55 and 5.58). The distance between these lines varies between 4 and 8 microns, which is the amount of dentin formed in 24 hours. When the tooth comes into functional occlusion, the daily increment decreases. The course of these lines shows the growth pattern of dentin. Some incremental lines are accentuated due to disturbances of mineralization and are called as Contour lines of Owen. These lines indicate a phase of mineralization and follow the outer contour of dentin. These lines represent hypocalcified bands. Mineralizing lines represent variations in mineralization. These are seen in ground section by microradiography or in demineralized section. They lie at an angle to the von-Ebner lines.

Neonatal Lines

Neonatal lines are hypocalcified area represented by a wide contour line seen in those teeth, where dentin is partly formed before birth and partly formed after birth. These are found in all deciduous teeth and first permanent molars. The neonatal lines represent the disturbance in mineralization due to the abrupt change in environment that occurs at birth. The dentin matrix formed prior to birth

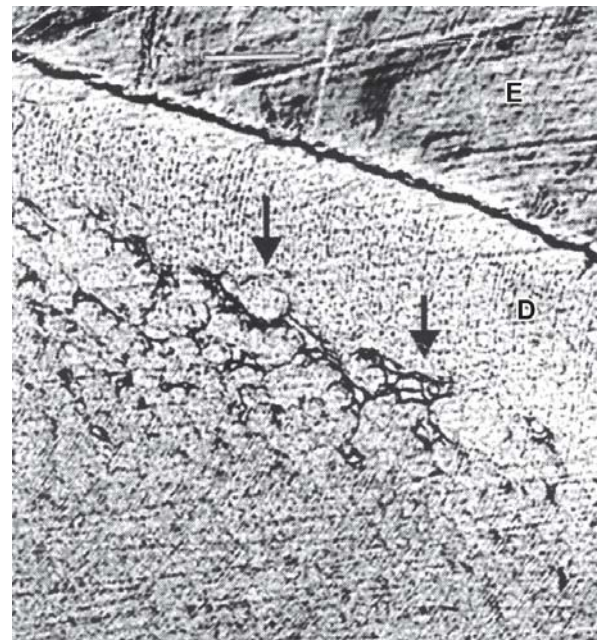


Figure 5.54: Interglobular dentin: Irregular shaped areas (shown by arrow) of hypomineralized dentin (D) enamel (E). Ground section x100

is usually of better quality than that formed after birth. The neonatal line is present in both enamel and dentin (Fig. 5.59).

GRANULAR LAYER OF TOMES'

This is a granular layer seen under transmitted light adjacent to cementum in ground sections of root dentin (Figs 5.60 and 5.62). This is known as Tomes' granular layer. This zone increases in amount from the cemento-enamel junction to the apex of the root. It appears dark in transmitted light and lighter under reflected light. The granular layer represents the looped terminal portion of the dentinal tubules in the root dentin. The cause of development of this zone is possibly similar to the branching and beveling of the dentinal tubules at the dentino-enamel junction. In the crown there is extensive branching of the odontoblast processes, and in the root, there is branching of adjacent processes. Tomes' granular layer and interglobular dentin are hypomineralized areas but the concentrations of calcium and phosphorus are higher in Tomes' granular layer and concentration of sulfur is higher in interglobular dentin.

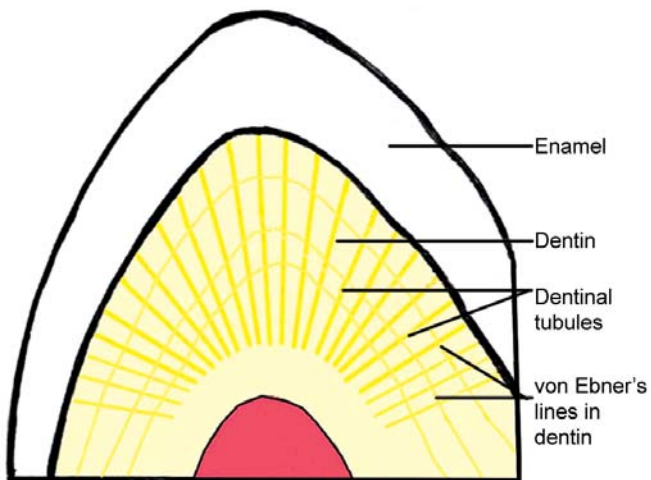


Figure 5.55: Incremental lines of von Ebner in the dentin run at right angle to the dentinal tubules (as seen in ground section)

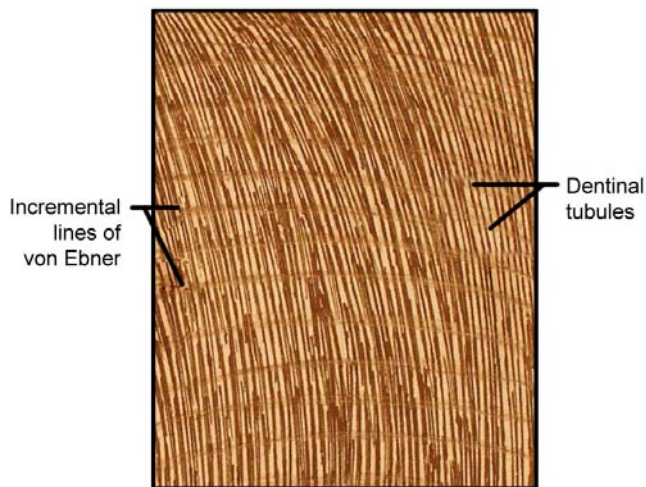


Figure 5.56: Incremental lines of von Ebner in ground section of dentin

DENTINAL JUNCTIONS

Dentinoenamel Junction

The junction between the enamel and dentin is irregular and is described as scalloped (Figs 5.63 and 5.64). The convexities face the dentin whereas the concavities face the enamel.

A membrane performativa (the basal lamina) is seen between the enamel and dentin during tooth development. After calcification starts, this membrane disappears and the interface is called dentinoenamel

junction. Under scanning electron microscope, the junction represent a series of ridges, which increases adherence between dentin and enamel. This ridging is more developed in coronal dentin where occlusal stresses are greatest.



Figure 5.57: Incremental lines of von Ebner (1) / Imbrication Lines: Ground section of deciduous incisor tooth: Curved lines perpendicular to the dentinal tubules (2) can be seen. The crest of the line corresponds to the edge of the incisor (x 500)

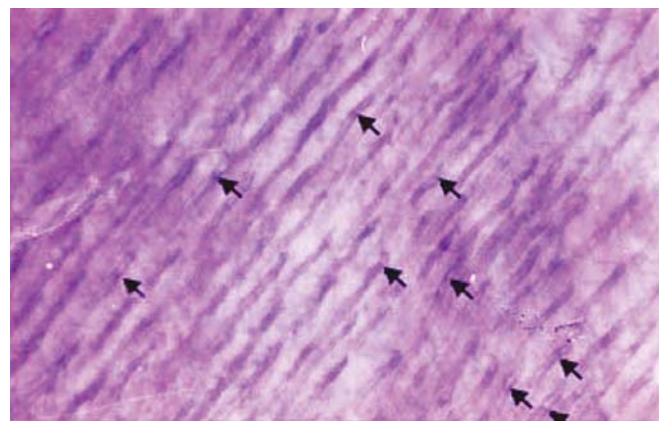


Figure 5.58: Dentin: High magnification of dentinal tubules showing small transverse lines at a distance of four to six microns (indicated by arrows) representing incremental lines of von Ebner

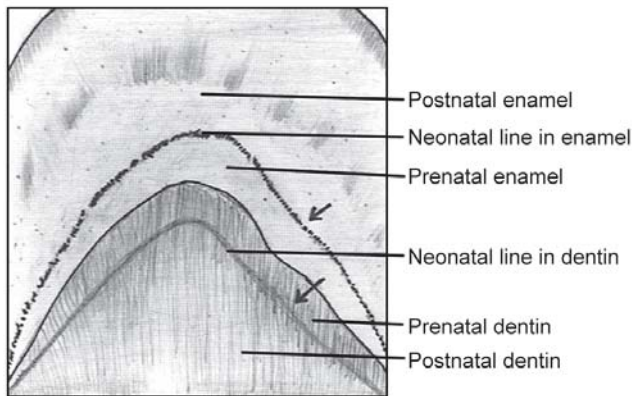


Figure 5.59: An accentuated incremental line separates prenatal and postnatal dentin. Neonatal lines in enamel and dentin (arrows) appear clear, in deciduous first molar (as seen in ground longitudinal section).

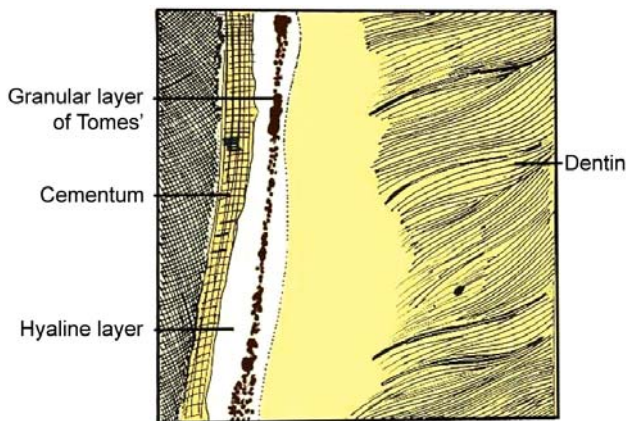


Figure 5.60: Root dentin showing the granular layer of Tomes' and hyaline layer (as seen in ground section)

Predentin Pulp Junction

Predentin-pulp junction is made up of dense collagenous fibers and is present between the uncalcified dentin (predentin) and pulp.

Dentin Predentin Junction

Dentin predentin junction is the interface between the calcified and uncalcified newly formed dentin called predentin.

Denting Cemental Junction

There is a smooth line junction between the dentin and cementum in permanent teeth. The cementodentinal junction in deciduous teeth, however, is sometimes slightly scalloped. The attachment of cementum to dentin is quite firm.

INNERVATION OF DENTIN

Dentin is a highly sensitive tissue because the dentinal tubules contain numerous nerve endings in the predentin and inner surface of dentin. Sensitivity is increased when the pulp is inflamed. The nerves and their terminals are found in close association with the odontoblast process within the tubules. The nerve endings are packed with

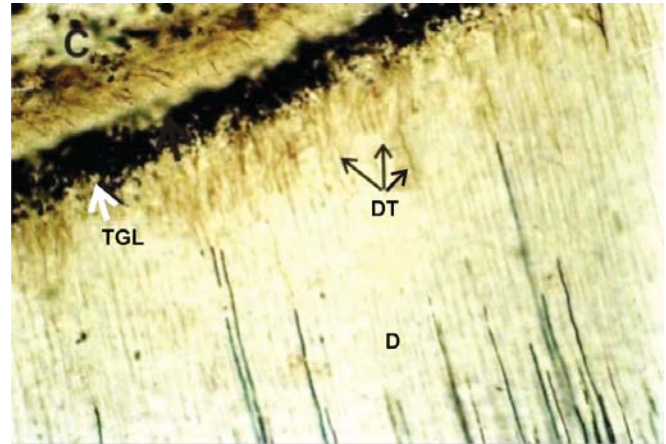


Figure 5.61: Tomes' granular layer (TGL) at cementodentinal junction seen as cementum black granular layer. (D= Dentin) The dentinal tubules (DT) near this layer show lateral branching. (C) (Ground section x 200).

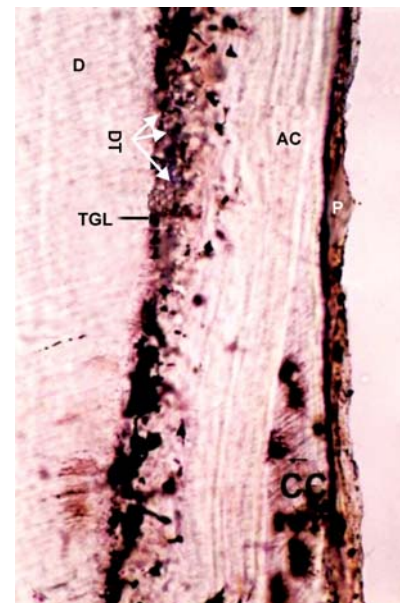


Figure 5.62: The empty ends of dentinal tubules (DT) can be seen in Tomes' granular layer (TGL) very clearly in high magnification. D = Dentin, AC = Acellular cementum, CC = Cellular cementum, P = Remnant of periodontal ligament adherent to cementum. (Ground section x 200)

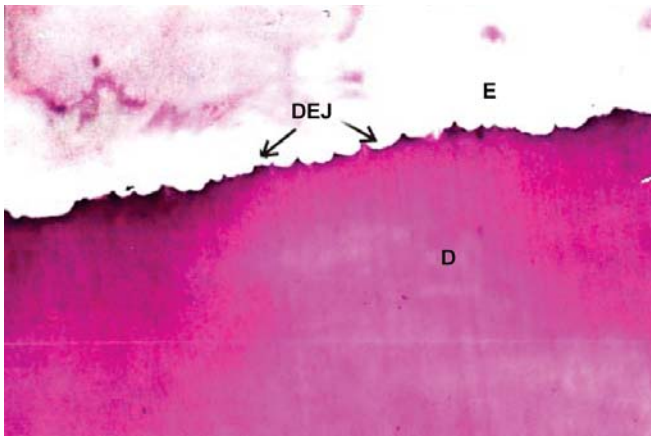


Figure 5.63: Dentinoenamel Junction: (DEJ) is scalloped and shows concavity towards enamel (E) dissolved during decalcification and convexity towards dentin (D). (H & E x 100)

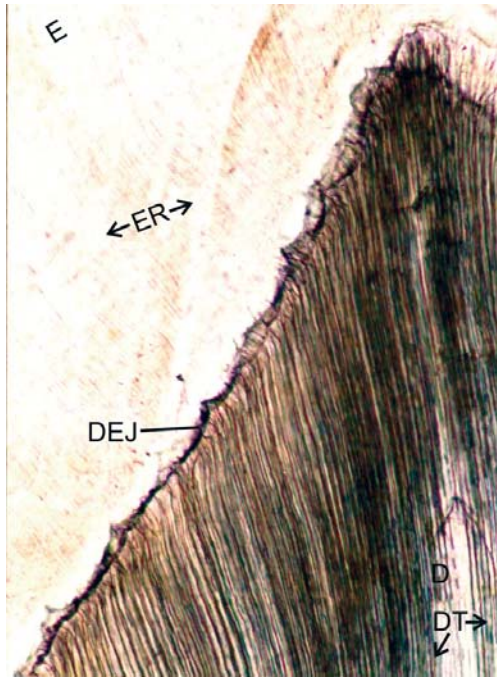


Figure 5.64: Scalloped dentinoenamel junction (DEJ) in ground section, with convexity towards dentin (D) and concavity towards enamel (E). Enamel projections fit snugly into shallow depressions of dentin

small vesicles. It is believed that most of these are terminal processes of the myelinated nerve fibers of the dental pulp. The primary afferent somatosensory nerves of the dentin and pulp project to the main sensory nucleus (subnucleus condalis) of the mid brain.

There are three theories explaining dentin sensitivity.

Direct Neural Stimulation

This theory stated that the nerve endings in the dentin, when stimulated, evoke a pain response. However, no nerve fibers could be demonstrated going to dentino-enamel junction, which is the most sensitive area. Thus dentin sensitivity does not solely depend on the stimulation of such nerve endings. There is little scientific support of this theory.

Odontoblast Receptor Theory or Transduction Theory

Odontoblasts are derived from the neural crest cells. The odontoblasts retain an ability to transmit and propagate an impulse. This theory states that the odontoblastic process is the primary structure excited by the stimulus. This is not a popular theory since there are no neurotransmitter vesicles present in the gap junctions between odontoblasts and between odontoblasts and pulpal nerve to facilitate the synapse or synaptic transmission.

Hydrodynamic Theory

The dentinal tubules contain fluid called dental lymph. Various stimuli affect fluid movement in the dentinal tubules and stimulate the pain mechanism by mechanical disturbance of the nerve closely associated with the odontoblast process (Fig. 5.65). The fluid movement is sensed by the free nerve endings in the plexus of nerves called subodontic plexus of Raschkow situated in cell-free zone of Weil. When dentin is exposed for the first time very small blebs of fluid are visible on the pulpal wall of the cavity or the wall of fresh cut dentin which is nearest to the pulp. If cavity is dried with air more fluid is lost resulting in more fluid movement leading to more pain. Due to profuse branching of tubules at the dentinoenamel junction the sensitivity is more. The stimuli may be heat, cold, desiccation, mechanical, chemical or osmotic pressure. Thus, these nerve endings may act as mechanoreceptors as they are affected by mechanical displacement of the dentinal fluid. This is most accepted theory of pain transmission through dentin. Hydrodynamic theory also explains the following. (a) Pain is produced by thermal change. (b) Local anesthetic is ineffective on application to cut dentin. (c) Pain is produced by dehydration, mechanical probing and application of hypertonic solutions.

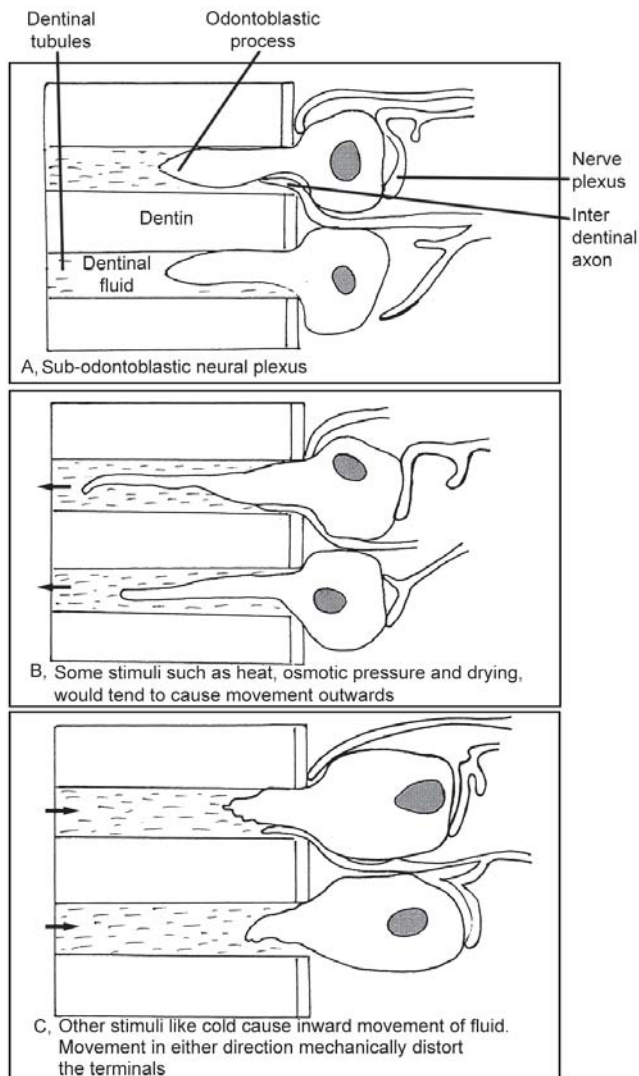


Figure 5.65 Sensory mechanism in Dentin

Age and Functional Changes

With advancing age and for functional requirements number of changes are seen in dentin. Important among them are the following:

1. Formation of secondary dentin
2. Sclerotic dentin
3. Formation of reparative dentin (Tertiary dentin)

Above three are already described in this chapter.

4. **Dead tracts:** Ground section of dentin under transmitted light sometimes shows a group of dark lines that follow the course of dentinal tubules. These are called dead tracts. They are groups of dead degenerated odontoblastic processes in the dentinal tubules

(Fig. 5.66). These tubules are empty and are filled with air and therefore appear dark in transmitted light (Figs 5.67 to 5.69). Dead tracts are probably the initial step in the formation of sclerotic dentin. These areas demonstrate decreased sensitivity and appear to a greater extent in older teeth due to the aging process. Their degeneration is most commonly observed in the area of narrow pulpal horns because of crowding of odontoblasts.

In case of teeth with vital pulp there may be loss of the odontoblast processes. This may be due to caries, attrition, cavity preparation or erosion and abrasion

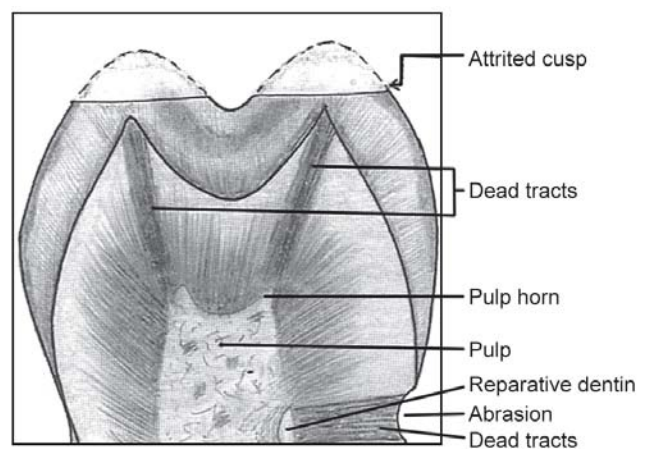


Figure 5.66: Degeneration of odontoblastic processes due to abrasion and attrition causes dead tracts in the dentin of a tooth with vital pulp (as seen in ground section)

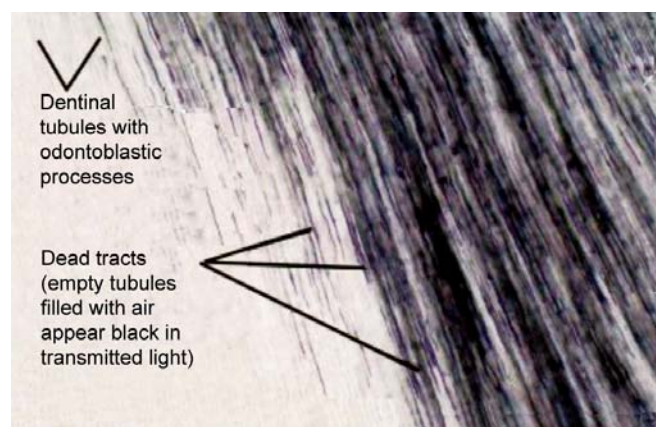


Figure 5.67: Longitudinal ground section of tooth showing dentinal tubules and dead tracts. Dentinal Tubules can be seen running longitudinally. The dead tracts appear black because of air replacing the odontoblastic process (x 150).

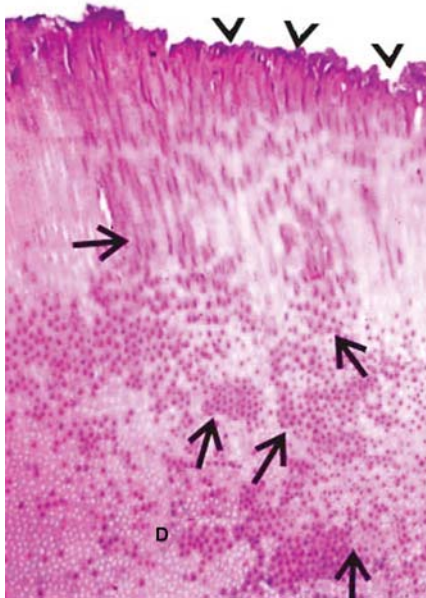


Figure. 5.68: Decalcified section of tooth showing dead tracts indicated by arrows in dentin (D) due to degeneration of the odontoblastic process as a result of caries. The margin of dentin at the floor of cavity indicated by arrow heads consists of trayed dentinal tubules open to environment (H & E x 100)

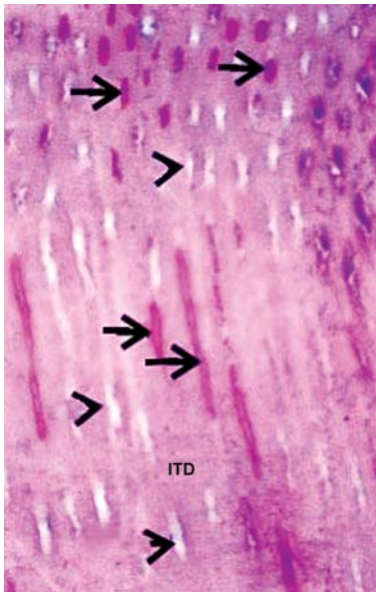


Figure 5.69: Higher magnification of figure 5.68. The degenerated dentinal tubules (dark colored indicated by arrows) show increased amount of collagen at places obliterating the tubule. There is widening of the intertubular dentin (ITD). These changes ultimately lead to the formation of sclerosed dentin. Normal dentinal tubules, white in color indicated by arrowheads are seen as empty tubules. (H & E x 400)

At some places reparative dentin seals dentinal tubules at the pulpal ends. These dentinal tubules get filled with fluid or gaseous substance. These tubules may entrap air and appear black in transmitted and white in reflected light.

These dentin areas where degenerated odontoblastic processes are present give rise to dead tracts.

CLINICAL CONSIDERATIONS

Unlike enamel, dentin is a living tissue. Trauma during operative procedure may cause pathology or death of pulp tissue. About thirty thousands to fifty thousands living cells are damaged when one square millimeter of dentine is exposed. The rapid penetration and the spread of caries in the dentin is the result of the tubule system in the dentin. The dentinal tubules form a passage for invading bacteria that may thus reach the pulp through a dentinal layer. The early decalcification of dentin involves the walls of the tubules allowing them to distend slightly as they become packed with masses of microorganisms (Figs 5.70 to 5.72). One tubule may be filled with coccal forms, while the adjacent tubules may contain bacilli or thread forms (Fig. 5.72).

During advanced stages of caries, walls of the individual tubules undergo further decalcification leading to their confluence. Along with this there is an increase in the diameter of the dentinal tubules. This occurs due to packing of the tubules with microorganisms. Small

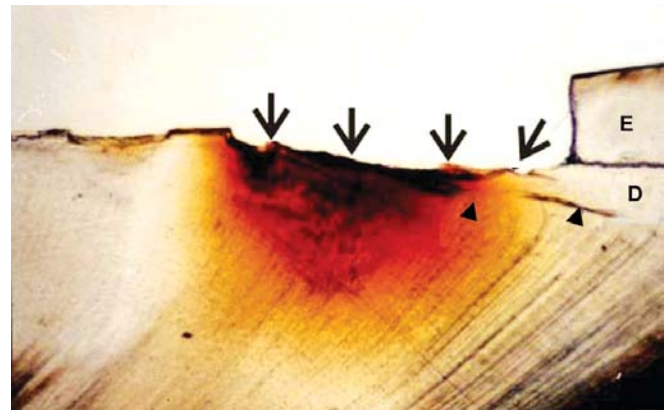


Figure 5.70: Spread of caries to dentin (D) after destruction of enamel by carious process indicated by arrows. The underlying area of dentin is exposed to the surface with deep reddish brown pigmentation showing effect of early caries. There is loss of dentinal tubules in the most superficial part and horizontal cracks indicated by arrow heads appear in the dentin. E= Unaffected enamel. (Ground section x 100)

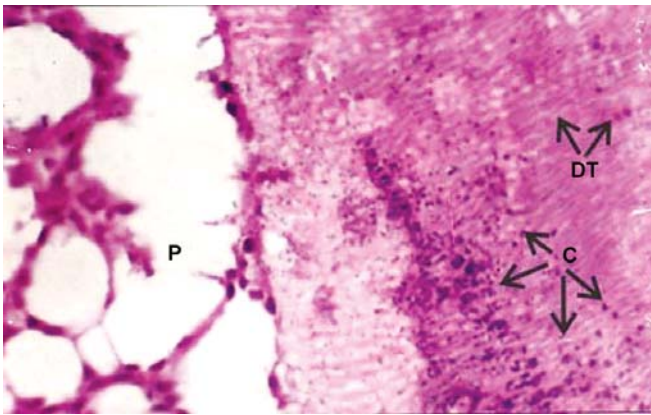


Figure 5.71: *Dental Caries*—Numerous minute round organisms (cocci) can be seen (C) in the dentinal tubules (DT). Some organisms are present in the intertubular dentin as well. Evidence of inflammation can be seen in the pulp (P) (H & E x 400)

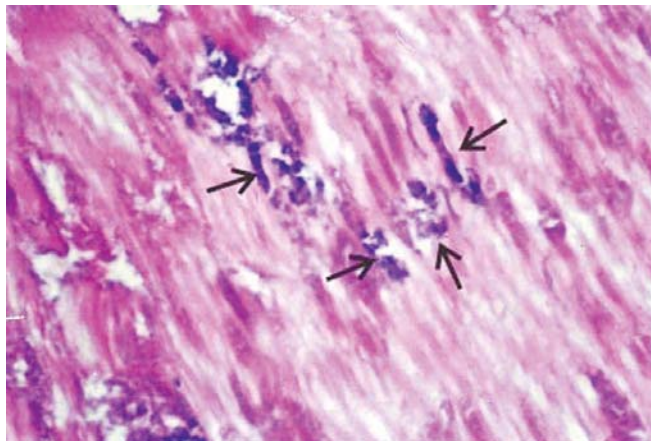


Figure 5.72: The dentinal tubules are packed with organisms (indicated by arrows) and thus form transport channels for infection to pass to the pulp. (H & E x 400)

'liquefaction foci' are formed by the breakdown of a few dentinal tubules (Figs 5.73 and 5.74). This 'focus' is an area of ovoid destruction, parallel to the course of the tubules and filled with necrotic debris that tends to increase by expansion. This produces compression, distortion and disruption of the adjacent dentinal tubules (Figs 5.75 to 5.77). Teeth with deep penetrating carious lesions can be treated only by partial removal of the deep carious lesion and insertion of calcium hydroxide or mineral trioxide aggregate containing dressing materials for a period of a few weeks or months. During this period, the odontoblasts form new reparative dentin along the

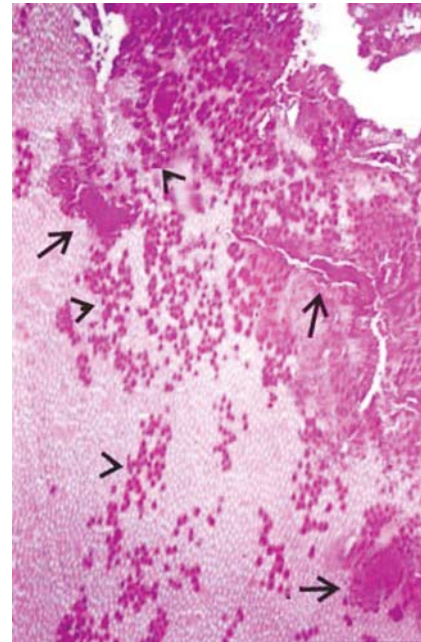


Figure 5.73: Decalcified section of dentin caries show large "foci of liquefaction" (indicated by arrows) due to break down of dentinal tubules. There are large number of degenerated tubules forming dead tracts indicated by arrowheads (H & E x 100)

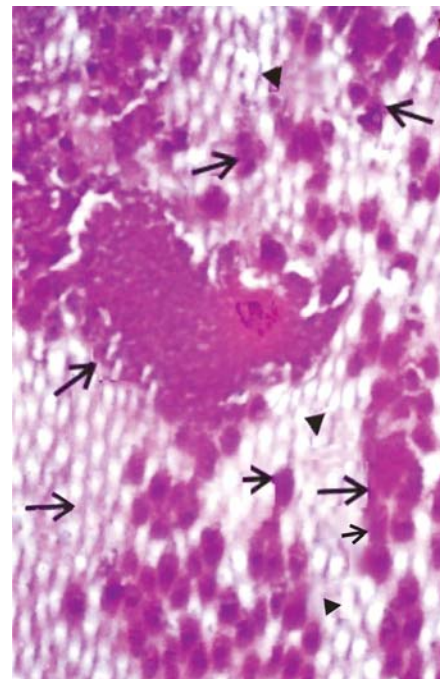


Figure 5.74: Higher magnification of figure 5.73 show "liquefaction foci" indicated by large arrow, degenerated dentinal tubules indicated by small arrows and normal empty dentinal tubules indicated by arrow heads in caries dentin. (H & E x 400)

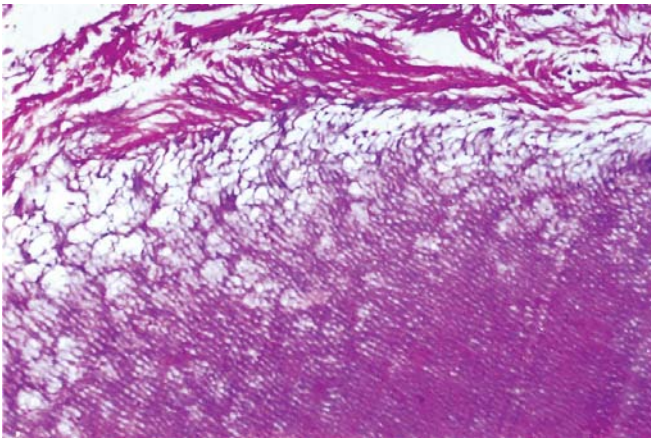


Figure 5.75: *Dentinal caries:* The dentinal tubules are dilated, disorganized, beaded and disrupted (H & E x 150)

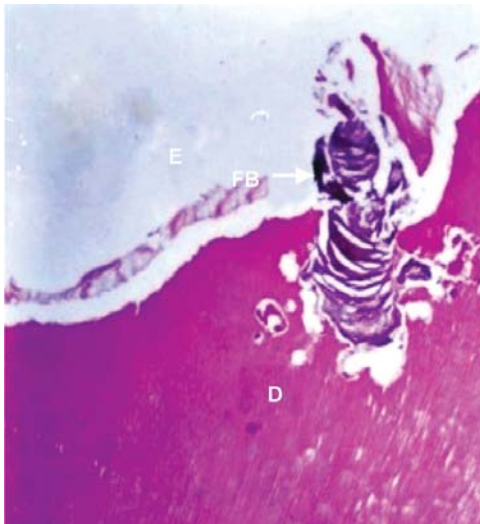


Figure 5.76: Caries involving both enamel (E) as well as dentin (D). The cavity has been filled with restorative material which is projecting into space left by dissolution of enamel and is occupying the dentinal cavity. FB =Foreign body (H & E x 100)

pulpal surface underlying the carious lesion (Figs 5.78 and 5.79). The cavity can be reopened and the remaining carious lesion removed without endangering the pulp. This treatment is known as indirect pulp capping. On removal of the caries, the mesenchymal cells of the cell rich zone differentiates into odontoblasts to replace those that have necrosed. These newly formed odontoblasts can produce well-organized dentin or an amorphous, poorly calcified, permeable dentin. The demarcation zone between secondary and reparative dentin is called the 'calciotraumatic line' (Figs 5.80 and 5.81).

Dentin is highly permeable with dentinal tubules running from the dentinoenamel junction to the pulp. Therefore, the dentinal tubules must be sealed for good restoration. Whenever dentin has been cut or abraded, a thin altered surface layer is formed, called the smear layer (Fig. 5.82). This layer is composed of denatured collagen,

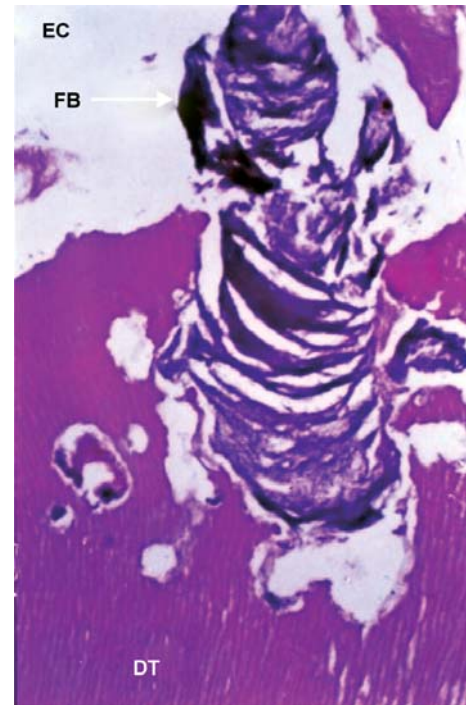


Figure 5.77: Higher magnification of (Fig.5.76) showing foreign body in the cavity of enamel (EC) (partially dissolved during decalcification) and dentin. The dentinal tubules (DT) in adjacent areas are markedly dilated due to caries (H & E x 400)

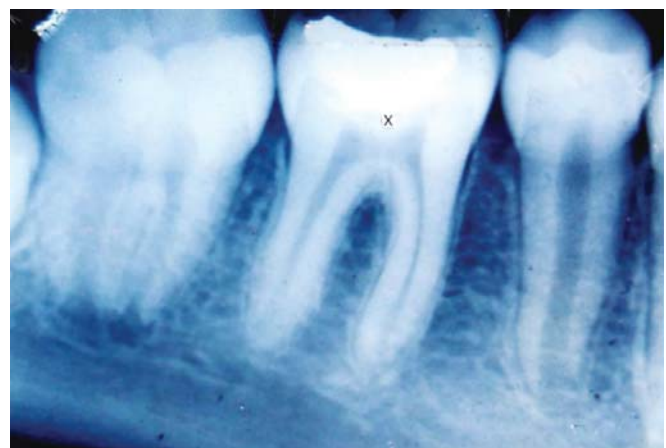


Figure 5.78: Radiograph showing Reparative Dentin (x) whiter than adjacent dentin under the filling

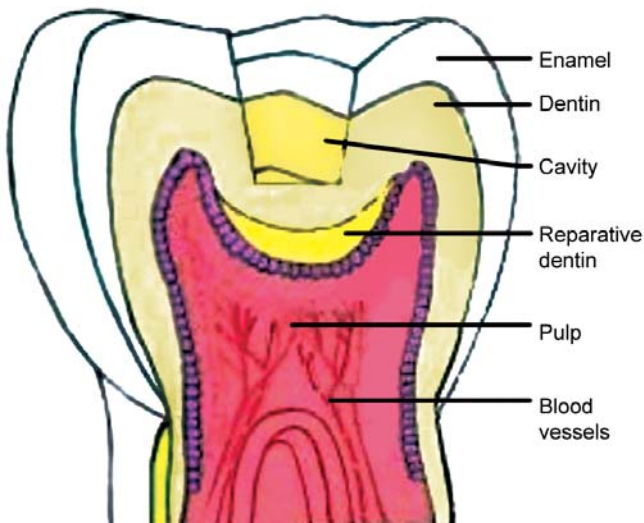


Figure 5.79: Reparative dentin formation in relation to the cavity preparation and subsequent restoration, is limited to zone of stimulation

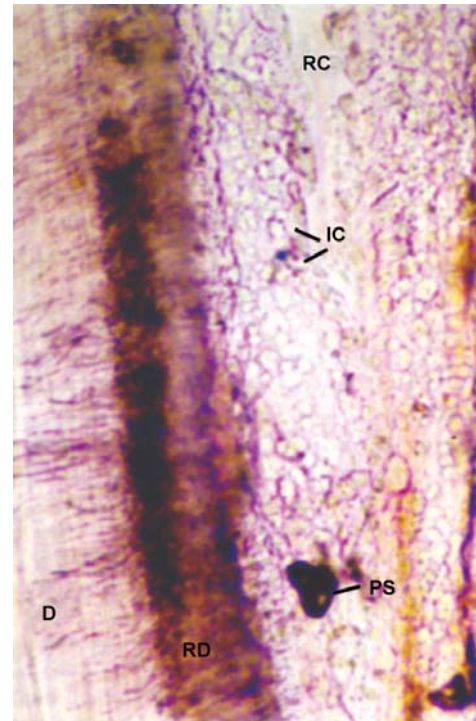


Figure 5.81: Thick layer of secondary dentin present at the floor of root canal sealing it off from the apical foramen. (H & E x 200)

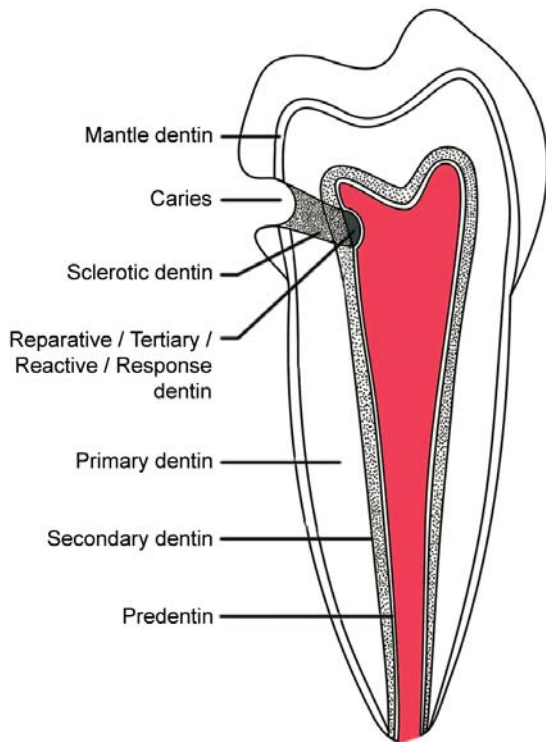


Figure 5.80: Secondary hard tissue apposition with time and/or irritation in mandibular molar. Small black arrows show physiologic secondary dentin and cementum apposition white arrows show formation of reparative dentin in response to irritants. Pulp space undergoes continual reduction in size and volume. At floor of pulp chamber, maximum secondary dentin formation takes place

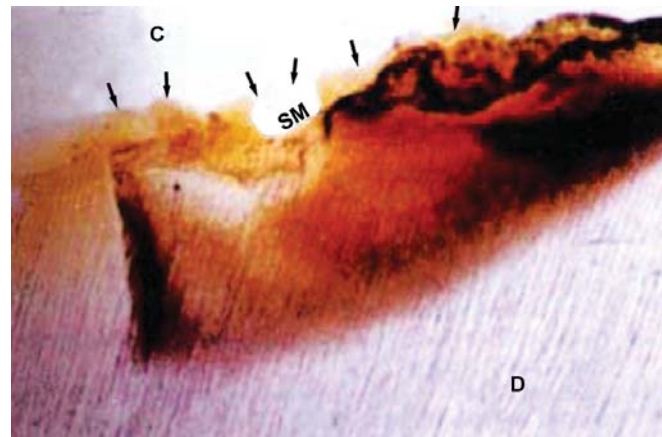


Figure 5.82: Ground section of a prepared cavity (c) in carious tooth. The dentin is covered by a thin layer of amorphous material indicated by arrows. This is called the "Smear Layer" (SM). It is composed of collagen and apatite crystals. Dentin up to certain distance from the caries cavity shows brownish discoloration. Normal dentin (D) x 100

hydroxyapatite and other cutting debris and is only a few microns in thickness. The smear layer acts as a natural barrier over the cut surface since it occludes many of the

dentinal tubules with debris called smear plugs. While the smear layer is a good protective barrier, it is relatively weak and subjected to dissolution by acid. Potassium nitrate is the best option for treatment of hypersensitive dentin. All metallic restorations are good thermal conductors. Therefore, a cement base must be placed under all metallic restorations to protect the pulp by minimizing conduction of heat and cold. Fluoride increases the hardness of dentin if incorporated during dentinogenesis. Deficiency of vitamin D does not affect the formation of dentin.

Reversible pulpitis ranging from hyperemia to mild to moderate inflammatory changes is limited to the area of the involved dentinal tubules, such as dentinal caries. Microscopically, one sees reparative dentin reparative pre dentin disruption of the odontoblast layer, dilated blood vessels and the presence of immunologically competent chronic inflammatory cells (Fig. 5.83).

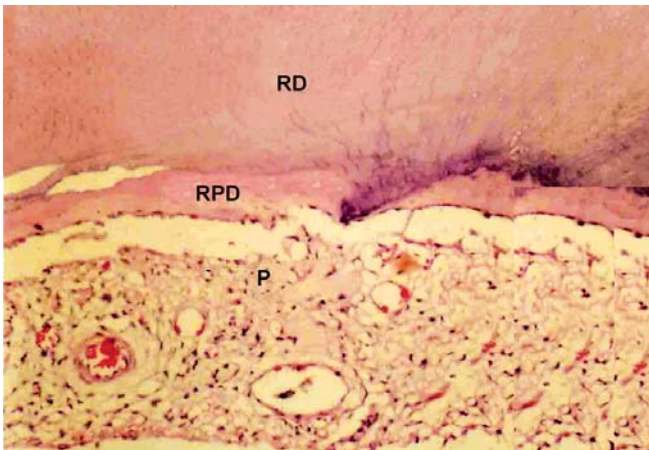


Figure 5.83: Thick layer of reparative dentin (RD) and thin layer of reparative predentin (RPD) are formed sealing off the pulp cavity from source of infection. (H & E x 200)

The localization of type III collagen in dentin is found in (a) dentinogenesis imperfecta, (b) osteogenesis imperfecta.

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Dental Pulp

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INTRODUCTION

The pulp has its genesis during the initiation of tooth development at about the sixth week of intrauterine life. The pulp is a vascular mesenchymal connective tissue that occupies the cavity in the center of the tooth within rigid dentinal walls. The pulp organ present within the pulp cavity can be distinguished into the coronal pulp and the radicular pulp. It is surrounded by dentin on all sides except at the apical foramen and accessory pulp canal openings, where it is in communication with periodontal soft tissue (Figs 6.1 and 6.2). Each individual has 52 pulp organs out of which 20 are present in the deciduous (primary) teeth, and 32 in the permanent teeth. The total volume of all the pulp organs that are present in permanent teeth is about 0.38 cc. The volume of a single permanent tooth pulp organ is about 0.02 cc. The number of pulp horns present in coronal pulp depends on the number of cusps present. The incisor pulp organs are three to four times smaller than molar pulp organs. The pulp horns gradually recede with age. Cells of the pulp convert to odontoblasts that form secondary and reparative dentin at the required site, for example, adjacent to the carious dentin, cavity preparation, attrition, abrasion and erosion, etc.

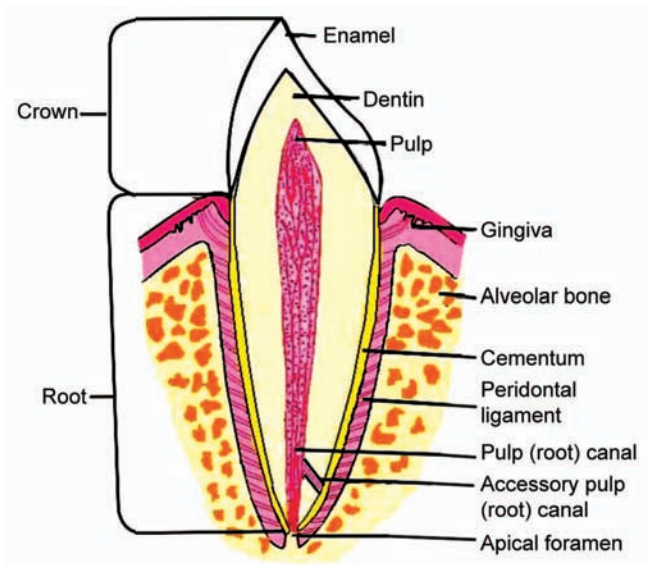


Figure 6.1: Longitudinal section of mandibular incisor and supporting structures

ANATOMY OF PULP

The pulp space, that is, the pulp chamber and root canals are enclosed by dentin. They are filled with soft connective

tissue, called pulp. The dentin is hard while pulp is soft tissue. Embryologically, histologically and functionally the dentin and pulp are similar tissues. The pulp is dried out and lost in dried tooth, leaving an empty chamber and root canals (Fig. 6.2). Because of this reason dentin and pulp are anatomically described in two separate chapters.

Shape of Pulp Chambers and the Pulp Canals of Permanent Teeth

Maxillary Teeth

Central incisor: In the crown, it is similar to the blade of the shovel with three small horns on the incisal edge. Gradually it tapers towards root end. In cross-section, it is almost a triangle with the point of triangle palatally.

Lateral incisor: Overall it is smaller than central incisor. Coronally it is spoon shaped. It gradually tapers towards the root apex.

Canine: Since it is the longest tooth, it has the longest pulp canal. In transverse section it is almost elliptical in shape. The root apex and the pulp inside the root apex are usually slightly distally inclined.

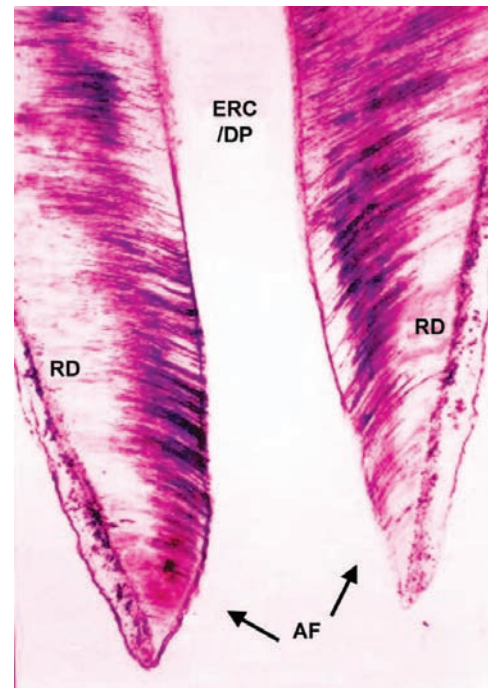


Figure 6.2: Empty root canal (ERC) due to dried pulp (DP) which gets lost in dried teeth. Root canal is surrounded by root dentin (RD) except at apical foramen (AF) where it communicates with periodontal tissue. Ground section x 100

First premolar: Its pulp chamber is occlusocervically large. On the mesial surface, there is a concavity extending from the cervical third of the crown to the root trunk. The crown portion divides into two small funnel-shaped roots. In crown, the buccal pulp horn is bigger than lingual.

Second premolar: Coronally, it is like the first premolar. It has only one root. In crown, the buccal and lingual pulp horns are almost equal.

Molars: Their pulp is generally similar to the premolar. Coronal portion is nearly rectangular but slightly larger mesio-distally. They have three roots, two buccal and one palatal. The buccal roots are one distobuccal and one mesiobuccal. Palatal root is the longest and distobuccal is the shortest and relatively straight.

Mandibular Teeth

Central incisor: Its pulp is very small in volume. It is long and narrow. In cross-section it has bucco-lingually flattened elliptical shape.

Lateral incisor: Its pulpal anatomy is almost the same as that of the central incisor, but it is slightly smaller in all dimensions.

Canine: Its pulpal anatomy is similar to the maxillary canine.

First premolar: Its pulpal anatomy is similar to the mandibular canine with a very small or insignificant lingual pulp horn.

Second premolar: Its pulpal anatomy is like maxillary second premolar. The buccal horn is bigger than the lingual pulp horn.

Molars: Pulpal anatomy of all the molars is almost similar, but gradually from first to third molar their size reduces. In coronal cross-section, pulp is mostly rectangular with greatest mesiodistal dimension. According to height the pulp horns from highest to lowest are (a) mesiobuccal, (b) mesiolingual, (c) distobuccal, and (d) distolingual. In ninety five percent of molars, there are two roots and three root canals. Mesial root contains two canals.

Cavity of Pulp and Pulp Organ

The cavity of pulp (pulp organ) can be divided into two parts.

1. Coronal pulp
2. Radicular pulp or root canal

Coronal Pulp

Coronal Pulp is the part of the pulp which remains in the crown of the teeth. In young teeth, the shape of the coronal pulp in the pulp chamber is similar to the outer surface of the dentin of the crown. There are six surfaces in the coronal pulp: (a) occlusal or roof or pulpal (b) floor or subpulpal, (c) mesial, (d) distal, (e) buccal, and (f) lingual or palatal. Below the cusps are pulp horns that extend into the cusp of the crown. The number of the horns are same as that of the number of cusps. Gradually with age, pulp chamber becomes smaller due to the continuous secondary and tertiary dentin deposition.

Radicular Pulp

The pulp that remains in the root of the tooth is called the radicular pulp. Anterior teeth have single and posterior teeth have multiple branches of pulp in the pulp canals of the root.

Radicular pulps are not always straight. They vary in size, shape and number. They are connected with the periapical tissues through the apical foramen or foramina and accessory pulp canal. The dentinal walls and radicular pulp taper apically.

During root formation, the apical opening is wide-open, limited by an epithelial diaphragm. Gradually, more dentin is formed as the growth proceeds. When dentin of the root is completely formed (in permanent teeth approximately after two to three years of eruption), the apical foramen and the pulp canal become narrower (Fig. 6.3).



Figure 6.3: Pulp organs of permanent teeth. Upper row-maxillary teeth central incisor to third molar. Lower row- mandibular teeth central incisor to third molar. Accessory pulps are also seen branching out from radicular pulp

Apical Foramen

In the maxillary teeth, apical foramen is wider (average 0.4 mm in diameter) than in mandibular teeth (average 0.3 mm in diameter). The shape and the location of the apical foramen undergo changes due to functional influences on the teeth. A tooth may migrate mesially, causing the apex to tilt in the opposite direction.

Occasionally the apical foramen is situated on the lateral side of the root, even in cases where the root is straight. Sometimes there are two or more foramina separated by dentin and cementum, or by cementum only (Figs 6.4A and B).

Accessory Canals

These are extra root canals, besides the main canal, which connect pulp with periodontal tissues. They may be present anywhere along the root but are most numerous in the apical third (Figs 6.1 and 6.5 to 6.7). They may spread infection both ways from the pulp to the periodontal ligament and vice versa.

Development: They usually develop in areas of premature loss of cells of root sheath. These root sheath cells initiate the formation of odontoblasts that form dentin. Lateral canal may develop if the developing root comes in contact

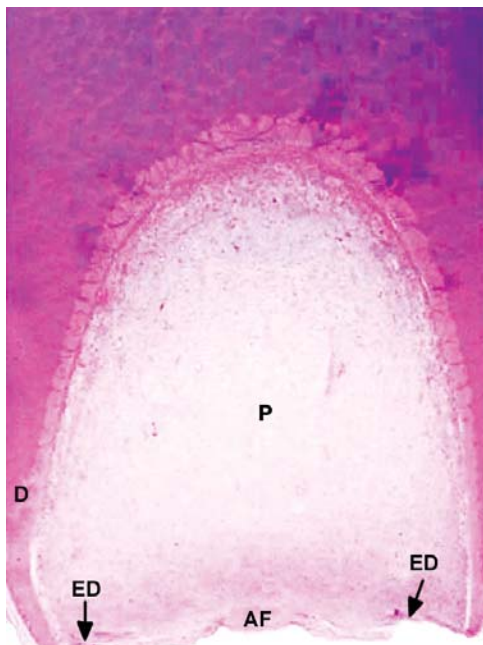


Figure 6.4A: Development of apical foramen: Section of developing deciduous tooth showing dentin (D), Pulp (P) and apical foramen (AF) limited on both sides by the epithelial diaphragm (ED) x 40

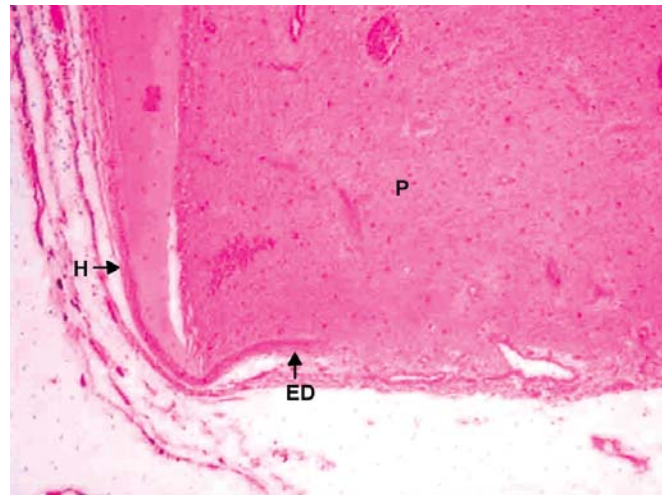


Figure 6.4B: Development of apical foramen: Higher magnification of developing deciduous tooth showing epithelial diaphragm (ED), from the column of epithelial cells of root sheath of Hertwig (H). Pulp (P) x 100

with a blood vessel. The dentin encircles the blood vessel and a lateral or accessory canal is formed, which connects the radicular pulp to the periodontal tissues.

Basic Structure of Pulp

The dental pulp is a connective tissue that is soft in nature. Two distinct regions are present in the pulp—the central region and the peripheral region.

Central Region

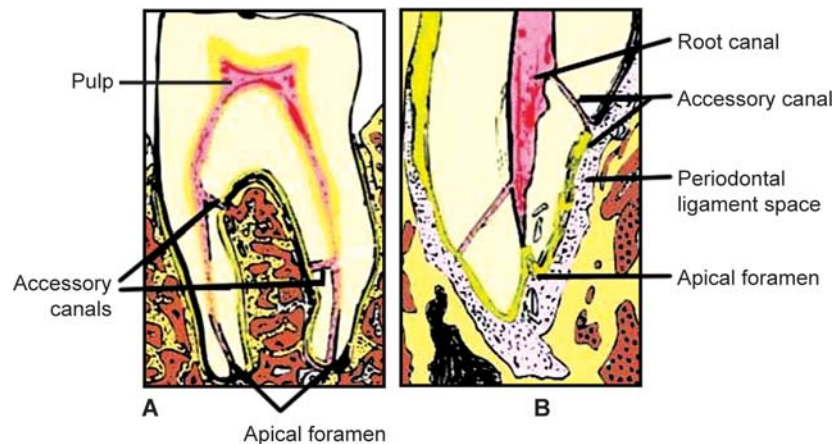
Central pulp core region is present in both the coronal as well as in the radicular pulp. Major blood vessels and nerves of the pulp are present in this region.

Peripheral Region

This region specially in young age is also known as the odontogenic region. Three zones are present in this region, these are as follows (Fig. 6.8).

1. Odontoblastic zone or the dentin forming zone
2. Cell-free zone or zone of Weil
3. Cell rich zone.

Odontoblastic zone or the dentin-forming zone: Odontoblasts are dentin-forming cells. A layer of these cells is found near the dentinal end of the pulp and this constitutes the odontoblastic zone.



Figures 6.5 A and B: Location of accessory canals: (A) In the cervical and middle thirds of the roots; (B) In the apical third of the root

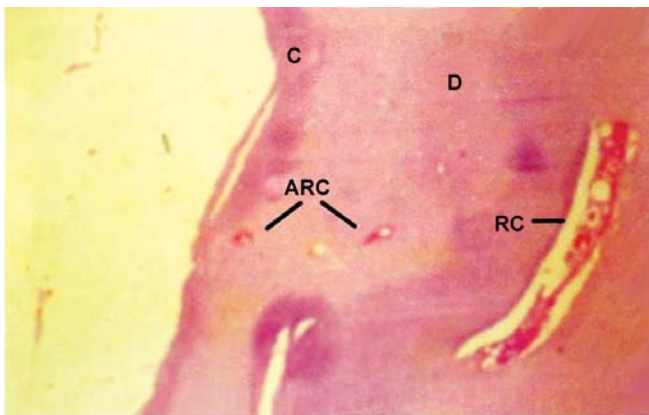


Figure 6.6A: Accessory root canal: Decalcified tooth showing cross sections of accessory root canal traversing the dentin (D) and cementum (C). (RC) Root canal, (H & E x 100)

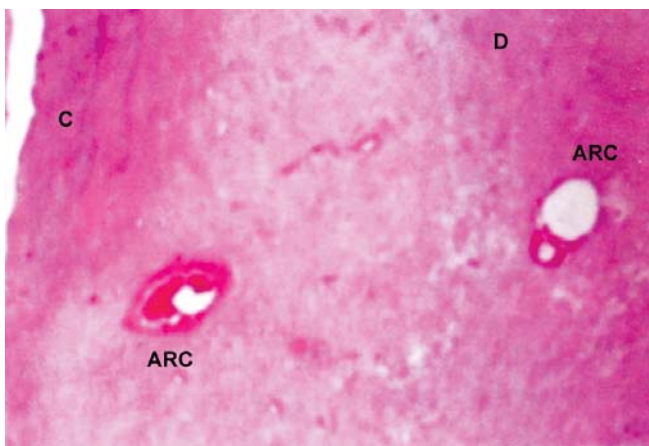


Figure 6.6B: Higher magnification of figure 6.6A. Accessory Root Canal: Blood vessel can be seen in accessory root canal (ARC). D = Dentin, C = Cementum (H & E x 400)

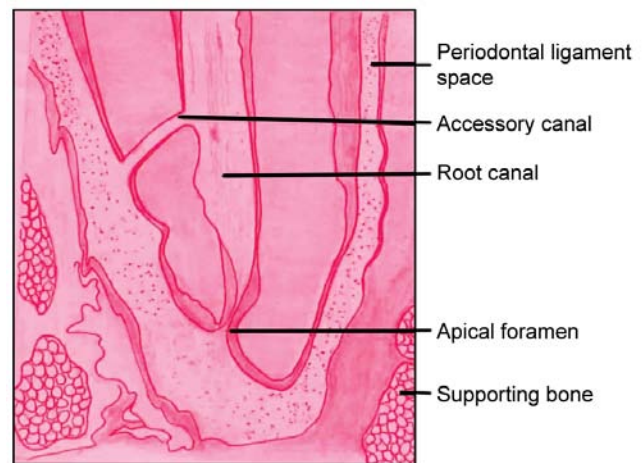


Figure 6.7: Apical foramen and accessory canal on the side of apex (as seen in longitudinal ground section of tooth)

Cell-free zone or zone of Weil: A cell-free zone is a space found below the odontoblastic zone. During the development of a tooth, odontoblasts may move toward the pulp in this zone. In functional teeth, there may be limited movement of the odontoblasts in this zone.

Cell-rich zone: It is present below the cell-free zone or outer to the central region. Mainly fibroblasts and undifferentiated mesenchymal cells are present in this zone.

Usually, shape of pulp cavity follows the general outline of the surrounding dentin of the individual tooth.

Contents of the Pulp

The soft tissue of the tooth, the pulp consists of Cells and Intercellular substances (Fig. 6.9 and Table 6.1).

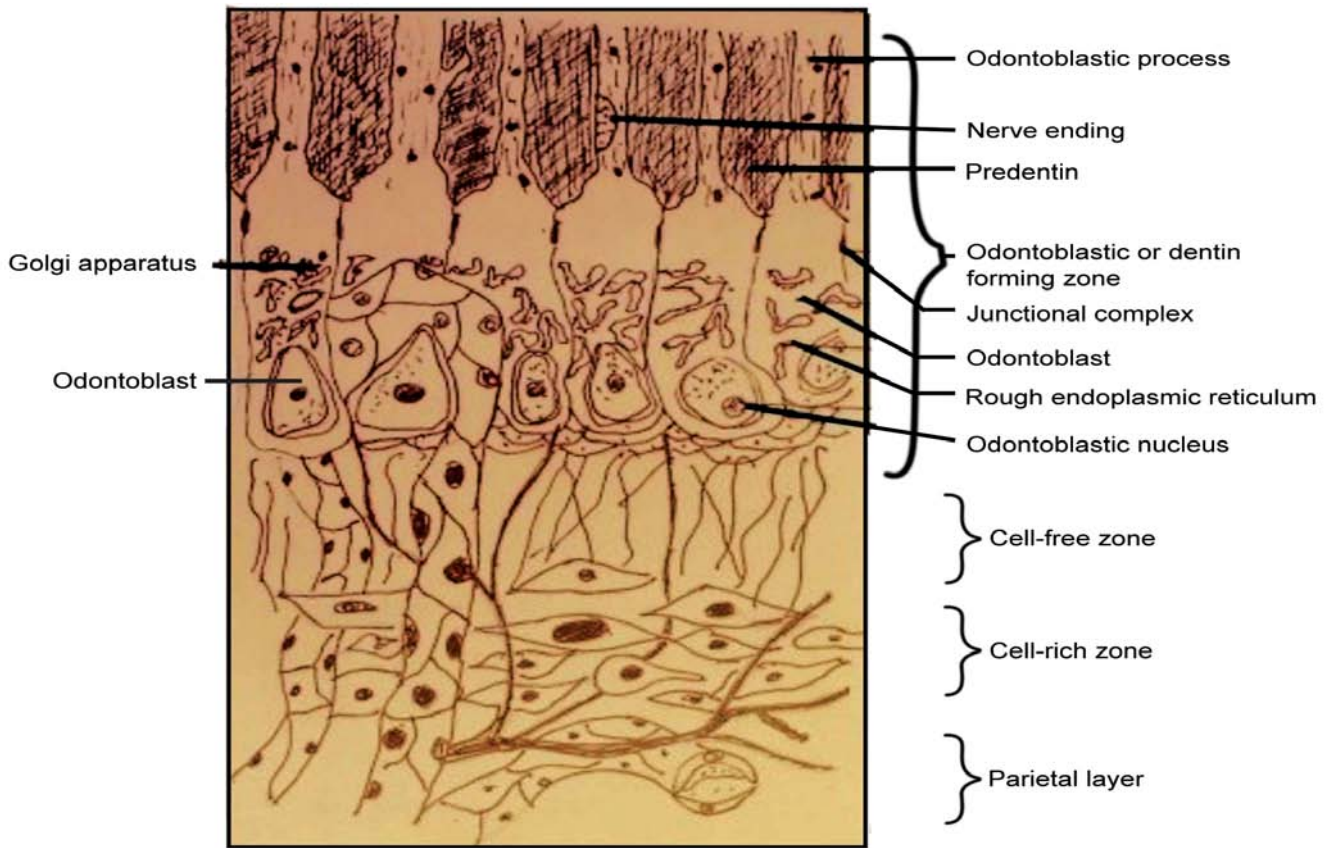


Figure 6.8: Section of the pulp at odontogenic zone showing odontoblast, cell-free zone, cell-rich zone, with blood vessels and non- myelinated nerves

Table 6.1: Contents of the pulp	
A. Cells:	1. Odontoblasts 2. Fibroblasts 3. Undifferentiated mesenchymal cells 4. Defense cells
B. Intercellular Substances:	5. Fibers: i. Precollagenous ii. Collagenous 6. Ground substance 7. Blood vessels 8. Lymph vessels 9. Nerves

CELLS

Odontoblasts

Odontoblasts are the second most prominent cells in the pulp. These cells line the periphery of the pulp adjacent to the predentin, which is also called as the “odontogenic

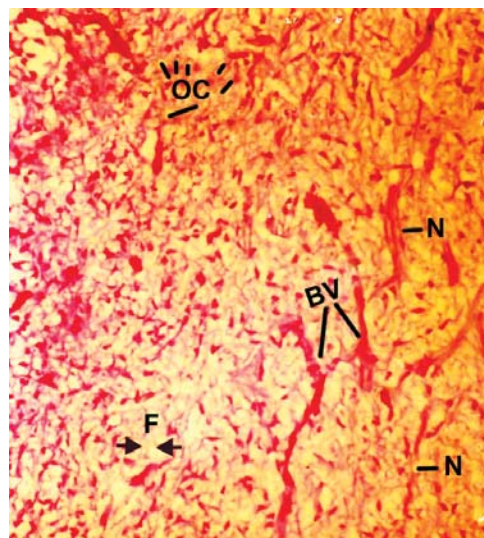


Figure 6.9: Normal pulp: Decalcified tooth: Pulp consists of fibroblasts (F), other cells (OC) and loose fibrous tissue, blood vessels (BV) and nerve twigs (N), that travel through the fibrous tissue. (H and E x 100)

zone of the pulp". The odontoblasts are tall, columnar in the crown, become cuboidal in the middle of the root and flat, spindle-shaped near the apex of the tooth (Figs 6.10 to 6.12 A and B).

Odontoblasts are approximately 5 to 7 microns in diameter and 25 to 45 microns in length. The cell bodies of the odontoblasts are columnar in appearance and contain large oval nuclei, which fill the basal part of the cell. The cells in the odontoblastic layer lie very close to each other, and the plasma membrane of adjacent cell exhibits junctional complexes and desmosomes (Fig. 6.13). Rough endoplasmic reticulum and Golgi apparatus are present in the cell body. This indicates an exchange of ions and small molecules. Odontoblasts that are in an active secretory phase have basophilic cytoplasm, prominent golgi zones, numerous vesicles, endoplasmic reticulum, mitochondria, open faced nucleus with many nucleoli, and scattered chromatin and membrane-bound granules, like lysosomes.

Cytoplasm of the young cell body differs from that of the older cell. During active phase of the cell, all cell organelles are more prominent and abundant. The cell increases in size and its process also lengthens as dentin formation occurs.

Odontoblasts that are in the resting period have hemotoxophilic (closed) nucleus, little cytoplasm, condensed chromatin, reduced number of endoplasmic reticulum, absence of secretory granules and flattened morphology. The terminal bar apparatus of the odontoblast consists of a clear terminal part of the cell

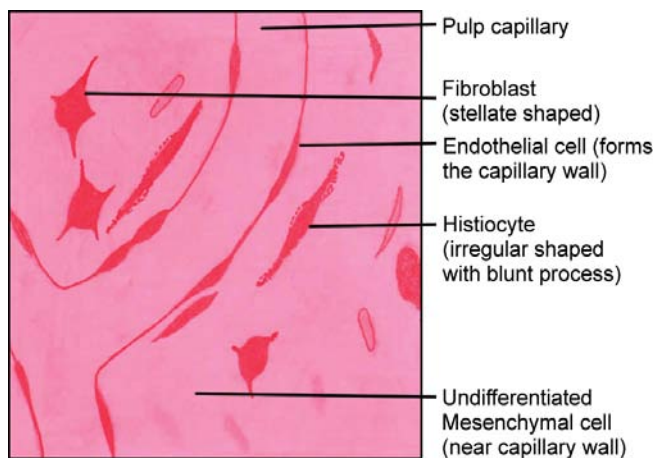
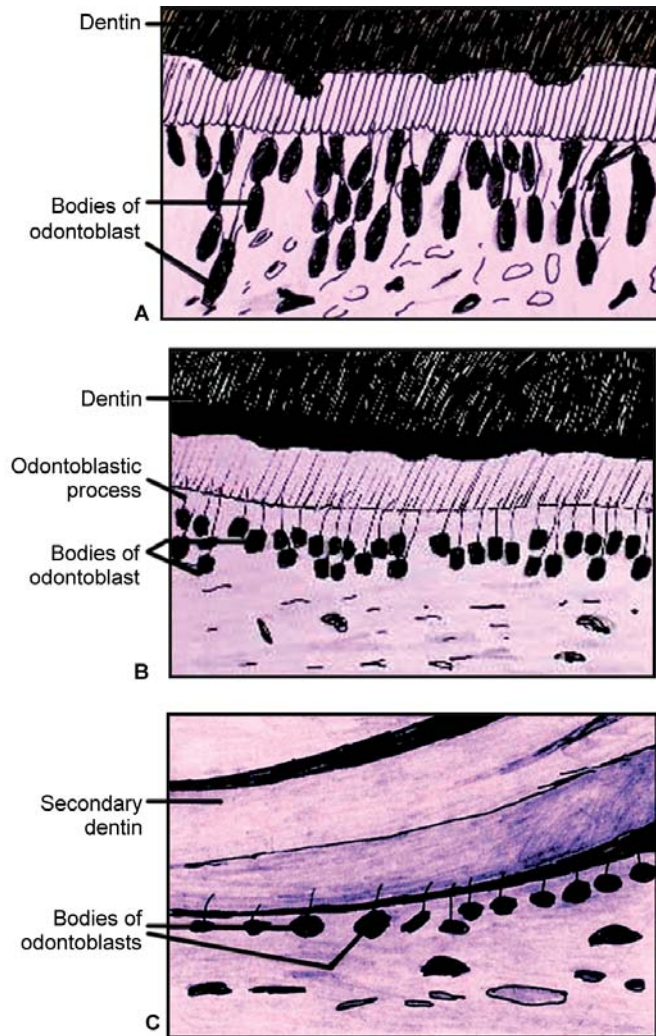


Figure 6.10: Different type of cells in pulp under high power magnification



Figures 6.11A to C: Variations of odontoblasts in different regions of one tooth—The formation and arrangement of the odontoblasts is not uniform throughout the pulp. (A) Tall columnar odontoblasts are present in the crown. (B) More cuboidal odontoblasts are present in the middle of root (C) Flat or irregular appearance of odontoblasts is present in the apical region of root

body and the adjacent intercellular junction. The terminal bars separate the odontoblasts from each other by intercellular condensation. Each cell gives off one main and several smaller cytoplasmic processes that extend into the dentin (Figs 6.14 and 6.15). The main process is called odontoblastic process. The odontoblastic process is devoid of major organelles. During early period of active dentinogenesis, it contains microtubular filaments and coated vesicles. These organelles are less frequently seen in the later stage of dentinogenesis.

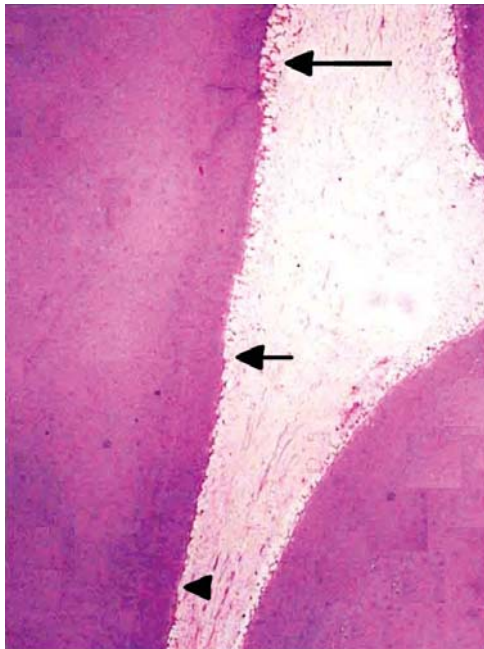


Figure 6.12A: Pulp: Length of odontoblast gradually decreases towards the root apex. Tall columnar odontoblasts (shown by long arrow) are present near pulp chamber. Shorter odontoblasts (shown by short arrow) are present towards root canal. Flattened odontoblasts (shown by arrow head) are observed towards the root apex (x 40)

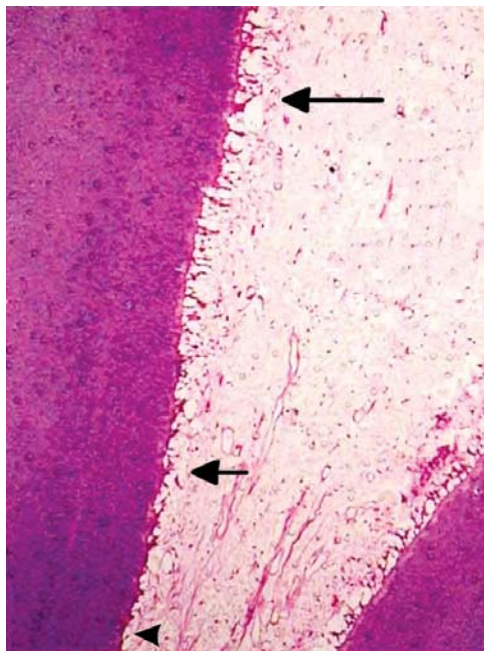


Figure 6.12B: Figure 6.12 A as observed under higher magnification (x 200)

Fibroblasts

Fibroblasts are cells that occur in greatest amount in the pulp tissue. The function of fibroblasts is to form and maintain the collagen fiber and ground substance throughout the life of the pulp. The fibroblasts also have the capability of ingestion and degradation of the collagen fibers and ground substance. In case of young teeth, the fibroblasts have abundant cytoplasm and cell organelles and are active in protein synthesis. The fibroblasts are stellate-shaped cells having extensive processes that communicate with the processes of other pulpal fibroblasts to form a syncytium (Figs 6.16 and 6.17 A

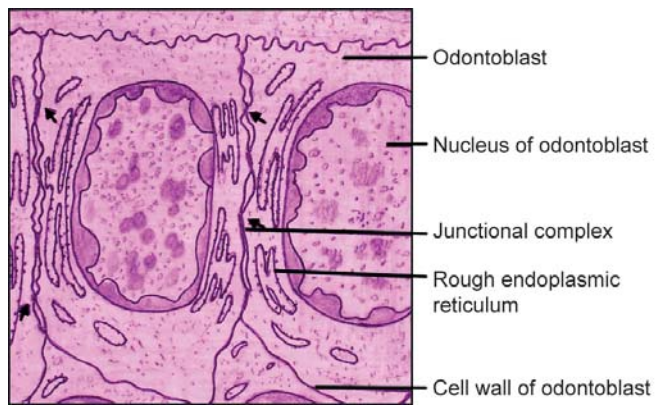


Figure 6.13: Junctional complexes (arrows) between cross-section of two adjacent odontoblasts (as seen under electron microscope)

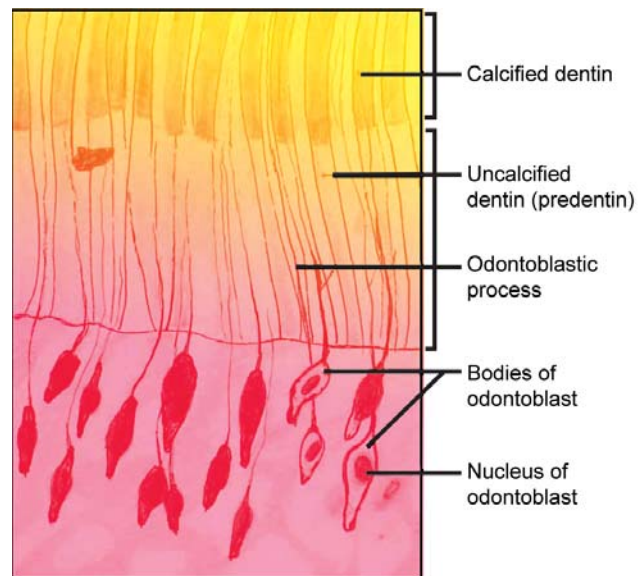


Figure 6.14: The odontoblast cells reside at pulp-predentin border and their processes extend into dentinal tubules

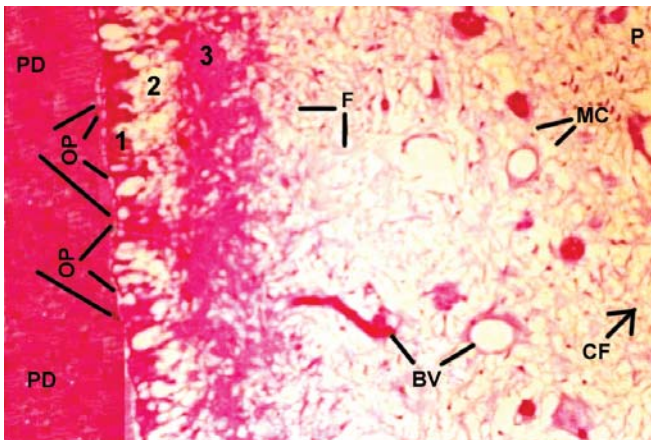


Figure 6.15: Decalcified section of tooth showing three distinct zones of peripheral pulp. Zone 1 represents odontoblastic or dentin forming zone. Adjacent to the odontoblasts, thin odontoblastic processes (OP) are seen entering the predentin (PD) to form dentinal tubules. Zone 2 represents cell-free zone or zone of Weil. Zone 3 represents cell rich zone. The central region of pulp (P) contains fibroblasts (F) and thin collagen fibers (CF), macrophages (MC) and many blood vessels (BV) (H & E x 100)

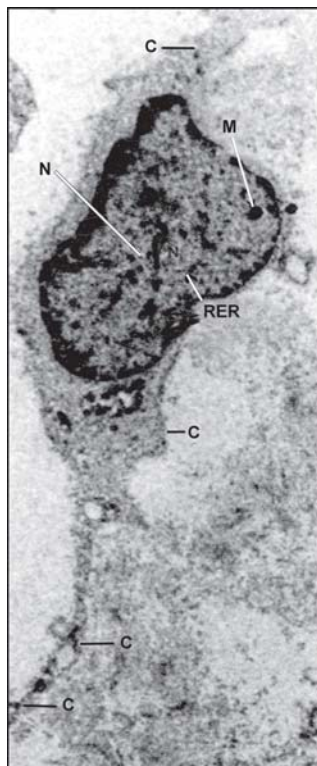


Figure 6.16: Electromicrograph of a fibroblast of pulp showing markedly elongated cytoplasmic process (C) of spindle-shaped cell. Nucleus (N) is vesicular in nature. M= Mitochondria RER= Rough Endoplasmic Reticulum H and E (H & E x 3300)

and B). Syncytium is a multinucleate mass of protoplasm produced by the merging of cells.

In older pulps, the fibroblasts appear spindle shaped or round shaped with short processes and have few cytoplasmic organelles. Such cells are termed fibrocytes. In the course of development, the relative number of cellular elements in the pulp decreases, whereas the fiber population increases. Deep stained nuclei with basic dye and homogenous light stained cytoplasm are seen under light microscope. Abundant rough endoplasmic reticulum, mitochondria and other organelles in fibroblast cytoplasm are seen under electron microscope (Fig. 6.16) Fibroblasts help in the inflammatory and healing process by the secretion of growth factors, cytokines, colony stimulating factors, FG F-2 and VEGF.

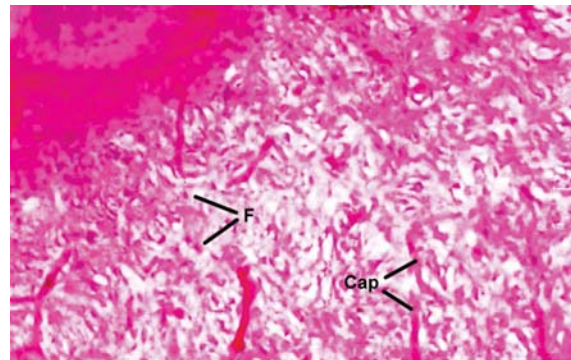


Figure 6.17A: Tooth of a young child showing cellular pulp, rich in fibroblasts (F) and capillaries (Cap). Collagen fibers are very thin and bundles of fibrous tissue are not present (H & E x 200)

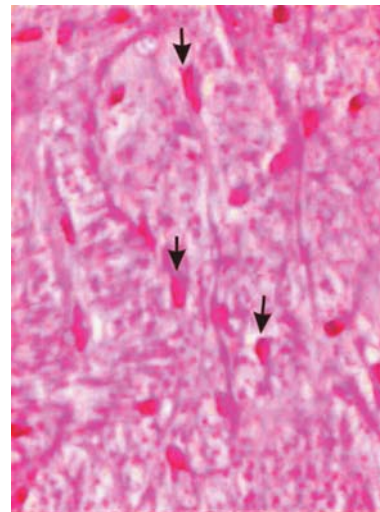


Figure 6.17B: Pulp: Pulp is made up of fine collagen fibrils which are laid down by fibroblasts (shown by arrows). Fibroblasts are thin spindle-shaped cells with elongated nuclei x 400

Undifferentiated Mesenchymal Cells

Undifferentiated mesenchymal cells are polyhedral shaped cells, larger than the fibroblasts, having a large oval staining centrally placed nucleus with peripheral processes. They are the primary cells in the young pulp. They are mostly found in the cell rich zone and few of them occur in the central area of the pulp. These cells are believed to be totipotent in nature. Totipotent cells are those cells that can differentiate into the cells of all types as per requirement of the situation. They are capable of differentiating into macrophages, odontoblasts or fibroblasts. These cells are mostly seen in the young pulp and decrease in number as age increases. They are also found along the blood vessels in the central pulp (Figs 6.18 to 6.20).

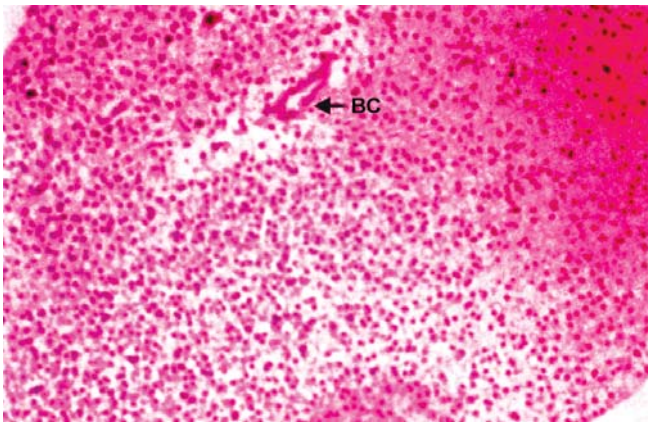


Figure 6.18: Pulp: Extremely cellular pulp in the developing tooth: BC= Blood capillary. The cells are round and represent immature mesenchymal cells. (H & E x 100)

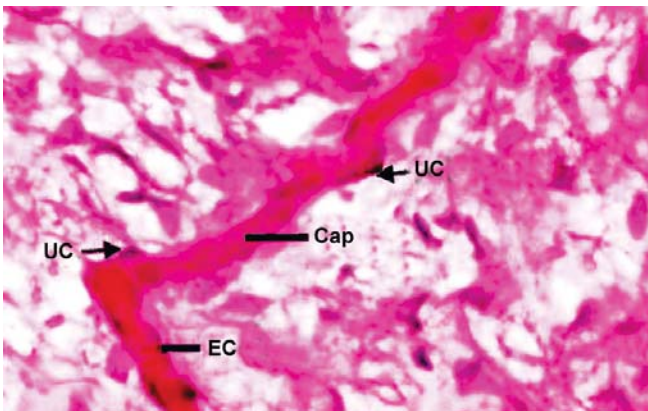


Figure 6.19: Pulp: Capillary (Cap) in the pulp shows thin flat endothelial cell (EC) lining. The plump cells located on the surface of the capillary represent undifferentiated cells (UC) (H & E x 400)

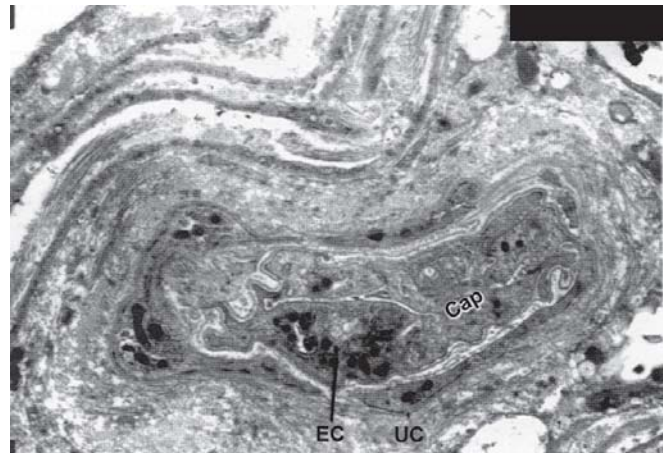


Figure 6.20: Capillary (Cap) wall is lined by swollen endothelial cells (EC). Flattened undifferentiated mesenchymal cells (UC) are seen on the outer surface of capillary. (Electron micrograph x 3800)

Defense Cells

A number of cells are found in the dental pulp that act as defense cells of the pulp. They include mast cells, plasma cells, histiocytes or macrophages. In addition, vascular cells such as the neutrophils, eosinophils, basophils, lymphocytes and monocytes are also present in the pulp (Fig. 6.21). These vascular cells emigrate from the blood vessels and develop a characteristic response to inflammation (Figs 6.21 to 6.25).

Histiocytes (Fixed Macrophages – Resting Wandering Cells, Adventitial Cells): The histiocyte or macrophage is an irregular spindle-shaped cell. These cells have a small round nucleus and granular cytoplasm with short blunt processes. During pulpal inflammation, these cells increase in size and their nuclei appear with a prominent nucleolus. This can be seen by toluidin blue, an intravital dye. These cells are involved in the elimination of dead cells and removal of bacteria. These cells are usually associated with small blood vessels and capillaries of the pulp. The vesicles or phagosomes, which contain phagocytized dense irregular bodies are a characteristic feature of macrophages.

Mast Cells: These cells are found in large numbers along blood vessels in an inflamed pulp. They have a small round nucleus and granular cytoplasm (Fig. 6.26).

Plasma Cells: These cells are seen during inflammation of the pulp. They are responsible for production of antibodies. Observed with the light microscope, the

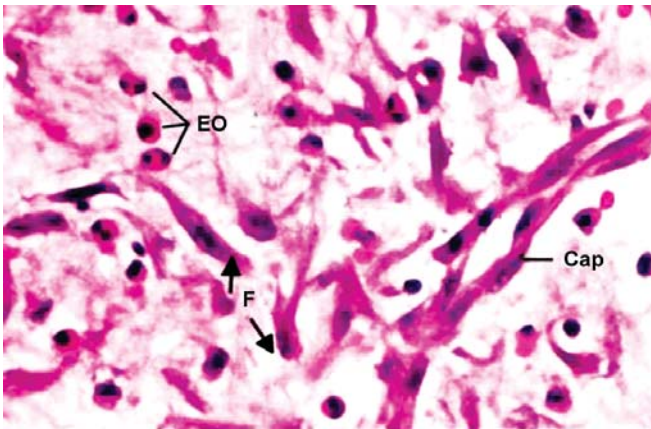


Figure 6.21: Pulp showing cellular infiltrate rich in eosinophils (EO), capillary (Cap) and plump fibroblast (F). (H and E x 400)

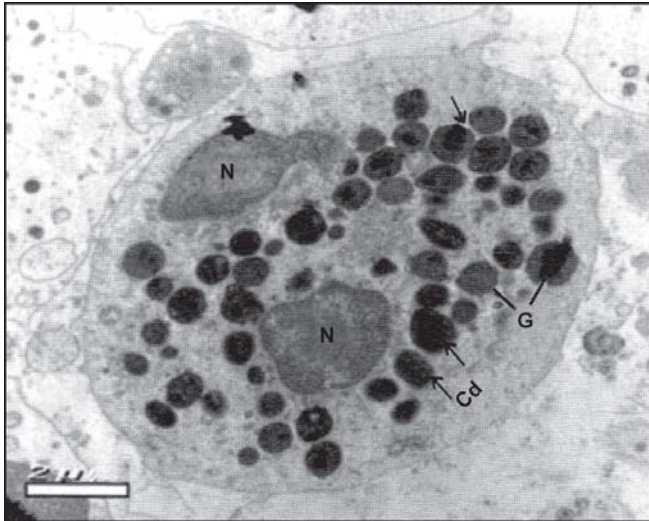


Figure 6.22: Eosinophil: Electromicrograph showing bilobed nucleus (N) with presence of dark granules (G) in the cytoplasm. Many granules show electron dense crystalloids (Cd) indicated by arrows in their center. These crystalloids are characteristic of eosinophils (x 10,000)

plasma cells have small concentric nuclei. Cytoplasm is basophilic and densely packed with rough surface endoplasmic reticulum.

The radiating chromatin adheres to the nuclear membrane and gives the appearance of a cartwheel. These cells contain densely packed rough endoplasmic reticulum. The mature and immature plasma cells may be found in the dental pulp. Both lymphocytes and eosinophils are found in the dental pulp. They increase in number during inflammatory conditions (Figs 6.27 to 6.34).

Pulpal stem cells: Pulpal stem cells are pluripotent cells and produce dentin in response to an injury. Stem cells contain dentin sialoprotein which is a marker of dentin synthesis.

Other Cells of Dental Pulp

In unerupted and in newly erupted teeth, some focal accumulations of lymphoid cells are observed. When monoclonal antibody labeling of normal human dental pulp was done, these lymphocytes were indicated as T lymphocytes, which are associated with the immune defense system.

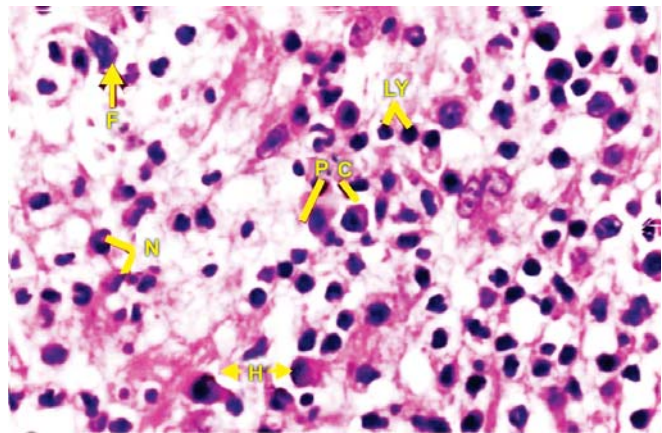


Figure 6.23: Increased cellularity of Pulp in pulp infection showing larger number of inflammatory cells. Pc= Plasma cell, Ly= Lymphocyte, H = Histiocyte, N = Neutrophil, F = Fibroblast (H and E x 400)

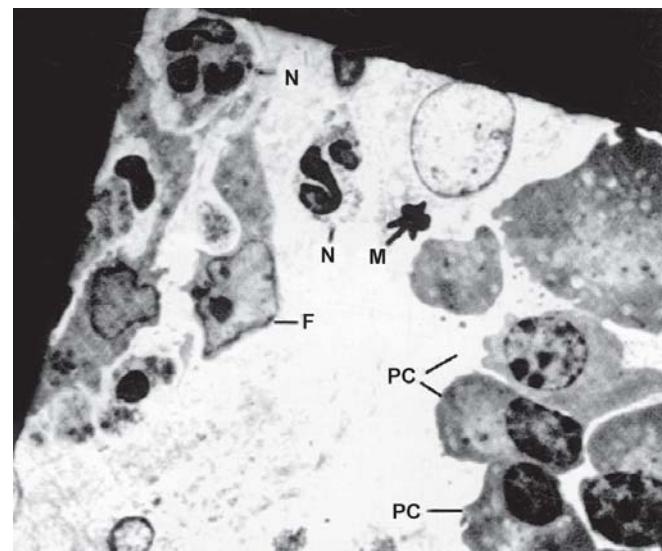


Figure 6.24: Inflammatory cells seen in pulp inflammation. N= Neutrophil, M= Macrophage, Pc = Plasma cell. Fibroblast (F) is also seen. (Electron Micrograph x 1800)

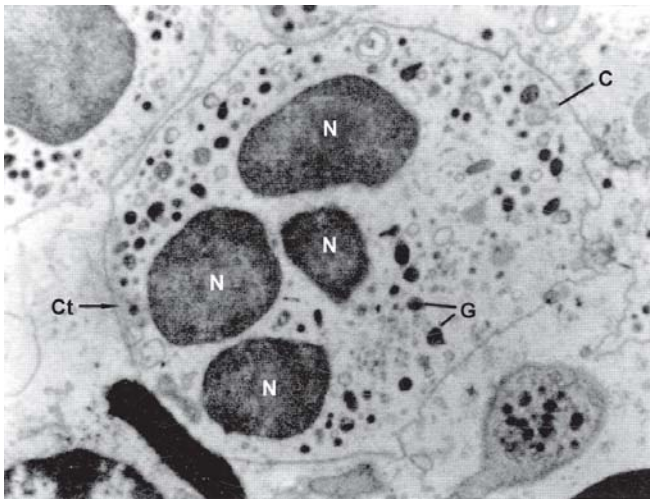


Figure 6.25: Neutrophil showing 4 lobes of the nucleus (N) and presence of dense granules (G) in the cytoplasm (Ct) which is rich in other cell organelles also (Electronmicrophotograph x 10,000)

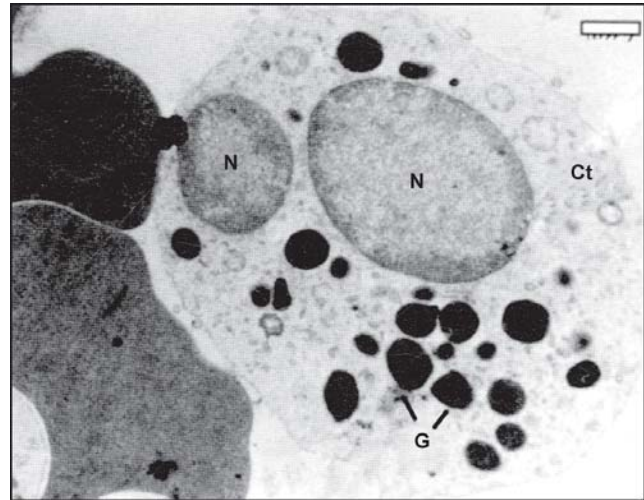


Figure 6.27: Electron micrograph of basophil showing bilobed nucleus (N) and presence of dark electron dense granules (G) in the cytoplasm (Ct) x 12000

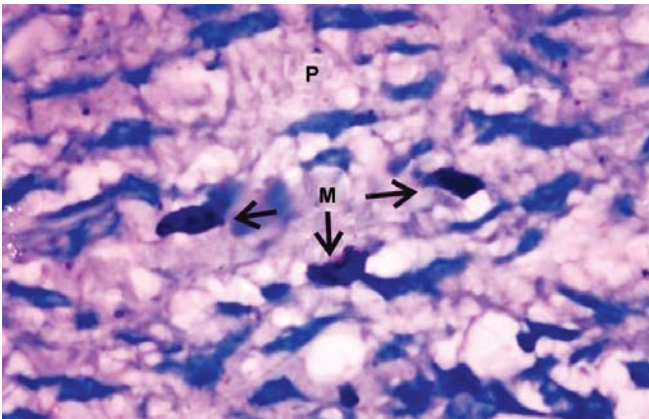


Figure 6.26: Mast cells in the pulp (P) showing deep basophilic granular cytoplasm. M= Mast cells, ZN stain x 200

Presence of dendritic cells is also indicated by monoclonal antibody labeling. The function of these cells is same as that of Langerhan's cells found in the epithelium as they capture foreign antigen and present it to T cell after processing (Fig. 6.35).

Dendritic cells help in immunosurveillance and these cells are more abundant in areas affected by caries, attrition or restorative procedures.

Intercellular Substance

Intercellular substance consists of ground substance and fibers. It is dense and gelatinous in nature and is also called as cement-substance.

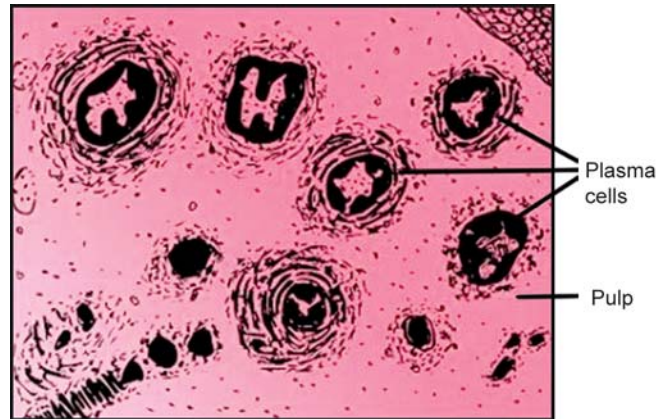


Figure 6.28: The clusters of plasma cells are seen during inflammation of the pulp

Fibers

Two types of fibers are present in the dental pulp.

I. Precollagenous or reticular or argyrophilic or Korff's fibers

II. Collagenous fibers

In the early stage of development, the fibers are mostly precollagenous in nature. With advancing age they become more collagenous. The collagen fibers of the pulp have typical cross-striations called banding at 64 nanometers, characteristic of collagen, and range in length from 10 to 100 nanometers or more (Figs 6.36 and 6.37).

The diameter of pulp fibers ranges from 10 to 12 nanometers (nm). The collagen fibers may appear in

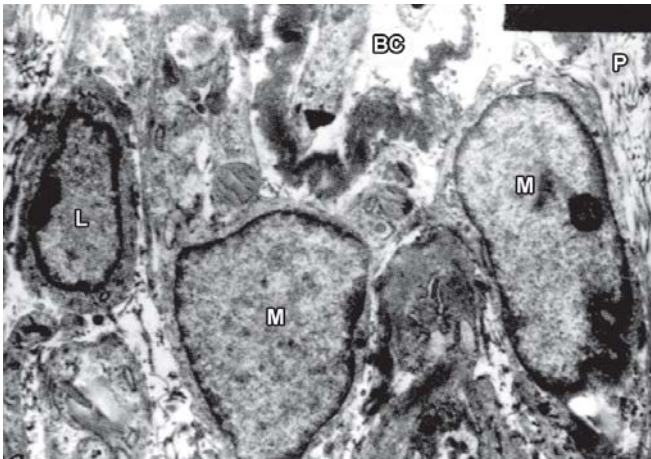


Figure 6.29: Inflammatory cell aggregate of lymphocyte (L) and macrophages (M). BC= Blood capillary P= Pulp. Lymphocytes have scanty cytoplasm and very few organelles in the cytoplasm. Macrophages show nuclear membrane folding, presence of mitochondria and lysosomes in the cytoplasm. (Electron micrograph x 3800)

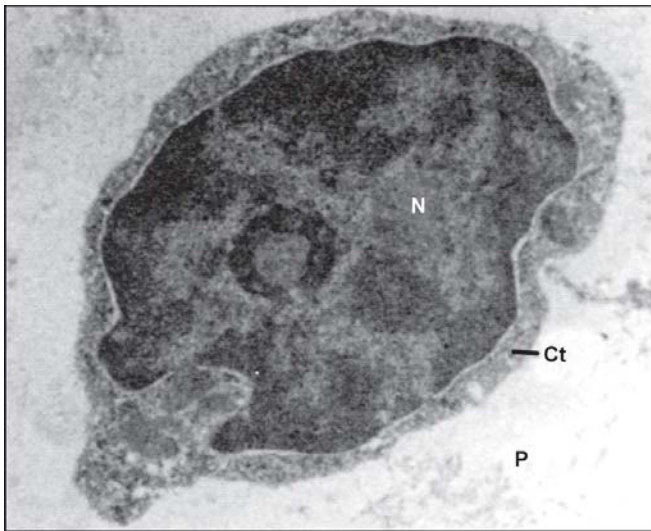


Figure 6.30: Electromicrograph of lymphocyte located in pulp (P). Cytoplasm (Ct) poor in organelles forms narrow rim around large oval to round dense nucleus (N) x 12,000

bundles or they may appear scattered throughout the pulp. These are known as bundle collagen or diffuse collagen. The collagen fibers found in the pulp are basically type I and type III in a 55 : 45 ratio. The collagen fiber bundles are mostly found in the apical region of root canal.

Ground Substance

The ground substance is composed of mucopolysaccharides, glycosaminoglycans, glycoproteins and



Figure 6.31: Electronmicrograph of macrophage showing highly lobulated nucleus (N) and abundant amount of cytoplasm (CT) rich in organelles P= Pulp x 8000

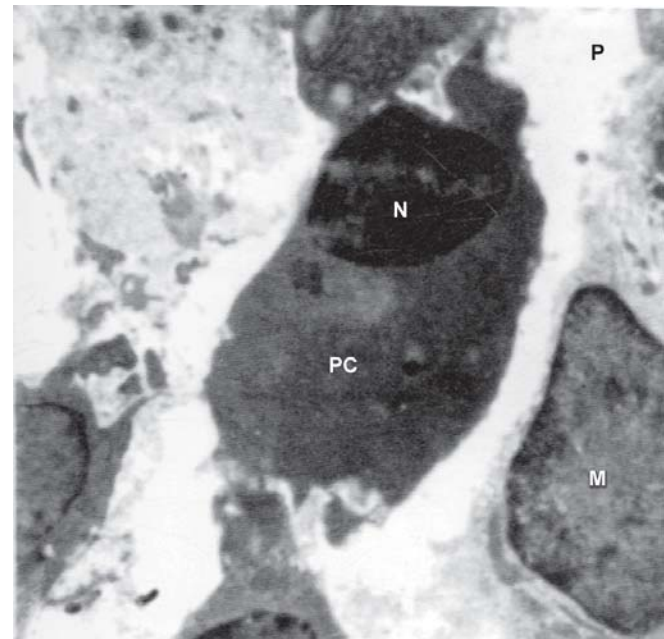


Figure 6.32: Plasma Cell (PC): Nucleus (N) is eccentric and shows nuclear chromatin condensations at the periphery giving cart wheel appearance P = Pulp M = Macrophage. There is slight perinuclear clearing of the cytoplasm (Electron micrograph x 2500)

water. Chondroitin A, chondroitin B and hyaluronic acids are also present in the ground substance. Their appearance varies from finely granular to fibrillar or denser in some area. The intercellular substance supports the cells and acts as a medium for nutrient transport from the vasculature to the cells, as well as for transport of metabolites from cells to blood vessels. Ground substances also contains hyaluronan, versican, syndecan, tenascin, fibronectin, laminin and intergrins.

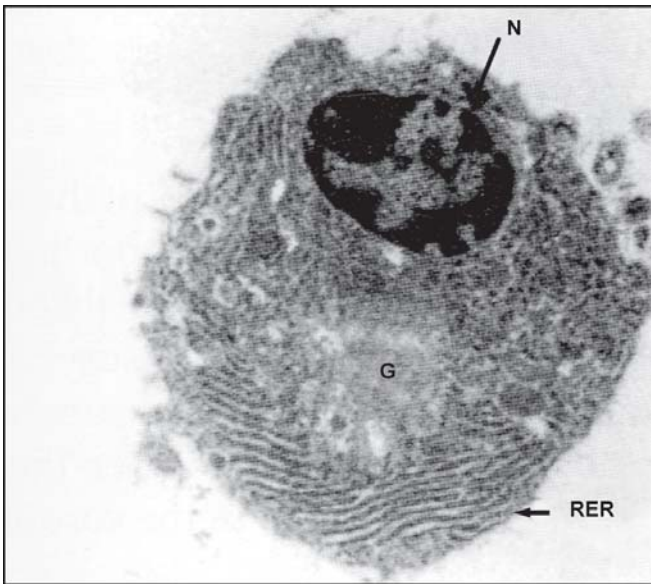


Figure 6.33: Plasma cell showing dense chromatin arranged at the periphery of eccentrically located nucleus (N). Cytoplasm shows stacks of rough endoplasmic reticulum (RER) at the periphery and ill-defined saccules near nucleus representing Golgi apparatus (G) x 10,000

venous system having thin walls and large lumen is seen in the pulp. The veins carry the blood away from the pulp. The blood capillaries that appear in the pulp form a loop close to the odontoblast and form a plexus in odontoblastic region (Figs 6.39 and 6.40 A and B). From this plexus, branches pass between the odontoblasts and reach towards the predentin.

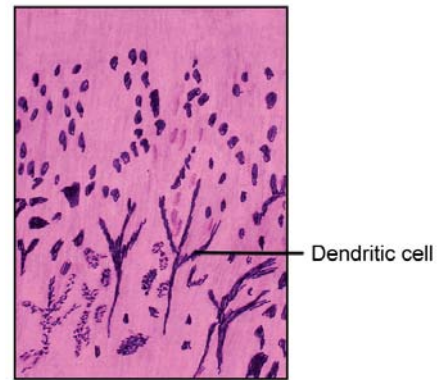


Figure 6.35: Dendritic cells (Melanocytes) present in odontogenic zone of the pulp

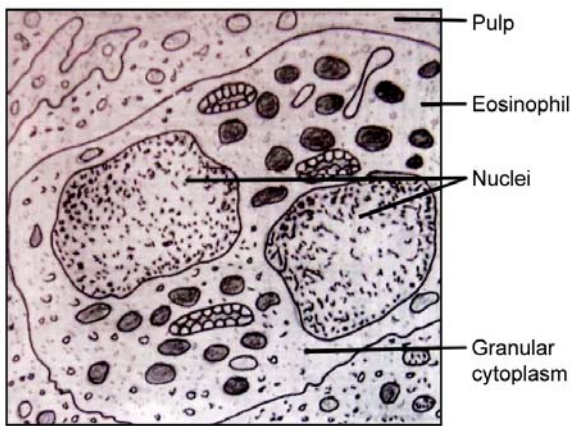


Figure 6.34: Eosinophil in extravascular location in pulp organ (as seen under Electron Microscope)

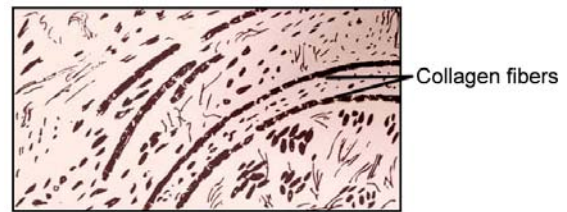


Figure 6.36: The collagen fibers in the pulp exhibit typical cross striation at sixty-four nanometers

Blood Vessels

The blood vessels enter into the pulp through apical foramen. Sometimes accessory blood vessels also enter pulp chamber through the accessory foramen. Pulp chamber is occupied by pulp, which is rich in blood supply (Fig. 6.38). The blood vessels of both the pulp and periodontium arise from the superior or inferior alveolar artery. An extensive

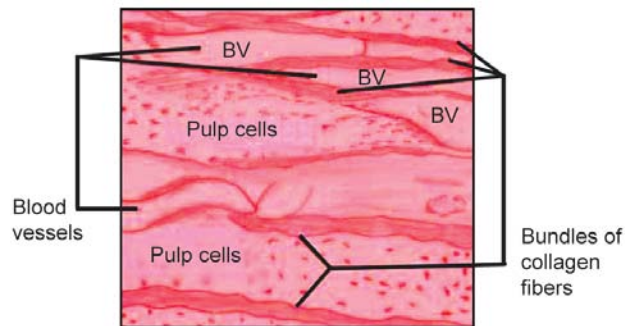


Figure 6.37: Blood vessels (BV) of pulp are surrounded by bundles of collagen fibers

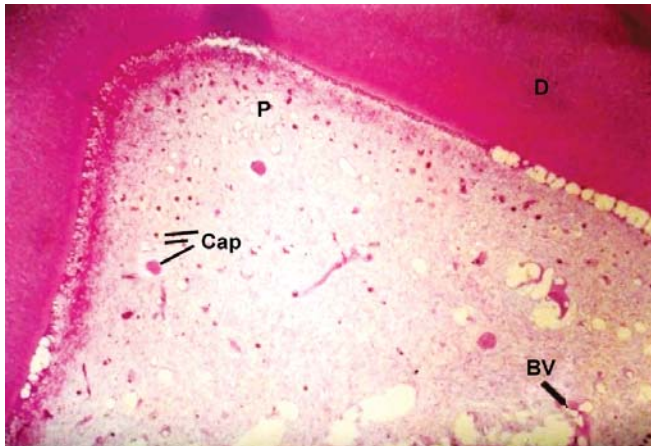


Figure 6.38: The peripheral region of the pulp (P) is rich in minute capillaries (Cap). Larger capillaries and blood vessels (BV) can be seen in the central region of the pulp. (H & E x 100)

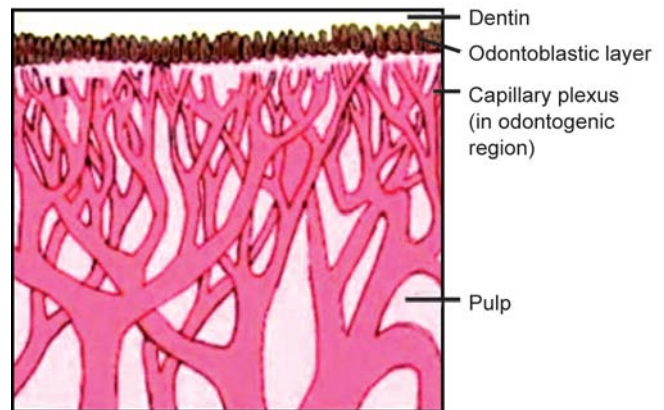


Figure 6.40A: Capillary plexus in the odontogenic region

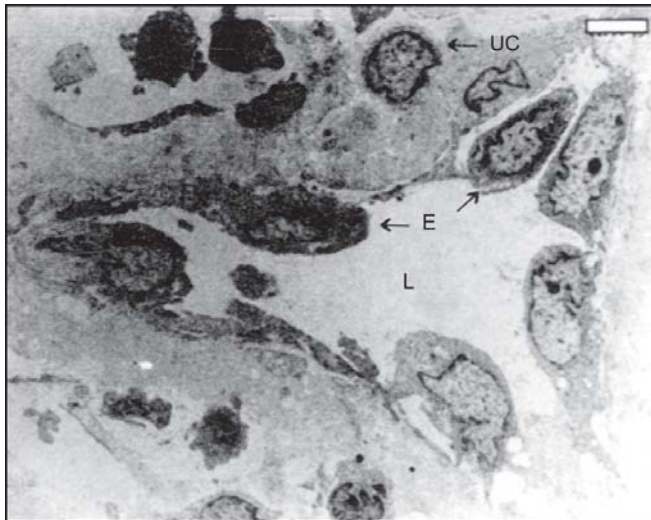


Figure 6.39: Capillary: Lumen (L) of the capillary is bounded by endothelial cells (E). Undifferentiated cell (UC) can be seen in the outer layer of the periphery (x 4600)

The arterial supply consists of arterioles which enter through apical foramen (sometimes through accessory foramina also) and divide into finer branches [terminal arterioles, metarterioles, precapillaries] and finally into capillaries at the arterial end. The capillaries form an extensive network in the cell poor zone, just central to odontoblastic layer. From this plexus arise small knob like capillary loops which extend between the odontoblasts in odontoblastic region and reach towards the predentinal (Figs 6.40 and 6.41 A and B). The capillaries

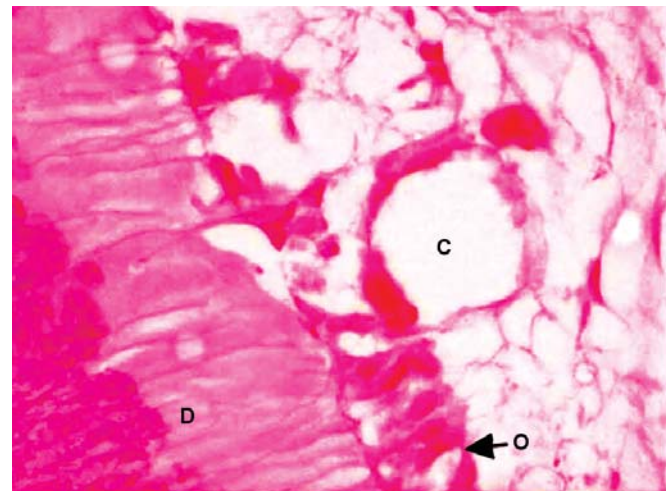


Figure 6.40 B: Pulp capillaries (C) come in close contact with odontoblasts (O) during active phase of dentin laying. Dentin (D) x 400

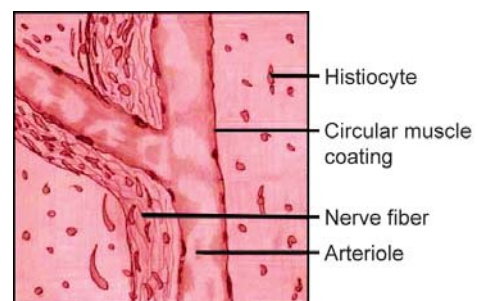


Figure 6.41: Pulp showing branching arteriole and nerve fibre

on the venous end unite to form post-capillary venules and collecting venules, which in turn unite to form a few venules finally exiting through the apical foramen.

The communication of the vessels of the pulp with the periodontium is important for its clinical significance, in the event of a pathologic condition in either the periodontium or the pulp. The branches of the alveolar arteries supply both the tooth and its supporting tissue. The periodontal vessels enter the pulp through the apical foramen and sometimes the accessory foramina. These are thinner walled than those that surround the tooth. Small arterioles enter the apical foramen and divide and redivide in the coronal pulp (Fig. 6.41).

The blood flow in arterioles is more rapid, approximately 0.3 to 1 mm / second, in venules it is 0.15 mm / second, and in capillaries about 0.08 mm / second. The largest diameter of arterioles in the pulp is 50 to 100 micrometers. The blood vessels have three layers. The first layer consists of squamous or cuboidal endothelial cells surrounded by basal lamina and is known as the tunica intima. The second layer consists of several layers of smooth muscle cells and is known as tunica media. Sometimes endothelial cell wall is in contact with the muscle cells. This is called as myoendothelial junction.

In central part of the pulp, arterioles have a thicker layer of muscle cells (Fig. 6.42) while at the periphery of pulp, small arteriole show thin layers (Fig. 6.43).

The outer or third layer consists of a loose network of collagen fibers around the large arteries and is known as the tunica adventitia. It mixes with the fibers of the surrounding intercellular tissue. The endothelial cells of

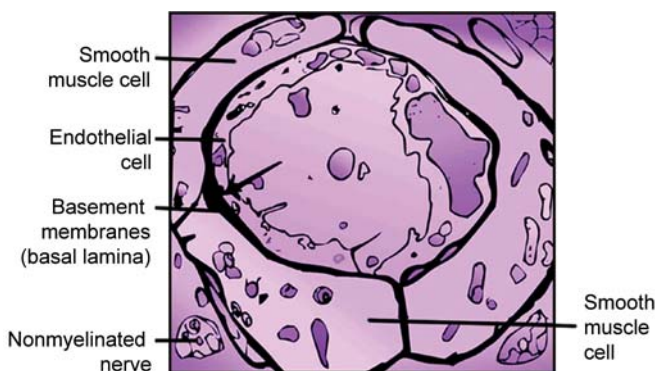


Figure 6.42: In central part of pulp arterioles have thicker layer of muscle cells and dense basement membrane interspersed between endothelial and muscle cells (arrow)



Figure 6.43: Small arteriole at periphery of pulp showing two thin layers of smooth cells which surround the endothelial cell lining of vessel

these vessels contain numerous micropinocytotic vesicles, which are responsible for transendothelial movement of fluid. Pericytes are fibroblasts associated with capillaries and occasionally lie on the surface of the vessels. Fine blood vessels are seen in the peripheral layer of pulp under cell rich zone (Figs 6.44 to 6.46).

In the radicular pulp, veins and venules are larger than the arteries. Venous-venous anastomosis and arteriovenous anastomosis appear in the coronal pulp.

The arteriovenous shunts play an important role in the regulation of flow of pulpal blood. The blood capillaries present in dental pulp appear as endothelium-lined tubes. They measure 8 to 10 micrometers in diameter. These fenestrated capillaries are responsible for rapid transport of metabolites in the process of dentinal matrix formation. Both continuous and fenestrated capillaries are responsible for rapid transport of metabolites and in the process of dentinal matrix formation. Both continuous and fenestrated terminal capillaries are found in the odontogenic region.

Lymph Vessels

The lymphatic vessels are seen as small, thin walled endothelium lined tubes that join lymph venules in the central region of the pulp. They pass through the apical foramen as a single or two large lymph vessels (Fig. 6.47). They are characterized by the presence of lymphocytes and absence of red blood cells. The lymphatic vessels have discontinuities in their walls. Lymph vessels drain the periodontal ligament and pulpal tissue of the anterior



Figure 6.44: Blood vessels and nerves as seen in the pulp along the surface of dentin

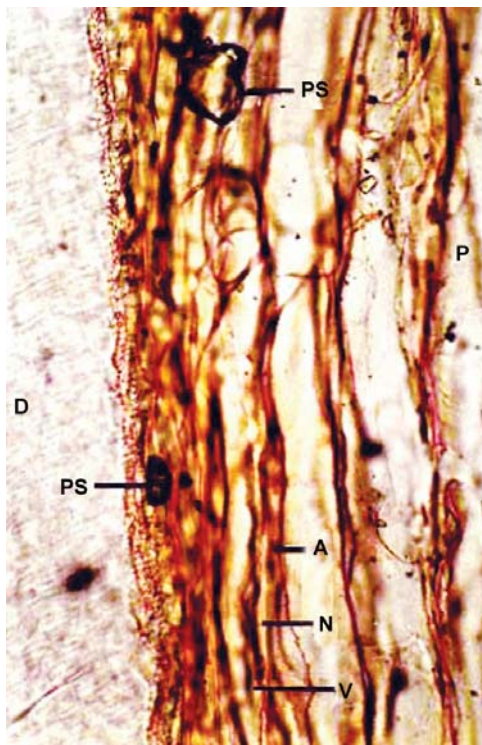


Figure 6.45: Ground section of tooth showing higher vascularity and innervation of pulp (P) at the periphery near dentin (D). A = Arteriole, V = Vessels, N= Nerve, PS= Pulp stone x 200

and posterior teeth to the submental, submandibular and deep cervical lymph nodes respectively. The presence of lymph vessels in the pulp is questioned by some investigators and agreed upon by other investigators.

Nerves

The dental pulp has abundant nerve fibers along with blood vessels. Both the myelinated and non-myelinated

nerves are found in pulp (Fig. 6.48). The nerve bundles enter through the apical foramen and follow the course of blood vessels and travel along the central core of the pulp. The number of fibers in these bundles varies from 150 to more than 1200. Majority of fibers in these bundles are smaller than 4 microns.

The nerve fibers divide and redivide into smaller branches as they migrate coronally, they lose their myelin sheath and form a rich plexus in the cell free zone. This is known as the parietal plexus or plexus of Raschkow or the circumodontoblastic plexus. Both myelinated axons and minute non-myelinated fibers make up this layer of nerves. From this plexus terminal branches are given off and form a plexus around the odontoblasts, and some nerve endings are also seen in the dentinal tubules. In this circumodonto-blastic plexus, capillaries are also present. The endothelial lining of these capillaries is occasionally surrounded by pericytes, which are also known as the capillary associated fibroblasts (Figs 6.49 to 6.54).

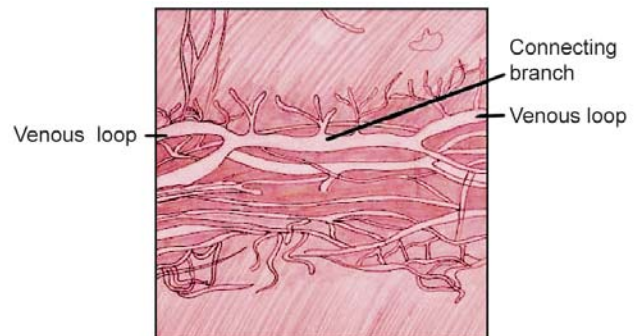


Figure 6.46: In the central pulp venous loops are present. Such loops are seen at the left and right with a connecting branch at the center (as seen under scanning Electron Microscope)

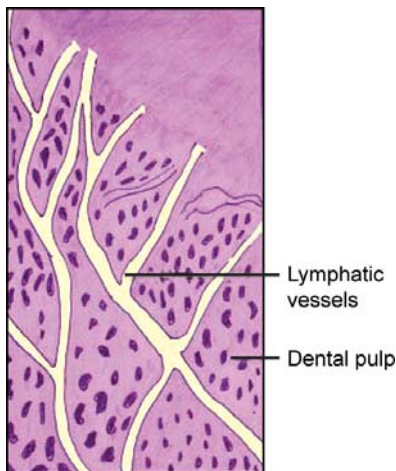


Figure 6.47: The presence of thin-walled lymphatic vessels in the dental pulp that exit through the apical foramen

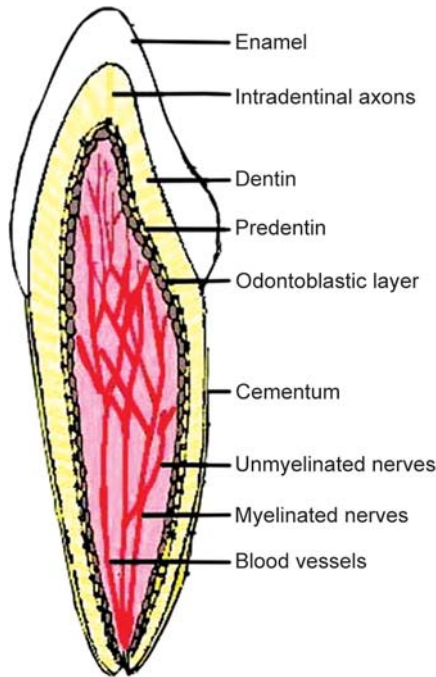


Figure 6.48: Both myelinated and nonmyelinated nerves and blood vessels are found in the pulp

The majority of the nerves of the pulp are non-myelinated and sympathetic in nature. The nerves are found in close association with the blood vessels. The nerve fibers terminate on the muscle layer of the larger vessels for regulating the lumen of the vessels (vasoconstriction and vasodilatation). The non-myelinated nerve fibers are also known as Remak's fibers.

The nerve terminals consisting of oval and round enlargements contain microvesicles, small, dark, granular bodies and mitochondria.

Myelinated (sensory) nerve fibers enter the pulp and mediate the sensation of pain, which may be caused by external stimuli. They cannot differentiate sensory response between heat, cold, touch, pressure or chemicals in the pulp. This is because the pulp organs lack those specific types of receptors that distinguish these types of stimuli. The parietal layer of nerves develop gradually and becomes prominent when root formation is completed.

The nerve axons of non-myelinated nerve fibers are enclosed in a Schwann cell covering. Most nerve endings are found in the pulp horns than in other areas of the coronal pulp. The types of neurotransmitters such as substance P, vasoactive intestinal peptide, 5-hydroxytryptamine, prostaglandins, acetylcholine, somatostatin and norepinephrine are present in the dental pulp. Many of these cause changes in vascular tone, thereby affecting the incremental growth of dentin indirectly by influencing the rate of blood flow.

Nerve endings – Nerve endings are located close to the odontoblast and contain small, dark granular bodies, mitochondria and microvesicles. Mature deciduous teeth contain more nerve endings near the odontoblast layer with few penetrating into the dentin.

FUNCTIONS OF THE PULP

The pulp is a soft vital tissue of the tooth that performs following functions. Important among them are as follows.

Inductive: (a) The basic function of the pulp primordium is to interact with the cells of oral epithelium. This interaction leads to the formation of dental lamina and enamel organ. (b) To determine the shape of each tooth the pulp primordium interacts with the developing enamel organ. (c) Therefore in the development of the tooth the pulp precursor has an important role to play.

Formative: (a) The function of pulp is the formation of dentin organic matrix, which surrounds and protects the pulp. (b) By the development of odontoblastic process the dentin is formed at the pulp-predentin border and along the tubule wall.

Nutritive: The dentin is nourished by the pulp through odontoblasts and their processes and by rich vascularity of the pulp.

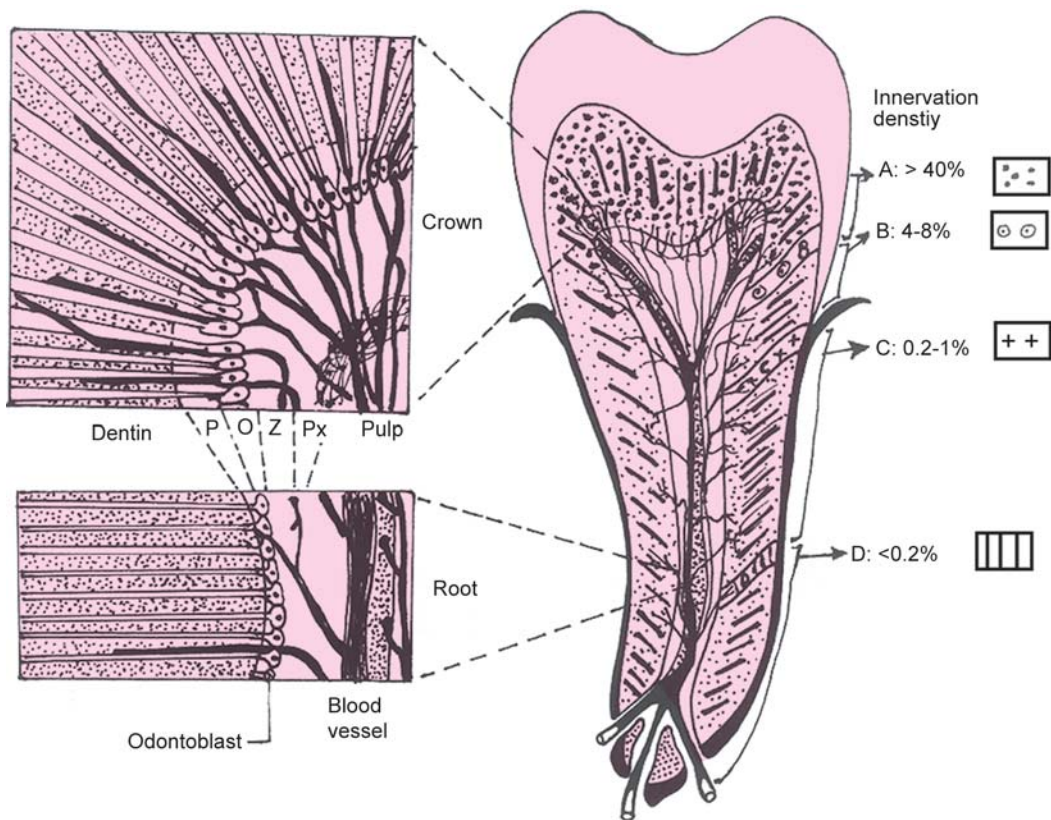


Figure 6.49: Innervation density at different areas of tooth - 'A' is magnified view of crown portion and 'B' is magnified view of root portion. P-Pre-dentin, O-Odontoblast layer, Z-Cell Free Zone, Px-Neural Plexus

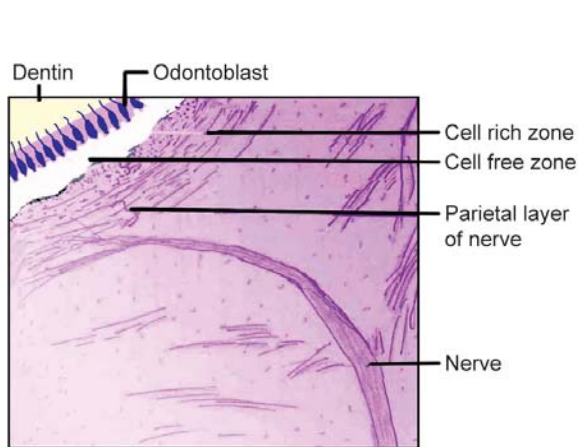


Figure 6.50: In pulp, nerve branch pass to parietal layer, which lies adjacent to cell rich zone

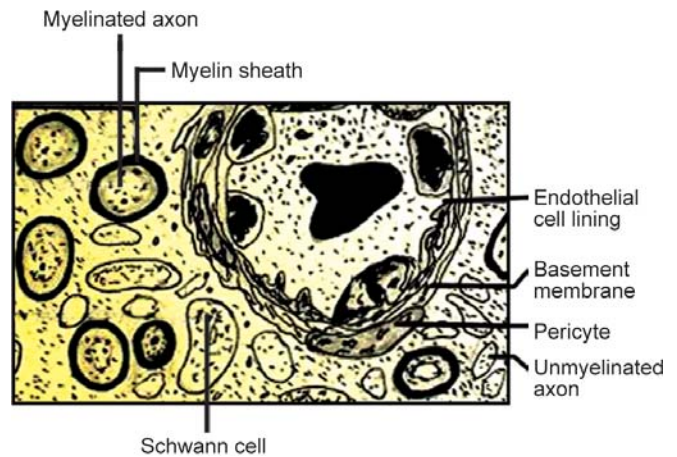


Figure 6.51: Both myelinated and unmyelinated axons lie adjacent to capillary. Endothelial cell lining surrounded by basement membrane and occasional pericytes

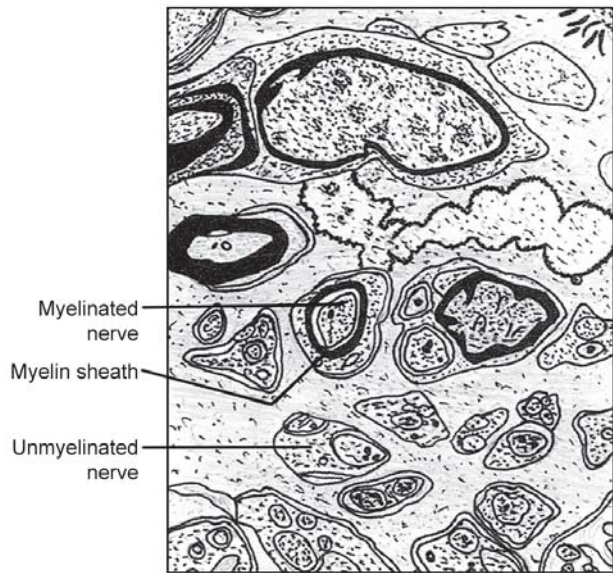


Figure 6.52: Cross-section of pulp. Both myelinated and unmyelinated nerves are present (as seen under Electron microscope)

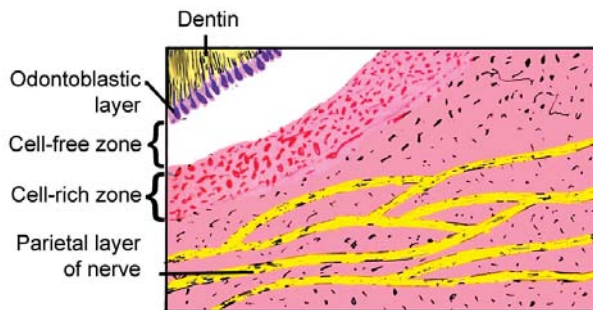


Figure 6.53: Parietal layer of nerve is made up of myelinated nerve fibers located adjacent to the cell rich zone

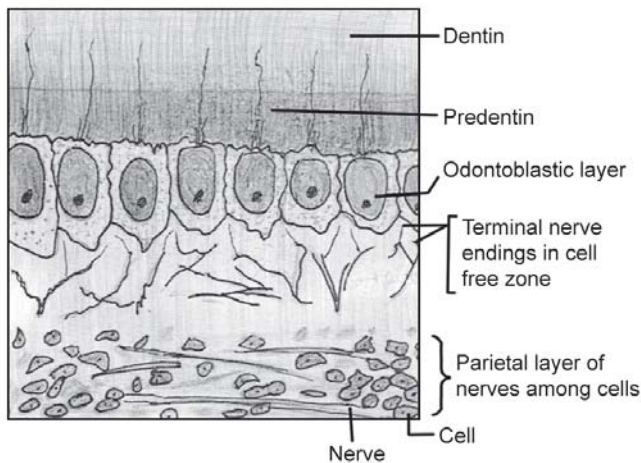


Figure 6.54: Dentinopulpal junction showing terminal nerve endings present among odontoblasts that arise from subjacent parietal layer (as seen under Electron Microscope)

Protective or Sensory: The sensory nerves present in the tooth respond to stimuli such as heat, cold, pressure, chemical agents and operative procedures by producing pain. Thereafter the preventive measures are initiated to prevent damage to the teeth. The sympathetic nerves initiate reflexes that control blood circulation in the pulp. This is done by stimulating the visceral motor fibers ending on the muscles of blood capillaries.

Reparative or Defensive (a) The pulp has very good reparative properties. (b) The pulp organ responds to all types of irritation, like bacterial, mechanical, thermal and chemical, by producing reparative dentin and the mineralization of the affected dentinal tubules. (c) These irritations may be cutting of dentinal tubules and placement of irritating dressing or filling material. (d) The pulp has macrophages, neutrophils, monocytes, lymphocytes, mast cells and plasma cells, all of which help in the repair process of the pulp. (e) When irritation is mild, hyperemia will be mild, leading to repair. It is called reversible hyperemia (Fig. 6.55). (f) If the inflammation is mild in nature, the pulp will heal because it has good regenerative capacity (Figs 6.56 to 6.58). (g) Toward off the source of irritation, reparative dentin is formed in pulp and calcification of the tubules, that is

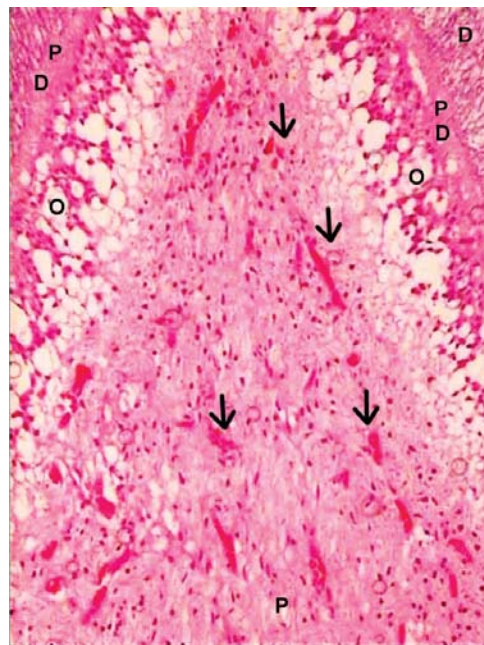


Figure 6.55: Marked hyperemia of pulp in very early infection. Capillaries are packed with red cells (shown by arrow). Pulp (P) Odontoblasts (O) Dentin (D) Predentin (PD). x 150

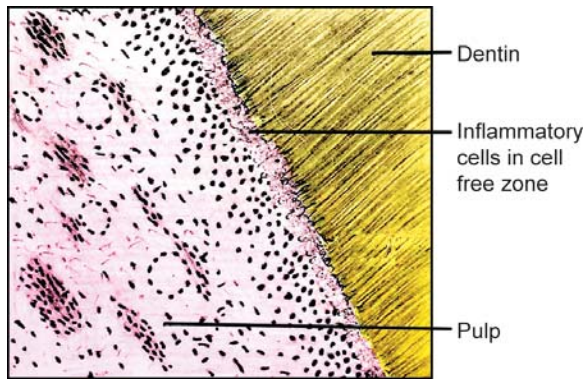


Figure 6.56: Mild inflammatory response of pulp is characterized by loss of odontoblast identity and inflammatory cells obliterating the cell free zone

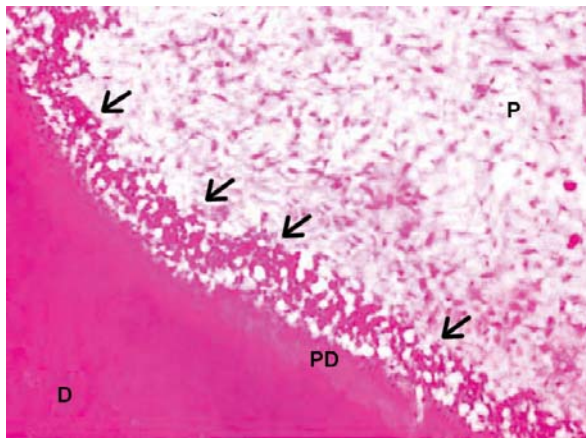


Figure 6.57: Mild inflammation of pulp (P) showing increased cellularity of the cell rich zone and presence of inflammatory cells indicated by arrows (\leftarrow). The zones of peripheral pulp are obliterated. D= Dentin, PD= Pre-dentin (H & E x 100)

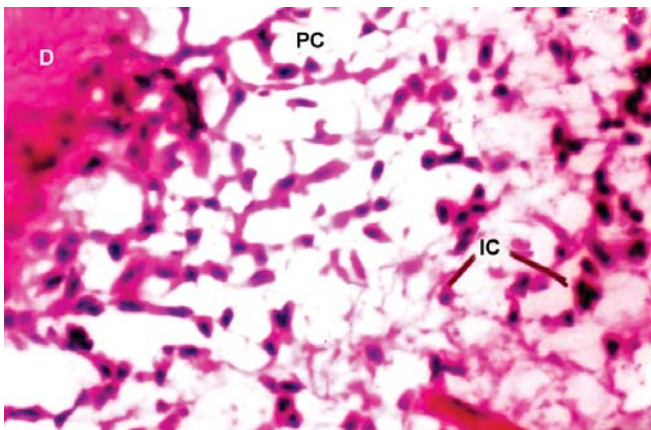


Figure 6.58: Pulp showing mild inflammation. The odontoblastic zone near dentin (D) is obliterated and pulp chamber (PC) is infiltrated by inflammatory cells (IC). (H & E x 200)

sclerosis, occurs (Figs. 6.59 and 6.60). (h) These are attempts of the pulp to isolate and wall off itself from the advancing source of any type of irritation. (i) The strong, rigid wall of dentin around pulp protects it from heavy masticatory forces, trauma and all other types of irritations. Sometimes this rigid wall may prove dangerous and fatal to the pulp. During severe irritation and inflammation there is accumulation of excess fluid due to hyperemia and increased permeability leads to edema of the pulp. When repair is not possible it is called irreversible hyperemia. This excess fluid of irreversible hyperemia is surrounded by unyielding rigid dentinal walls. This results into excessive pressure on the apical vessels leading to ischemia and finally the necrosis of the pulp.

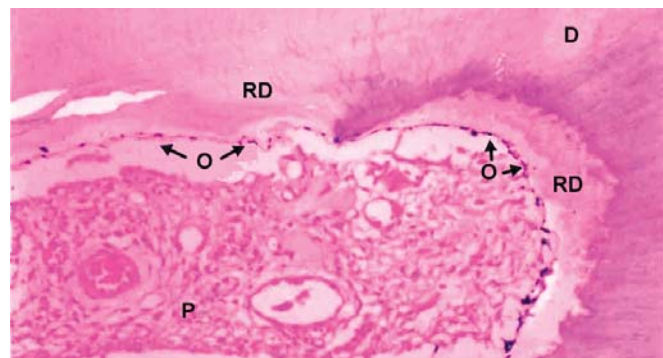


Figure 6.59: Laying down of reparative dentin (RD) in response to inflammation of the pulp (P). Pulp shows dilated blood vessels indicating hyperemia. Odontoblasts (O) become prominent D= Dentin. (H & E x 150)

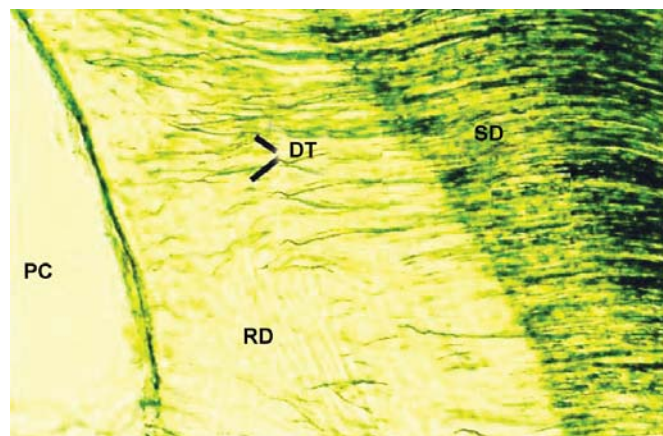


Figure 6.60: Ground section showing reparative dentin (RD) formation. The dentinal tubules (DT) are sparse and haphazardly arranged. PC= Pulp chamber, SD= Secondary dentin x 200

PULP INJURY AND REPAIR

Molecular events in pulp repair are as follows.

In mild pulpal injury, molecules like kinesin, actin and myosin produce dense skeletal network adjacent to the wound site and nestin, fibronectin and vimetin reestablish the normal architecture.

Cell adhesion molecules like N-cadherin help in cell adhesion, cell recognition, control of cell division, migration and differentiation. Factors like transforming growth factor, fibroblast growth factor, bone morphogenetic protein and insulin like growth factor initiate angiogenesis, migrate the odontoblastic process and recruit inflammatory cells. Injured cells release the calcium which helps in migration of cell and regulation of cytoskeletal organization.

DEVELOPMENT OF PULP

The area of ectomesenchymal condensation immediately subjacent to the enamel organ is known as dental papilla, which is primitive pulp. The initial stages of tooth development are controlled by dental papilla. It is due to proliferating future papilla, the oral epithelium invaginates and enamel organ is formed. Gradually the enamel organ grows and dental papilla is enclosed in their central portion. The outer shape of the dental papilla determines the future shape of the tooth that is whether the forming enamel organ is to be a canine or a molar. Recently it has been demonstrated that it is the epithelium that guides the shape of the future tooth.

In the region of deciduous incisors, the initiation of development of dental pulp begins at the eighth week of embryonic life. This is gradually followed by posterior teeth. Due to proliferation of cells and blood capillaries in the dental papilla, the cell density and vascularity are increased. The young papilla become highly vascularized with well organized network of capillaries before the odontoblasts begin laying down the dentin matrix. The cells of the dental papilla are called pulp after dentin forms around them. After the formation of ameloblasts from the inner enamel epithelium, the odontoblasts then form from the outer or peripheral cells of the dental papilla. The odontoblast is responsible for production of dentin. As this occurs, the dental papilla is now designated as the pulp organ.

Undifferentiated mesenchymal cells are present in the dental papilla. After formation of the pulp, these cells differentiate into stellate shaped-fibroblasts.

Vascularization of the developing pulp starts during the bell stage, with small branches arising from principal vascular trunks of the jaw. The numerous small branches form a bed of venules, arterioles and capillaries in the subodontoblast and odontoblast layer. The vascularity of the odontoblast layer increases as dentin is progressively laid down. The nerves are present close to the tooth germ from the very earliest stage of its development. The first fibers to enter the developing pulp are sensory, and the sympathetic nerve fibers follow later. A large number of nerves enter the pulp prior to root formation. The plexus of Raschkow is not established until root formation is complete. Nerve fibers in the dental follicle is first seen in eleventh week of intrauterine life and in the dental papilla in the eighteenth week of intrauterine life. Nerve fibres reach the subodontoblastic region in the twenty fourth week. Dental pulp cells produce nerve growth factor and Semaphorin 7A during the development.

PULP ORGANS

Deciduous (Primary) Pulp Organ

The deciduous pulp organ functions in the oral cavity for a shorter duration than do the permanent pulp organs. It functions for only about 8 years and 3 months duration. The length of this duration may be divided into the following three time periods:

- a. *Period of growth of pulp organ*: It is the time period from the beginning of root formation to root completion. Usually, in about one year's time, the crown and roots of the teeth develop.
- b. *Period of maturation of pulp*: It is the time period after root formation is completed until root resorption begins. The maturation of pulp takes place in this period of time. The root is completed and resorption of root begins at about 3 years and 9 months of age.
- c. *Regression of pulp*: It is the period of time starting from the beginning of root resorption until the time of exfoliation of tooth. It usually lasts for about 3 years and 6 months. The maximum life of primary pulp organs in the oral cavity including growth, maturation and regression periods of pulp, is approximately 9 years and 6 months. The period of regression of the deciduous radicular pulp depends on the time from the completion of the permanent crown till the time of permanent tooth eruption.

Permanent Pulp Organs

Pulps of deciduous and permanent teeth are morphologically almost identical during formation of crown. The development of permanent pulp organs requires longer period of time than that for the development of pulp organ in the deciduous teeth. The period of time for formation of complete crown and its calcification is about 5 years and 5 months. During this period, the organs are highly cellular and show high mitotic rate, particularly in cervical region. The pulp is highly vascular. The pulps of both deciduous and permanent teeth are almost alike in structure, like cell-free zone, cell-rich zone, parietal layer containing thin and fine nerves and blood capillaries at the periphery and central pulp containing slightly thicker nerves blood vessels.

The developmental processes take much longer time in permanent teeth than in deciduous teeth. The period of time of crown completion to eruption of teeth in upper and lower arches is about 3 years, 6 months. The period of time from eruption of teeth to completion of root on an average is three years, eleven months.

The average length of time for the development of permanent pulp organ is about 12 years, 4 months, which is based on the length of time from prenatal crown formation to postnatal root formation. The time taken for

completion of the root of permanent tooth is longer on an average is 7 years, 5 months, than in the deciduous teeth, where the average is 3 years, 3 months. In the deciduous teeth, the period of aging of pulp is much faster than in the permanent teeth. In permanent teeth, the aging of pulp takes full adult life span (about 40 years), while in deciduous teeth it takes time from root completion to exfoliation (about 7 years and 5 months).

The mandibular arch requires shorter period of time to complete each process of development for both the primary and permanent pulp organs in comparison with the maxillary arch.

AGE OR REGRESSIVE CHANGES IN PULP

The change from developmental tissue (the dental papilla) to the mature tissue (the dental pulp) is gradual. During the developmental stages the synthetic activity of the odontoblasts and fibroblasts is high. The cells are larger with prominent organelles associated with protein synthesis. As age advances continuous deposition of dentin by odontoblasts causes reduction in pulp volume (Figs 6.61 to 6.63). The pulpal cells gradually decrease in number. The sensitivity of pulp is reduced with age due to degeneration and loss of unmyelinated and myelinated axons. Due to resorption and deposition of cementum, position of apical foramen shifts (Fig. 6.64).

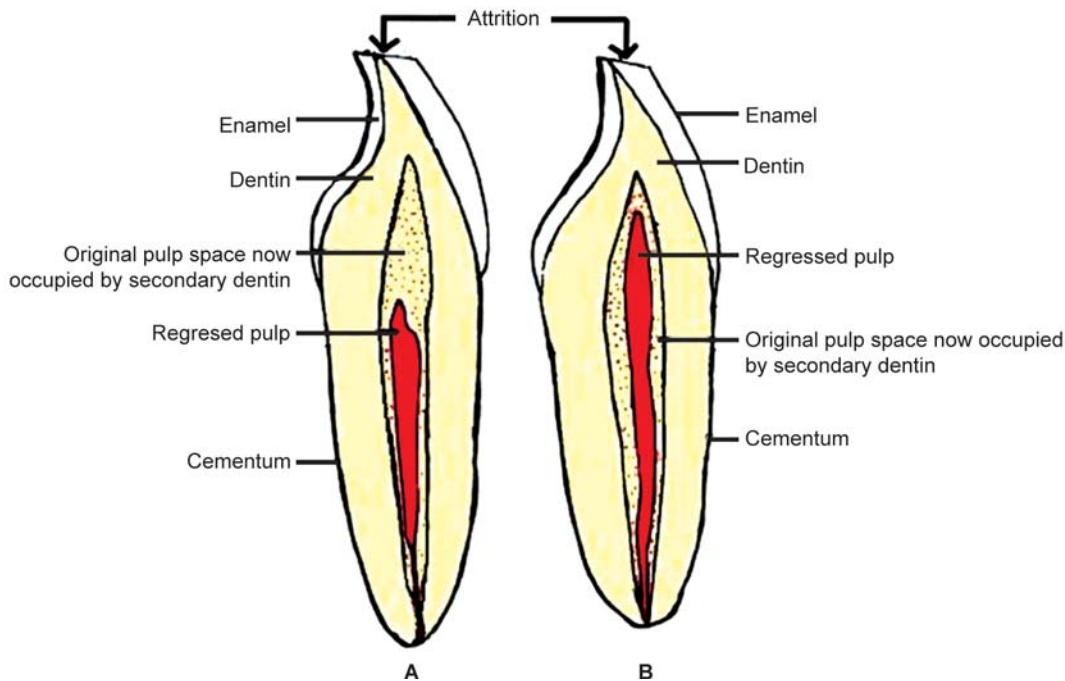


Figure 6.61: Regressive changes in pulp. About sixty percent of original pulp space is occupied by secondary dentin at the age of 60 years. In (A) secondary dentin shown by dots deposition is more in pulp chamber. In (B) secondary dentin deposition is uniform all around

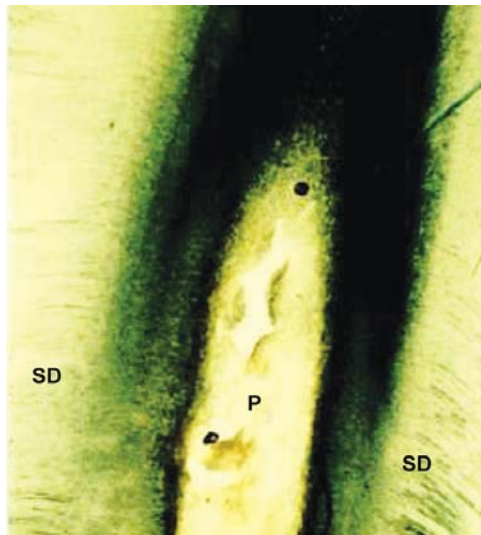


Figure 6.62: Ground section of tooth showing marked increase in secondary dentin (SD) formation and reduction in the size of pulp (P) x 100

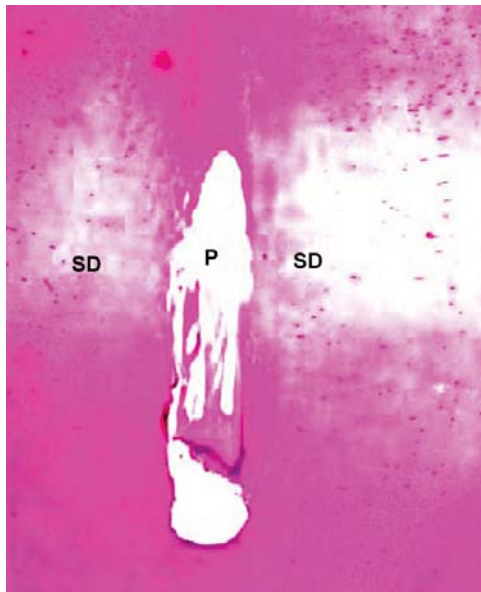


Figure 6.63: The pulpal tissue (P) is markedly reduced in amount due to large amount of secondary dentin (SD) formation. Decalcified tooth. (H & E x 100)

Pulp Stones (Denticles)

Pulp stones, or denticles are calcified, shiny nodular masses that occur either single or multiple in numbers within the coronal and radicular pulp. The pulp stones usually are asymptomatic in nature, but may be

symptomatic if the stones impinge on nerves or blood vessels. Usually they are present in teeth that appear normal in all other respects. They are present in embedded unerupted teeth or in functional teeth.

The pulp stones start as very small bodies. They may gradually grow and become large and impinge on nearby structures. There are reports in the literature that these tiny small innocent looking bodies, rarely, when impinge on the nerves may give rise to mild to severe neuralgic type of pain. The pain disappears after removing these pulp stones by pulpotomy or pulpectomy.

Occurrence: The incidence as well as size of pulp stones gradually increase with age. By radiological examination only large pulp stones are seen (Fig. 6.65). By histological examination most of the pulp stones, large and small almost all are seen. Therefore, they appear more prevalent by histological studies than radiological studies.

The average percentage of the presence of pulp stones of three histological studies according to the age groups is given in Table 6.2.

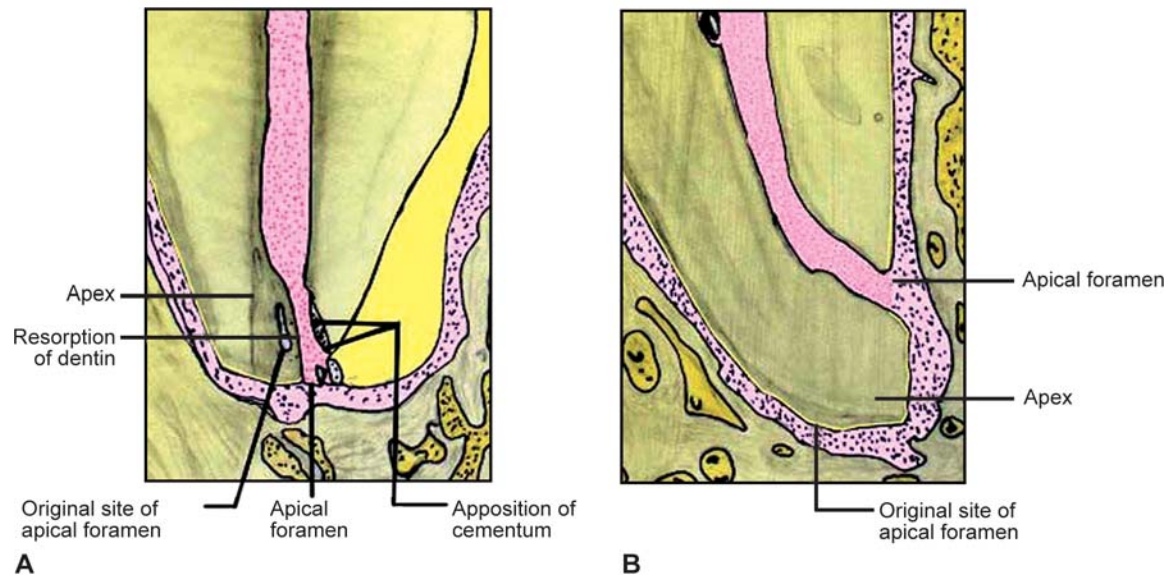
Table 6.2: The average incidence of pulp stones in teeth as observed by three radiological and histological studies in various age group of the persons

S.No.	Age group	Percentage of teeth having pulp stones as observed by	
		Histological studies	Radiological studies
1.	15 to 20	50	0.5
2.	20 to 30	60	1
3.	30 to 40	75	2
4.	40 to 50	85	4
5.	50 to 60	90	6
6.	60 and above	95	7

Pulp stones according to their histological structures, have been classified into two types— true and false (Fig. 6.66). They may be present in erupted, impacted and unerupted teeth.

True Pulp Stones

The structure of true pulp stones is similar to dentin, as they possess dentinal tubules and odontoblastic processes and may show surrounding odontoblasts. They are comparatively rare and are mostly located in the apical



Figures 6.64 A and B: Changes in location of apical foramen (A) Resorption of dentin and cementum occurring on one side and cementum is laid down on the opposite side resulting in relocation of apical foramen (B) Apical foramen is located on side of apex

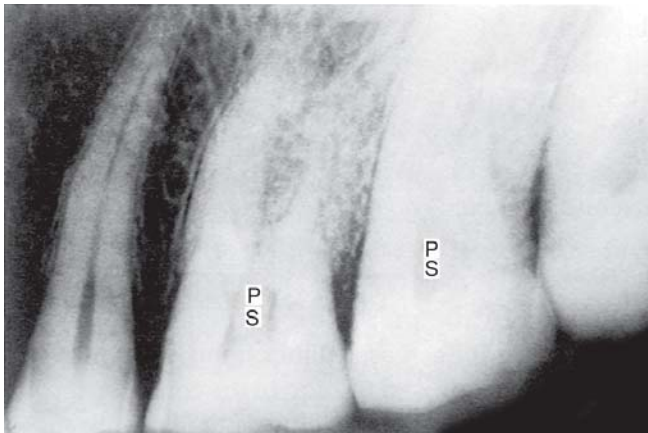


Figure 6.65: Radiograph showing first and second maxillary molars with large pulp stones (PS) in the pulp chambers

third of the root. True pulp stones are caused due to inclusions of the remnants of the epithelial root sheath present within the pulp. These epithelial remnants induce the mesenchymal cells of the pulp to differentiate into odontoblasts, which then secrete dentin mass, thereby forming true pulp stones (Figs 6.67 to 6.72).

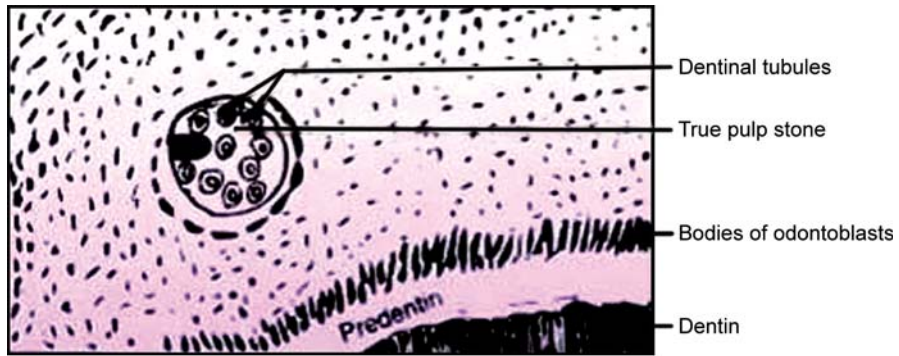
False Pulp Stones or Denticles

False pulp stones are calcified masses that do not exhibit dentinal tubules (Figs 6.66 and 6.73). They are seen as a

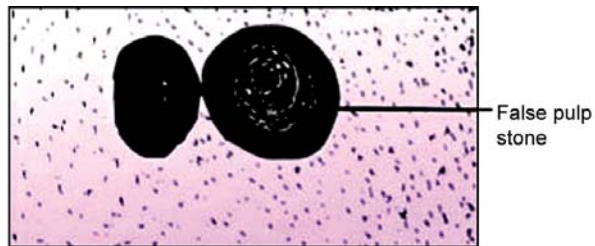
calcified mass having concentric layers of hard tissue. It is believed that usually these calcification sites appear within a bundle of collagen fibers very rarely small portion of the pulp tissue is also included in the center of the pulp stone (Figs 6.73 and 6.74). The calcified thrombi present in blood vessels may act as centers for inducing the formation of false pulp stones. Thrombi in blood vessels, when they get calcified, are called phleboliths. Sometimes false pulp stones arise around blood vessels. They consist of calcified remnants of necrotic cells. Beginning with small nodule, all denticles increase in size by incremental growth on their surface

Pulp stones, according to their relation with the dentin of the tooth, may be divided into three types—(a) free, (b) attached (c) embedded (Figs 6.75 and 6.76). The free pulp stones are completely surrounded by pulp. The attached pulp stones are partly surrounded by pulp and partly fused with dentin. The embedded pulp stones are completely surrounded by dentin. (Fig. 6.77) Embedded pulp stones are also called as interstitial pulp stones.

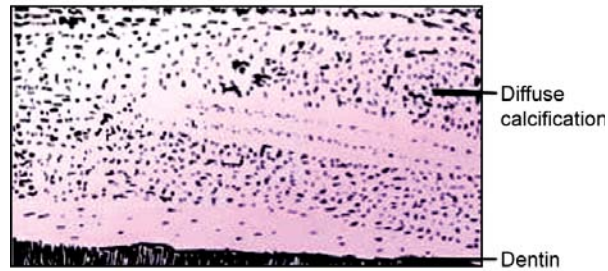
The pulp stones may appear close to nerve trunks and blood vessels of pulp (Figs 6.78 to 6.81). The incidence and the size of pulp stones increases as age advances. All pulp stones are formed free in the pulp and then afterwards become attached or embedded as secondary dentin formation progresses by the sides.



I (A) True pulp stone



I (B) False pulp stone



II - Diffuse calcification

Figure 6.66: Calcifications in the pulp. (I) Pulp stones (A) True, (B) False, (II) Diffuse calcifications

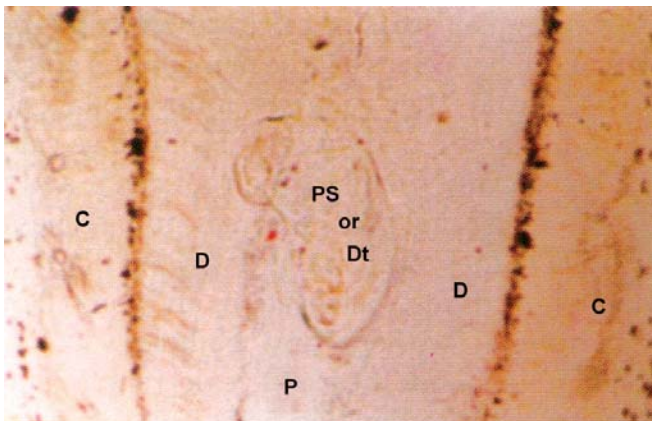


Figure 6.67: True Pulp Stone or Denticle (Dt) in the pulp (P) surrounded by dentin (D) and cementum (C) on both the sides. Dentinal tubules are seen in the pulp stone (PS) (Ground section x 200)

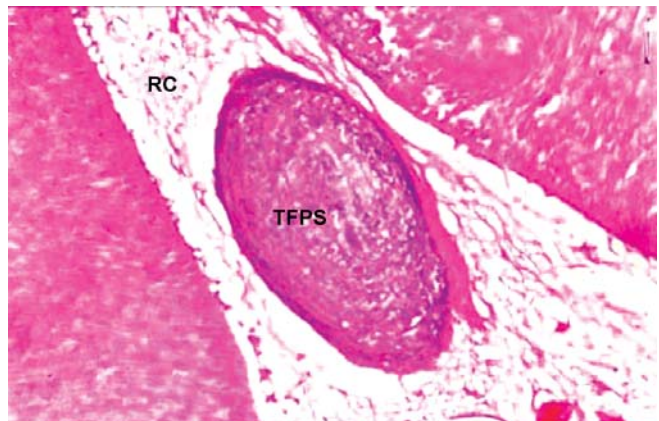


Figure 6.68: True free pulp stone (TFPS) in root canal (RC). (H & E x 100)

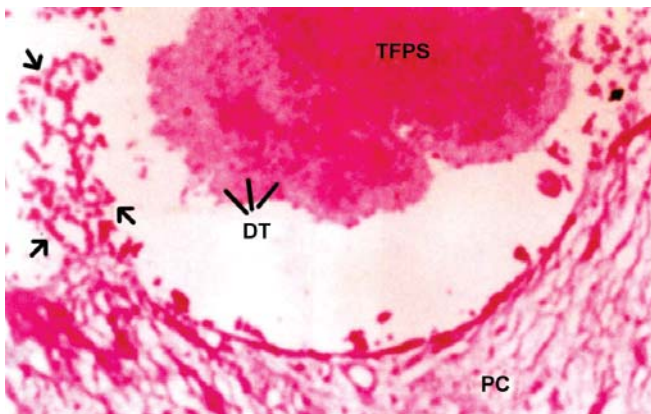


Figure 6.69: Dental tubules (DT) can be seen very clearly in True Free Pulp Stone (TFPS) lying in the pulp chamber (PC). Inflammatory cells indicated by arrows are present in pulp (H & E x 200)

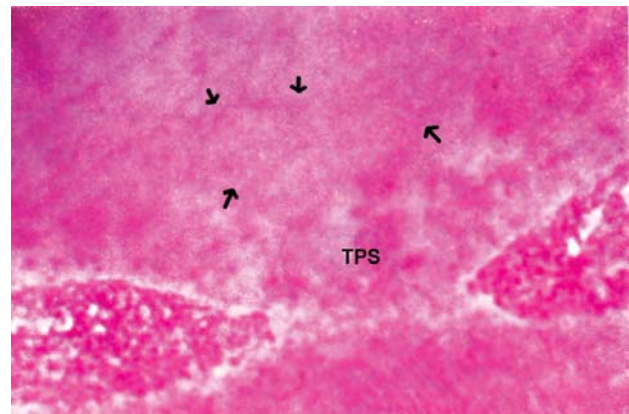


Figure 6.72: Higher magnification of Figure 6.72 showing dental tubules indicated by arrows in the True Pulp Stone (TPS) (H & E x 200)

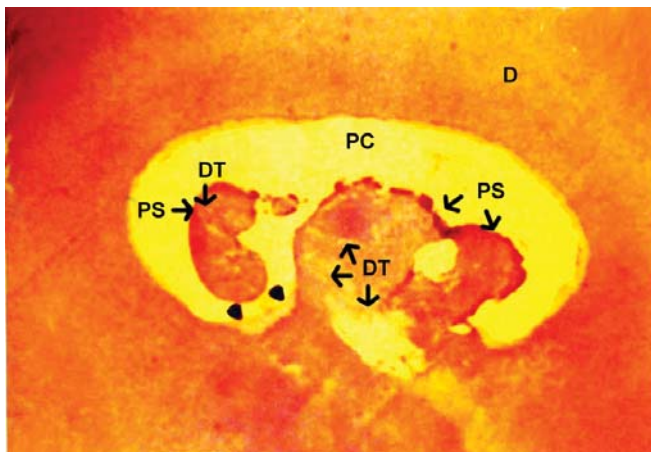


Figure 6.70: True pulp stone (Denticles): Decalcified section of tooth showing narrowing of root canal shown by arrow heads (▶) Dentin (D) is extending into the root canal as large round globular masses forming true pulp stones (PS). Tiny blue spots are dental tubules (DT). PC = Pulp chamber. (H & E x 200)

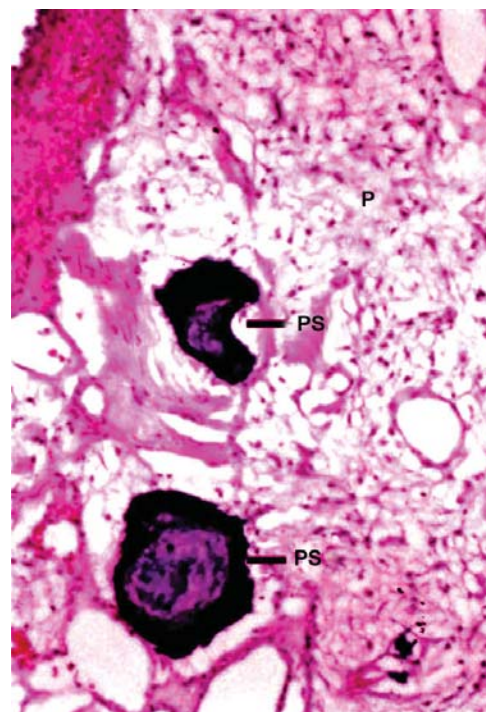


Figure 6.73: Free false pulp stones (PS): Two large, irregularly round deep blue deposits of calcium can be seen in the pulp chamber (P). Center of the stones is lighter than periphery indicating cell inclusion. Pulpitis is present at the periphery of pulp chamber where inflammatory cells have collected (H & E x 200)

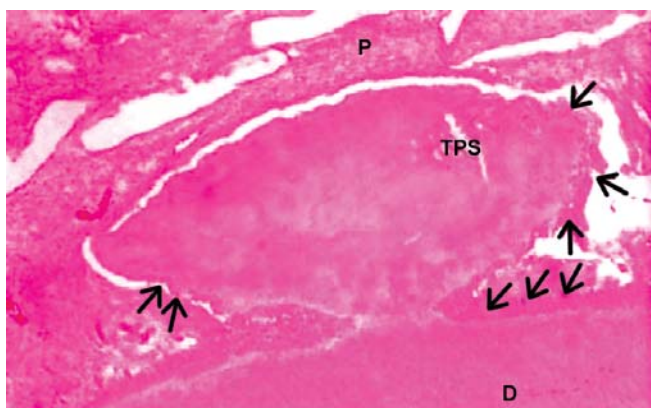


Figure 6.71: True pulp stone (TPS) or denticle seen attached to the dentin (D) and projecting into the pulp chamber (P) (H & E x 100)

Diffuse Calcifications

They are irregular areas of calcification in the pulp tissue that can be seen as a large mass or fine spicules of calcified tissue. They generally follow collagenous fiber bundles and blood vessels. The diffuse calcifications are usually

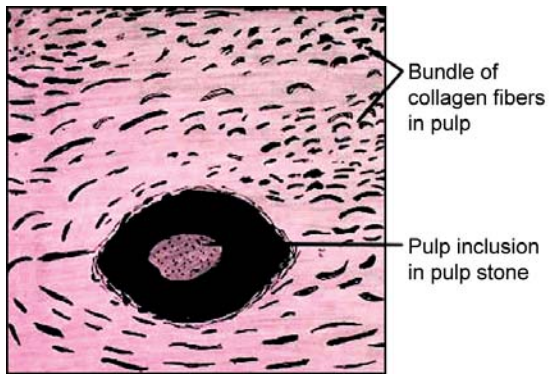


Figure 6.74: Pulp in the center of free, false pulp stone which is seen within collagen bundle of coronal pulp

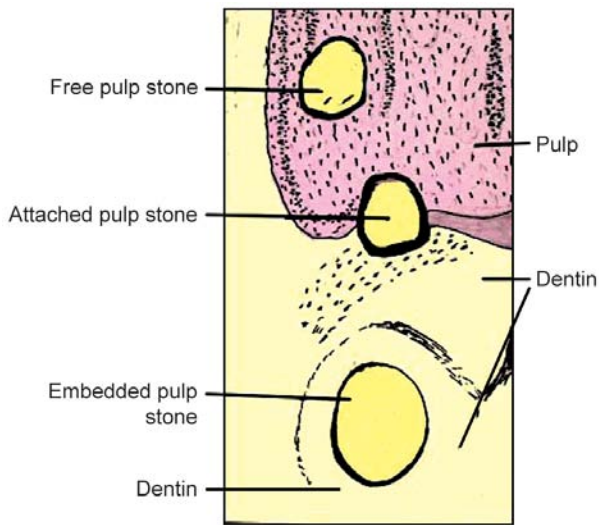


Figure 6.75: Typical appearance of pulp stone as free, attached and embedded, depending on their relation to the dentin of the tooth

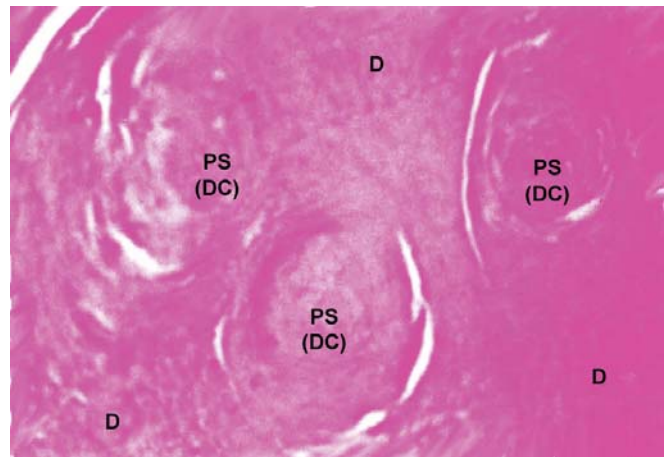


Figure 6.77: Embedded pulp stones (Interstitial pulp stones). Three true pulp stones (PS) or Denticles (DC) embedded in the dentin (D) (H & E x 50)

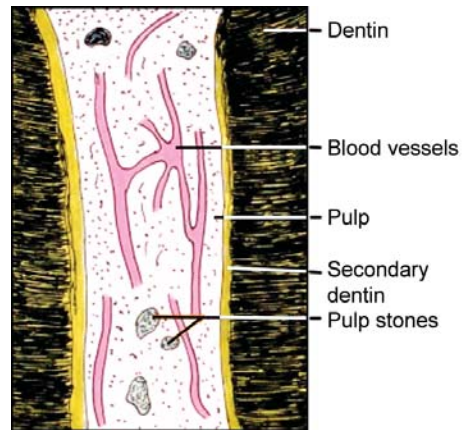


Figure 6.78: Longitudinal section of the pulp showing the anastomosis of blood vessels and free small pulp stones

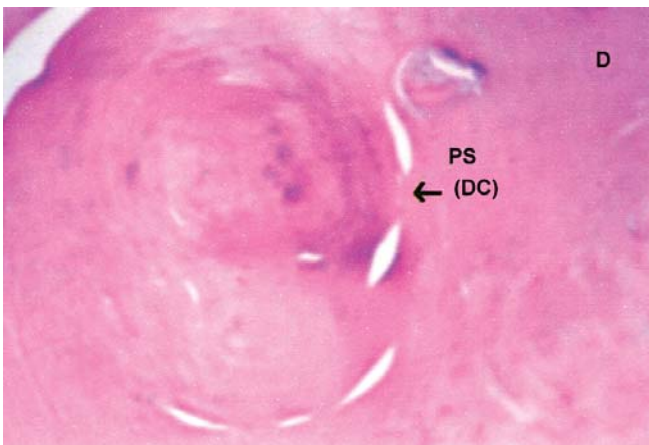


Figure 6.76: A round circular Pulp stone (PS) or Dentine (DC) embedded in the dentin (D). Decalcified Tooth (H & E x 200)

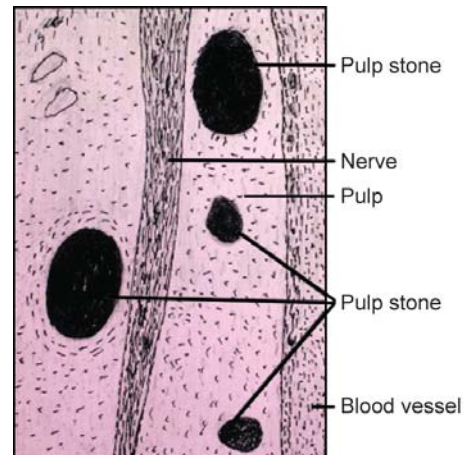


Figure 6.79: Pulp stones close to nerve in pulp and blood vessels

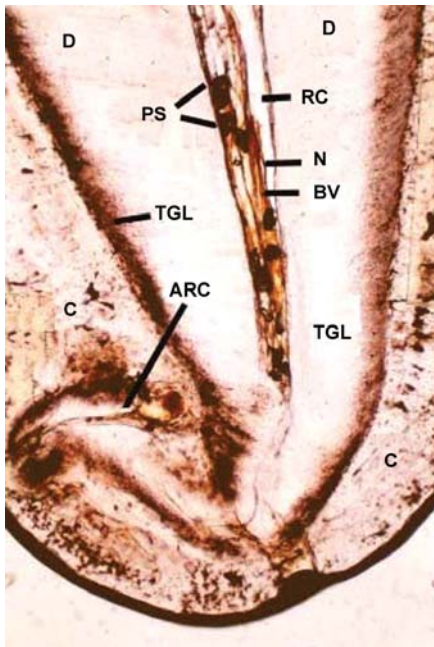


Figure 6.80: Low power Photomicrograph of ground section of tooth showing many pulp stones (PS) in the root Canal (RC) and accessory root canal (ARC). Thin blood vessels (BV) and nerves can be seen in the root canal. C = Cementum, D = Dentin, TGL = Tomes' granular layer

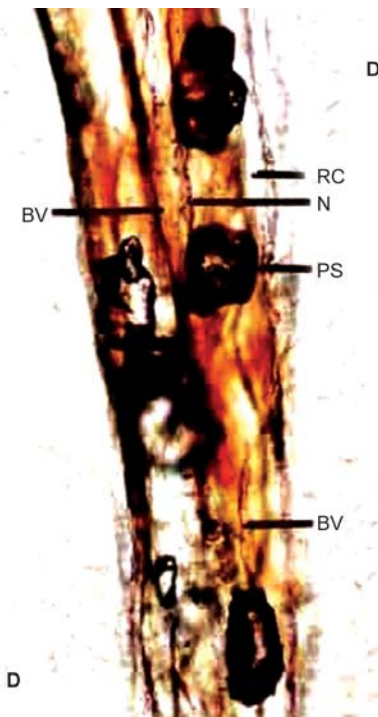


Figure 6.81: Higher magnification of a portion of main canal. Ground section showing pulp stones (PS) in the root canal (RC). Vascular supply of the pulp is seen distinctly. D= Dentin, BV= Blood vessel, N= nerve

found in the root canal and are rarely seen in coronal pulp, whereas pulp stones are mostly seen in the coronal pulp (Fig. 6.82). Radicular pulp may show diffuse calcifications and coronal pulp may be normal without any pathology. Diffuse calcification surrounds the blood vessels and is known as dystrophic calcification (Fig. 6.83). This calcification may occur in a linear or concentric pattern (Figs 6.85 to 6.87).

Regressive Changes (Aging)

Cell Changes

In the aging pulp, the cells are present in lesser number. They are characterized by decrease in the number and size of cytoplasmic organelles. The pulpal fibrocytes or fibroblasts have abundant, rough surfaced endoplasmic reticulum, prominent Golgi apparatus and numerous

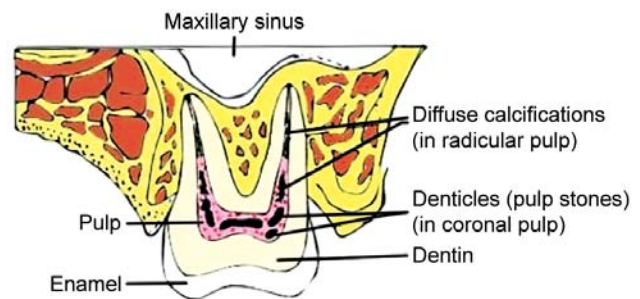


Figure 6.82: Longitudinal section of a maxillary molar showing the coronal and radicular pulp with pulp stones and diffuse calcifications respectively

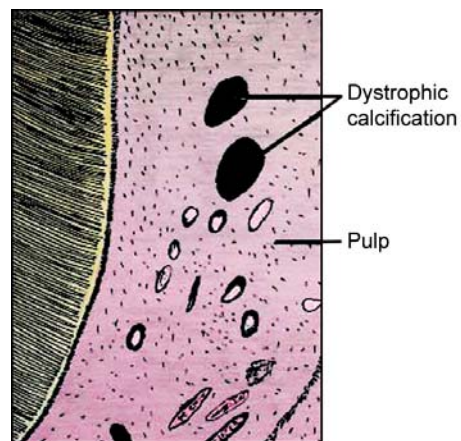


Figure 6.83: Dystrophic calcification may occur in concentric or linear pattern in the center of the pulp chamber

mitochondria. The fibroblasts of aged pulp have less perinuclear cytoplasm with long, thin, cytoplasmic processes. The intracellular organelles are reduced in size and number.

Fibrosis

With aging of the pulp, fiber content increases and cellular content decreases. Fiber bundles arrange in a random, more diffused fashion in the coronal pulp and longitudinally in bundles in the radicular pulp. In the medial and adventitial layers of blood vessels, collagen increase. Due to a decrease in the total volume of pulp, the collagen fibers also appear condensed and concentrated without actual increase in their total volume. Therefore accumulations of bundles of collagen fibers and diffuse fibrillar components are usually seen in the aging pulp (Figs 6.84 and 6.85). Any external trauma produces localized fibrosis or scarring effect. The fiber increases in the pulp slowly and it is generalized throughout the pulp tissue (Figs 6.86 and 6.87).

Vascular changes: Changes in capillary endothelium due to aging are the presence of (a) numerous pinocytic vesicles, (b) microfilaments, (c) glycogen granules, (d) microvesicles and many Golgi complexes. Vascular changes in the form of decreased vascularity also exist with aging pulp (Fig. 6.86). The calcification occurs in the walls of blood vessels near the apical foramen. There is an increase in collagen in medial and adventitial layers of blood vessels, which appears more than actual (Fig. 6.88). Atherosclerotic plaque may appear in pulpal vessels.

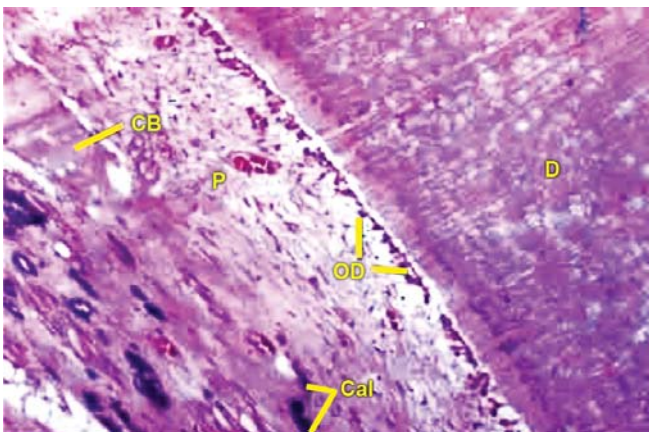


Figure 6.84: Low power view of root canal showing diffused linear calcification (cal) of pulp (P). Collagen bundles (CB) are increased in amount and bundles are thick. OD = Odontoblasts, D = Dentin x 100

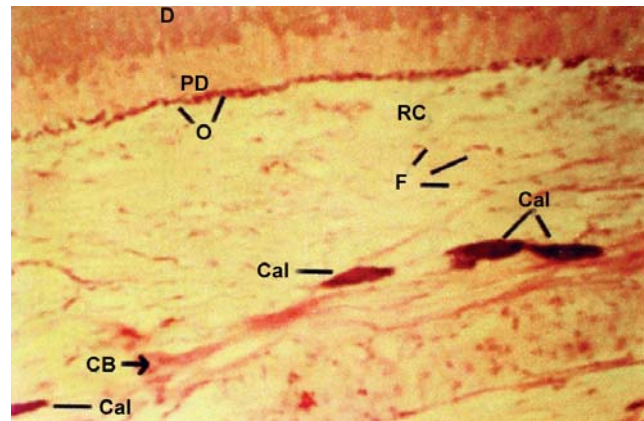


Figure 6.85: Linear calcification (Cal) deposits in the root canal (RC) which otherwise appears normal. Thin fibroblasts (F) and short thick collagen bundles (CB) are present. O = odontoblast PD= predentiin, D = Dentin (H & E x 200)

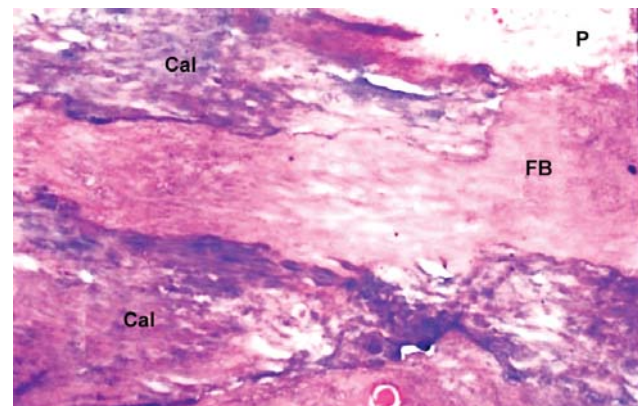


Figure 6.86: Pulp fibrosis and diffuse calcification generally are seen together. High magnification of pulp (P) showing areas of fibrosis (FB) and calcification (cal). There is marked decrease in vascularity of pulp (H & E x 200)

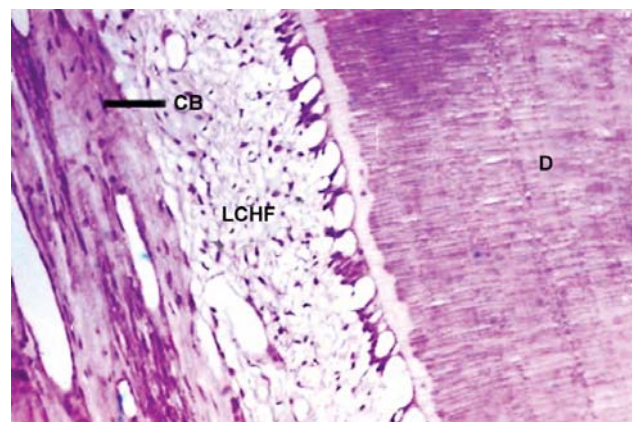


Figure 6.87 A: Pulp from an elderly person showing low cellularity, high fibrosis (LCHF) and presence of thick collagen bundles (CB), D = Dentin (H and E x 100)

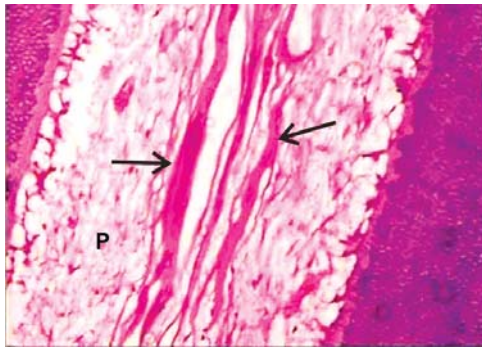


Figure 6.87B: Pulp from an elderly person show low cellularity and presence of thick collagen bundles (shown by arrows) in the pulp (P) x 150

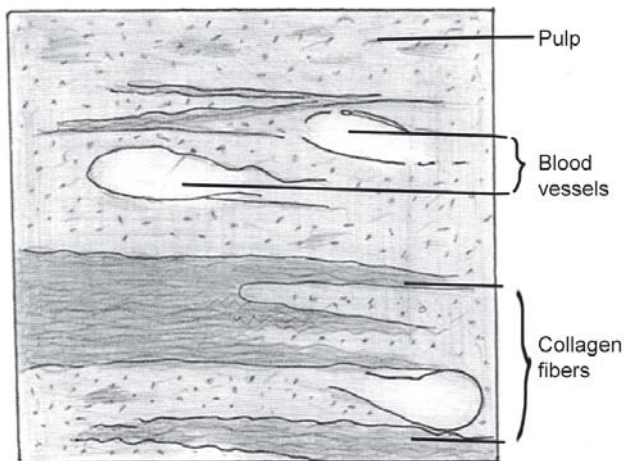


Figure 6.88: Bundles of collagen fibers around and among blood vessels of pulp (as seen under electron microscope)

CLINICAL CONSIDERATIONS

The dental pulp has a rich vascular supply. It responds to external stimuli by forming reparative dentin as a protective measure. The shape and size of pulp cavity is different in permanent and deciduous tooth. The pulp horns are comparatively higher in deciduous teeth. The predetermination of the shape and size of pulp chamber and the extension of the pulp horns is important for avoiding hazardous cavity preparation. While performing root canal treatment, the shape, size and number of root canals and shape and location of the apical foramen must be kept in mind for successful treatment of root canals. The size and variation in shape of the root canals and pulp chamber should be considered before opening of pulp chambers.

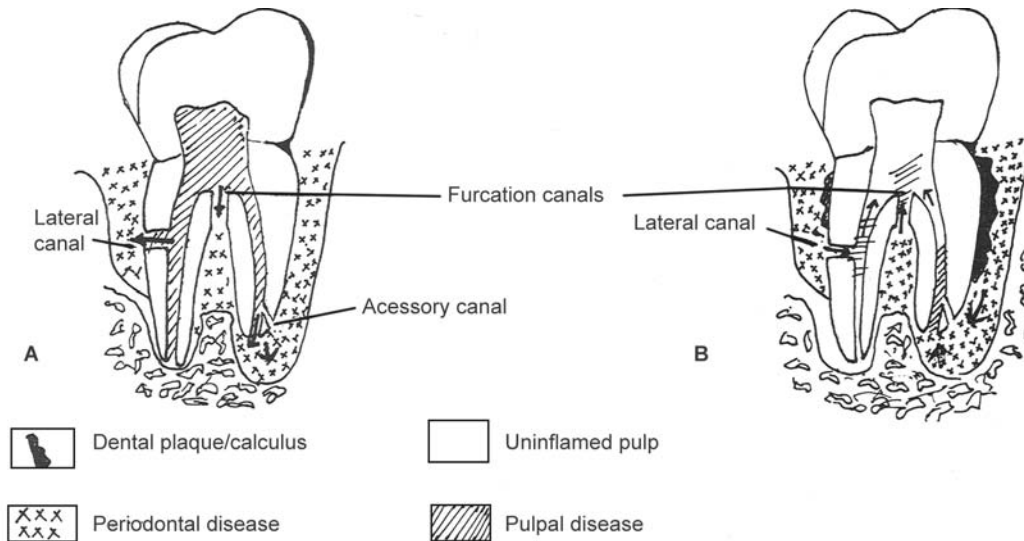
As age advances, gradually coronal and radicular total pulp space becomes smaller. There is an excessive secondary dentin formation at the roof and floor, so it is difficult to locate root canals in aged patients. To overcome this problem it is better to first advance towards distal root in lower molar and towards palatal root in upper molars before searching for other canals, as these canals are wider and easier to locate.

For treatment of root canals shape of the apical foramen should also be considered. When apical foramen is narrowed by cementum, it is readily located as further progress of endodontic instrument is stopped.

When accessory canals are present close to coronal part of the root or in the bifurcation region, they may play an important role in the spread of disease from pulp to periodontium or from periodontium to the pulp (Figs 6.89A and B). An accidentally, non-infected or minimally infected exposed pulp can be preserved by pulp capping procedure. In this process reparative dentin is formed at the site of exposed pulp. Thus, a dentin bridge or barrier is formed at the site of exposure, and the vitality of pulp is maintained. The pulp capping procedure is more successful in deciduous and young permanent teeth than in the older permanent teeth. Most restorative materials containing calcium hydroxide readily induce the formation of reparative dentin. Enamel matrix derivative promote the reparative process in the wounded pulp. It has been observed that Mineral trioxide aggregate formed thick dentin bridge and produce less inflammation, necrosis and hyperemia. Pulp capping materials of future are bioactive molecules like (a) bone morphogenetic protein, (b) TGF β -1, or purified dentin protein fractions, (c) tissue cultured dentin, and (d) stem cells.

The inflammatory or defense cells are generally present in small numbers in normal pulp (Fig. 6.90). The main characteristic clinical feature of acute pulpitis is extensive acute inflammation (Figs 6.91 to 6.93).

This is characterized by increased cellularity in the early stages of the pulpal disease (Fig. 6.91). When only a small area of the pulp is involved, severe pain is elicited. This pain is especially caused by cold. Large diameter A- δ fibers are fast conducting myelinated fibers and produce a sharp pain. Small diameter C- fibers are slow conducting non-myelinated fibers and produce dull pain on stimulation. Any pulpitis shows increased sensitivity to heat and cold (Figs 6.91 to 6.94).



Figures 6.89 A and B: Pathway of communication between pulpal and periodontal tissues. A. Pulpal irritants exit through the apical foramen, furcation, lateral and accessory canals to affect periodontal tissues. B. Periodontal irritants enter to affect the pulpal tissues through apical foramen, furcation, lateral and accessory canals

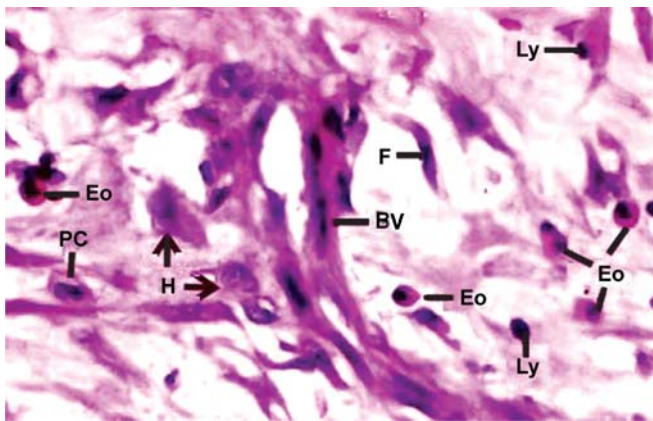


Figure 6.90: Pulp showing presence of small number of inflammatory cells which are present normally as defence cells. A small blood vessel (BV) and fibroblasts (F) are also seen. Eo = Eosinophil, H = Histiocyte, PC = Plasma cell, Ly = Lymphocyte (H & E x 400)

Vitalometer is the instrument which tests the reaction of the pulp to the thermal stimuli or electrical stimuli. This instrument checks the sensitivity of the pulp by providing the information about the status of the nerves supplying the pulpal tissue. Laser Doppler flowmetry and Transmitted light Photoplethysmography check the vitality of the tooth by recording the pulpal blood flow.

Age causes certain important changes in pulp. There is continuous deposition of secondary dentin throughout life. There is deposition of reparative dentin in response

to stimulation. Both of these reduce the size of the pulp chamber thereby decreasing the size and volume of the pulp. Along with this, there is continuous deposition of peritubular dentin also. Some of the dentinal tubules close completely and form sclerotic dentin. There is also a reduction of fluid content of the dentinal tubule.

The decrease in pulpal volume reduces the cellular, muscular and neural content of the pulp. The collagen fibers increase in number and size.

All these changes make the dentin less permeable and more resistant to external stimulus.

Presence of lateral and accessory canals is a normal feature of pulp. These canals pose a problem in cleaning and obturation because pulp tissue lies within these canals.

These lateral and accessory canals frequently occur in apical third of the root. These are also found in areas of bifurcation and trifurcation of multirooted teeth. All these canals may contain vital pulp after the pulp is removed from the main canal. However with age, calcification occurs in the contained soft tissues of these canals (Fig. 6.95). Hence with advancing age, the number of accessory canals normally decreases.

Those operative procedures, which produce dehydration of dentinal tubules and damage to the pulpal tissue, should be avoided. A tooth without vital pulp becomes brittle and is subject to fracture. Therefore, every effort should be made to preserve the vitality of the pulp.

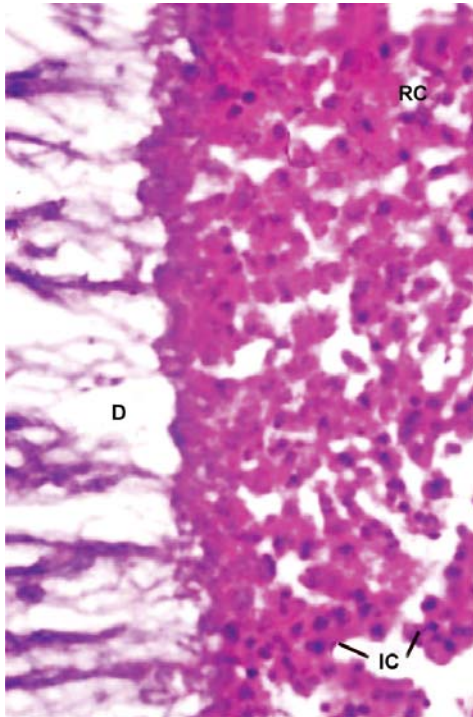


Figure 6.91: Severe acute inflammation of pulp. Root canal (RC) is packed with acute inflammatory cells (IC) obliterating all the normal zones of pulp. D= Dentin (H & E x 200)

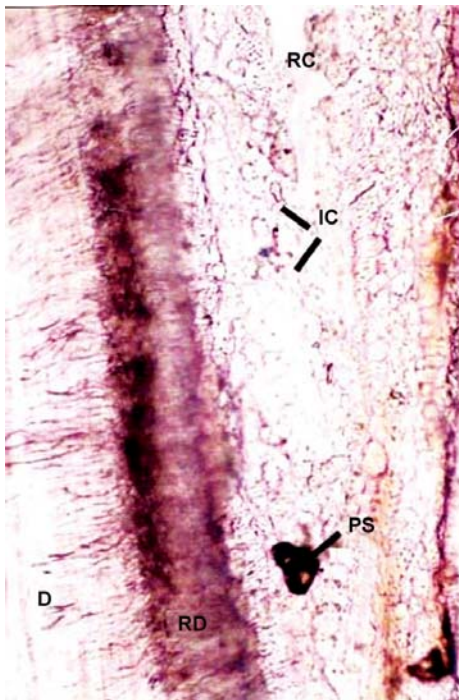


Figure 6.92: Pulpitis- Ground Section of a tooth showing presence of numerous inflammatory cells in root canal (RC) in a case of severe pulpitis. Unstained round inflammatory cells (IC) are present in the root canal. Two pulp stones (PS) can be seen lying in the canal. D = Dentin

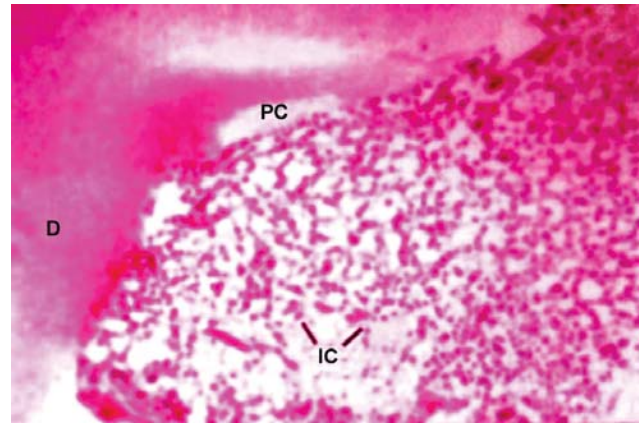


Figure 6.93: In severe chronic pulpitis, the pulp chamber (PC) is infiltrated diffusely by chronic inflammatory cells (IC). D= Dentin (H & E x 100)

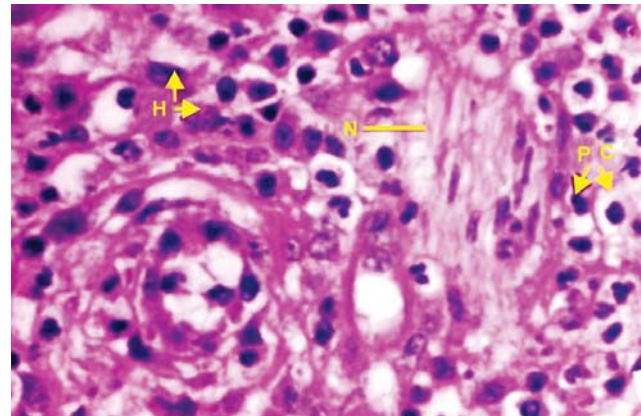


Figure 6.94: Nerve twig (N) running through the pulp in a tooth showing pulp infection. H= Histiocytes, PC= Plasma cell (H & E x 400)

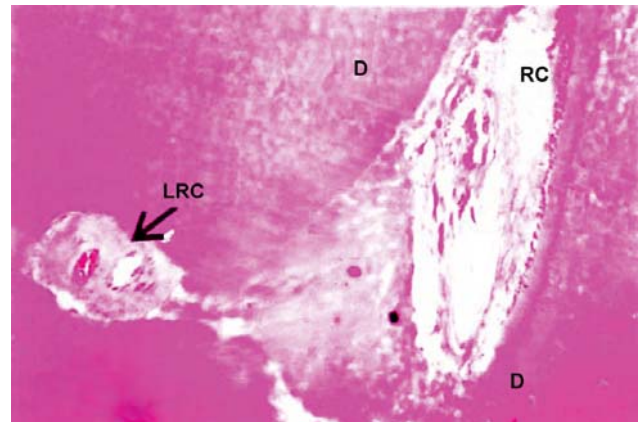


Figure 6.95: Horizontal section at apical third of root showing lateral root canal: Horizontal extension of root canal (RC) is seen laterally into the dentin (D) and is called lateral canal. Such lateral root canals (LRC) pose problem in successful treatment of pulp infection. (H & E x 100)

The successful management of pulpal diseases and preservation of vitality of dental pulp are the most important challenges to the dental surgeon.

Due to heavy masticatory forces on a sharp object, margin of tooth or a filling may fracture or crack. Through this, microorganisms and their toxins may penetrate into pulp resulting in pain, pulpitis and pulp pathology.

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Cementum

- Introduction
- Physical and chemical properties
- Cementogenesis
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- Structure of cementum
- Classification
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- Functions of cementum
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INTRODUCTION

Cementum is a hard layer of calcified mesenchymal tissues which forms the outer covering of the anatomical root. It is also called as substantia ossea. It was first microscopically demonstrated by two pupils of Purkinje in 1835. Cementum extends from the cemento-enamel junction to the apex of tooth. It forms a covering on root surface, which seals the open dentinal tubules of root dentin and provides an attachment for periodontal ligament that holds the tooth in its socket.

The outer surface of the cementum provides a medium for the attachment of periodontal ligament fibers that support the teeth within the jaw. Cementum is one of the four tissues that forms the periodontium. Other tissues of periodontium are the periodontal ligament, alveolar bone and the gingiva. The inner surface of the cementum is firmly attached to the dentin. Human cementum is avascular and has no innervation. It has some physical, chemical and structural properties, which are same as that of the surrounding alveolar bone.

It is thickest near the apex and bifurcation area of root and is thinnest at the cervical region of the tooth.

PHYSICAL AND CHEMICAL PROPERTIES

The hardness and calcification of the cementum, even when it is fully mineralized, are less than that of the dentin. Cementum is light yellow in color, having a dull surface and a slightly darker hue. It does not have a shine. Cementum is lighter in color and is also softer and more permeable than dentin. On becoming nonvital it becomes darker in color as is usually seen in teeth exfoliated or extracted due to periodontal diseases. The permeability decreases as calcification increases with age. The softness of cementum makes it susceptible to abrasion, when it is exposed in the oral environment due to recession of gingiva. Cementum does not have the ability to remodel. It is more resistant to resorption than bone. This is clinically important specially for various orthodontic procedures.

Cementum consists of organic matter, inorganic matter and water. By volume, cementum contains 35 percent organic materials, 45 percent inorganic material and 20 percent water. On a dry weight basis, cementum contains 45 to 50 percent inorganic substance, 50 to 55 percent

organic materials and water. The organic matter of cementum consists primarily of type I collagen fibers, and interfibrillar ground substance consists of proteoglycans (Protein polysaccharides). Collagen types III, V, VI and XII are also seen. The principal inorganic material is hydroxyapatite - $\text{Ca}_{10}[\text{PO}_4]_6[\text{OH}]_2$, which is less than that in bone (65%), enamel (97%) and dentin (70%). The hydroxyapatite crystals consist of calcium and phosphate. Numerous trace elements are also present in the cementum. The fluoride content of cementum is highest in comparison with all the other calcified tissues of body. Noncollagenous proteins are important in matrix deposition, matrix remodeling, initiation and mineralization. Osteopontin regulates mineralization and cementum derived attachment protein (CAP) helps in the attachment of mesenchymal cells to the extracellular matrix. Sialoprotein and osteopontin fill the large interfibrillar spaces. Chondroitin sulfate, heparan sulfate, hyaluronate, keratan sulfates – fibromodulin and lumican, versican, biglycan and osteoadherin are present in bone and cementum.

CEMENTOGENESIS

Cementum is deposited on the surface of root dentin. In cementogenesis, the formation of an acellular (primary) cementum and cellular (secondary) cementum takes place. Cementum develops from the activity of mesenchymal cells of dental follicle after fragmentation of the epithelial root sheath. After formation of dentin, loss of continuity occurs in the epithelial root sheath. This allows adjacent cells of the investing layer of the dental follicle to come to lie on surface of the root dentin and these are induced to differentiate into cementoblasts. The cementoblasts secrete collagen and components of the ground substance.

Cementum formation starts after the first layer of dentin of root is laid down. There is formation of a structureless, highly mineralized layer of about 10 micron thickness on the surface of the root dentin. This is known as intermediate cementum. As the collagen fibers of mantle dentin form, they are not deposited immediately against the basement membrane, supporting the root sheath. A gap of about 10 microns is left which is filled up with ground substance and fibrillar material. The basement membrane supporting the root sheath breaks up and the root sheath develops rough endoplasmic reticulum and secretes a material. This material helps in the formation of an intermediate cementum.

When the root dentin formation has begun, the Hertwig's epithelial root sheath that is continuous, fragments and forms a network. This network enables mesenchymal cells to pass between the cells of root sheath. Now, these come into apposition with newly formed root surface. These mesenchymal cells are known as

cementoblasts. These cementoblasts increase in size and they develop all the cytoplasmic organelles that synthesize and secrete proteins. Now these cells start depositing the organic matrix of cementum, which consists of the intrinsic collagen fibers and ground substance (Figs 7.1 to 7.6).

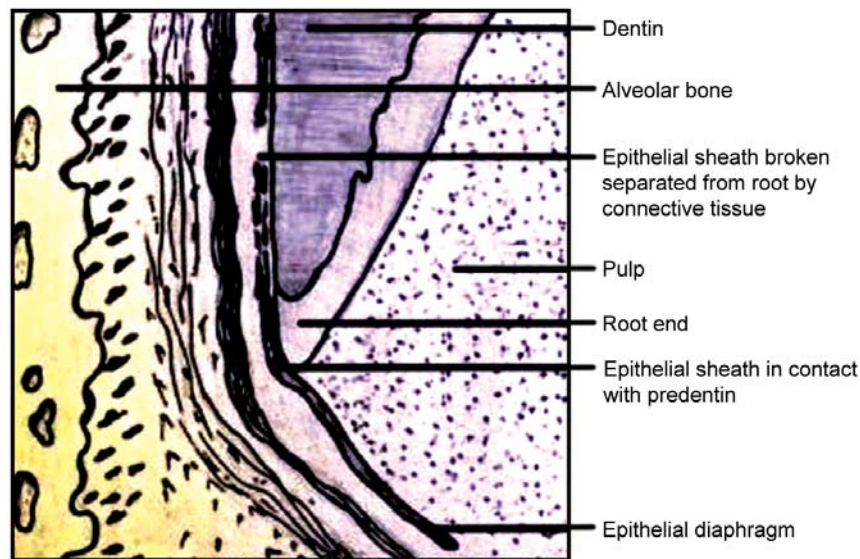


Figure 7.1: Hertwig's epithelial root sheath at the end of forming root. Cementum formation begins at the side of root where the sheath is broken up

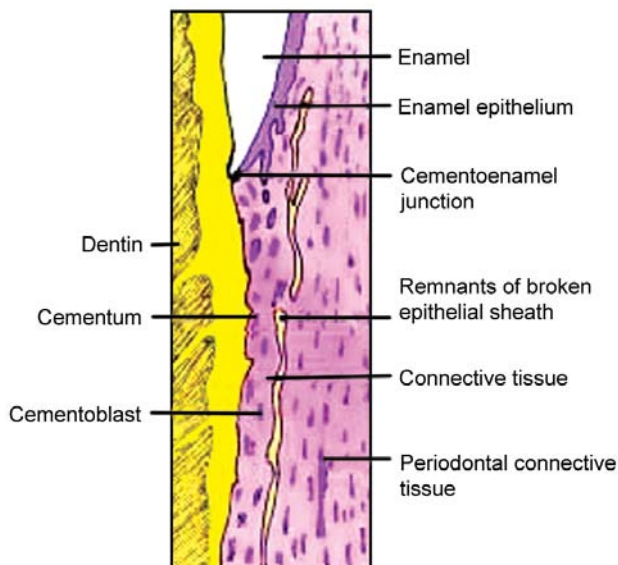


Figure 7.2: Broken epithelial sheath is separated from root surface by connective tissue

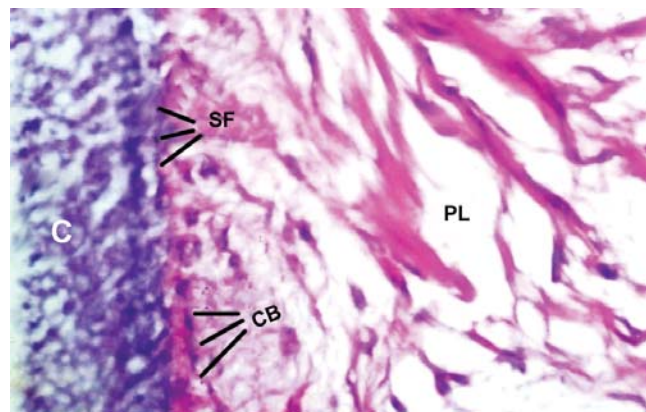


Figure 7.3: Cementogenesis: cells of periodontal ligament (PL) at the periphery become activated and specialized to produce cementum (C). These cells are called cementoblasts (CB). The collagen fibers of periodontal ligament enter cementum for anchorage and are called Sharpey's fibers (SF) x 500



Figure 7.4: Cementum: Collagen fibrils make up the bulk of the organic portion of the tissue and ground substance is interspersed between collagen fibrils. (As seen under electron microscope)

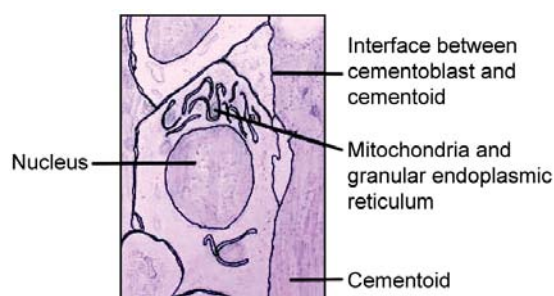


Figure 7.5: Cementoblasts on cementoid surface as seen under electron microscope

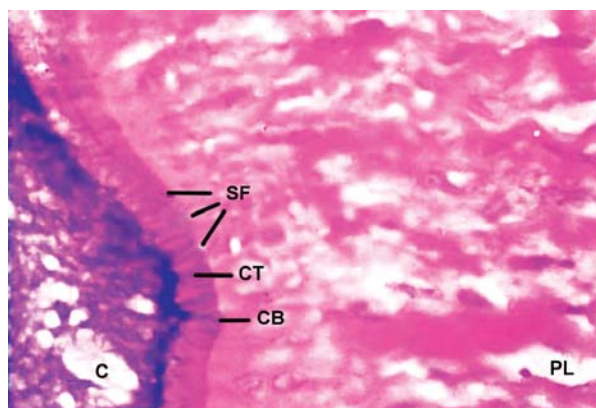


Figure 7.6: Cementogenesis: Cementoblasts (CB) lay down matrix or ground substance called cementoid tissue (CT) which soon becomes mineralized to form cementum (C). Periodontal ligament (PL) shows horizontal collagen fibers (SF) Sharpey's Fibers (H & E x 500)

Cementum is laid down much slowly while the tooth is erupting. This cementum is acellular or primary. When the tooth comes in occlusion, more cementum forms around the apical two-thirds of the root, which has greater proportions of collagen. The cementoblasts become

trapped in lacunae within this matrix, hence this cementum is called cellular (secondary) cementum. Cementoid is laid down in successive layers followed by mineralization. This process is continuous and the thickness of cementum on root surface gradually and slowly increases with age.

During cementogenesis, the cementoblasts have intracytoplasmic organelles that are responsible for synthesis and secretion of organic matrix. Most of the collagen in primary cementum is derived from the Sharpey's fibers of periodontal ligament. These are also known as extrinsic fibers of cementum. The Hertwig's epithelial root sheath cells migrate away from the root surface towards adjacent dental sac. The cells of Hertwig's epithelial root sheath form epithelial rests of Malassez that are found in the periodontal ligament of completely formed teeth (Fig. 7.7).

CELLS OF CEMENTUM

The cells that are concerned with cementum formation are cementoblasts and cementocytes. The cementoblasts synthesize collagen and protein polysaccharides that form organic matrix of cementum. After Hertwig's epithelial root sheath breaks up, the cementoblasts are formed by differentiation of adjacent undifferentiated mesenchymal cells.

Cementoblasts are of two types (Table 7.1).

- A. Cementoblasts producing cellular cementum
- B. Cementoblasts producing acellular cementum

Table 7.1: Differences between two types of cementoblasts

A. Cementoblast producing cellular cementum	B. Cementoblast producing acellular cementum
1. Cementoblast derived from dental follicle	Cementoblast derived from Hertwig's epithelial root sheath.
2. E 11 antibody strongly reacts with this cementoblast.	E 11 antibody does not strongly react with this cementoblast
3. Cementoblast express receptors for parathormone	Receptors for parathormone are not expressed
4. Cementoblast express osteopontin and osteocalcin	Cementoblast express osteopontin only.

The cementoblasts are responsible for producing cementum. When active, they are round, plump cells with a basophilic cytoplasm. These cells have well formed Golgi apparatus, numerous mitochondria and large amounts of granular endoplasmic reticulum. (Fig. 7.8).

The uncalcified matrix of cementum is known as cementoid (Figs 7.5 to 7.8). The mineralization of cementum begins after formation of some organic matrix of cementum. Mineralization of cementoid is a highly ordered event by deposition of calcium and phosphate ions in the form of hydroxyapatite. It is a rhythmic process, as new layers of cementoid are formed after the old one calcifies. Osteopontin regulates growth of apatite crystals and Gla proteins like osteocalcin and osteonectin promote mineralization.

It is usually seen on the cemental surface. The cementoid tissue is lined by cementoblasts. Several connective tissue fibers from the periodontal ligament pass between cementoblasts into the cementum. These fibers get embedded into the cementum and attach the tooth to surrounding bone. Their embedded portion is known as Sharpey's fibers. These fibers are composed of numerous collagen fibrils.

The deposition of cementum continues in phases throughout life. Resting cementoblasts have a closed nucleus and little cytoplasm. When acellular cementum is formed, the cementoblasts retreat leaving behind cemental matrix. Development of acellular cementum is related with Hertwig's epithelial root sheath (HERS). HERS secrete cementum related proteins like osteopontin, fibrillar, collagen and bone sialoprotein (BSP).

During formation of cellular cementum, the cementoblasts become incorporated into the cemental matrix. These cells are known as cementocytes. The cementocytes have sparse amount of cytoplasm and possess numerous cell processes or canaliculi, radiating

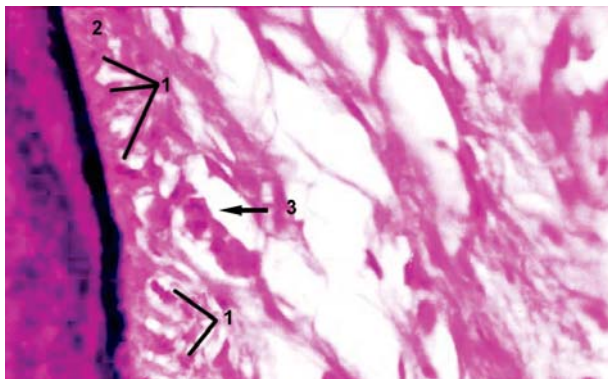


Figure 7.7: Cementum: Cementoblasts (1) lining the surface of cementum. These cells differentiate from the undifferentiated mesenchymal cells of periodontal ligament. These cells first lay down pink matrix cementoid (2) which later gets mineralized to form cementum. Small island of epithelial rest of Malassez (3) is also seen (H & E x 200)

from their cell body. These processes may branch and anastomose with similar processes of the adjacent cementocytes. The cementocytes lie in space known as lacunae (Figs 7.9 to 7.11).

The cementocytes receive their nutrients from periodontal ligament by the process of diffusion. As formation of cementum continues, the existing cementocytes become progressively farther from periodontal ligament. The cementocytes present in deeper layers of cementum contain few organelles in their cytoplasm. At a depth of 60 microns or more, the cementocytes exhibit definite signs of degeneration like cytoplasmic clumping and vesiculation due to nutritional problems of these cells. Thus, they leave empty lacunae in the deeper cementum (Fig. 7.12).

STRUCTURE OF CEMENTUM

Under light microscope, two types of cementum are visible. Based on the presence or absence of cells, they are called: (a) acellular (primary) and (b) cellular (secondary) cementum (Fig. 7.13).

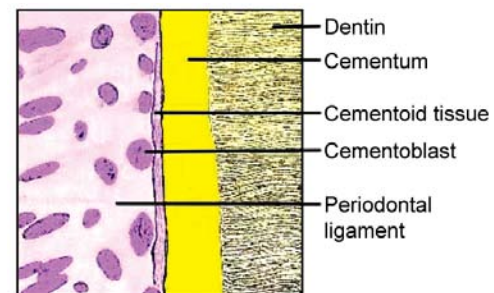


Figure 7.8: Cementoid tissue on the surface of calcified cementum. In between periodontal ligament fibers cementoblasts are present

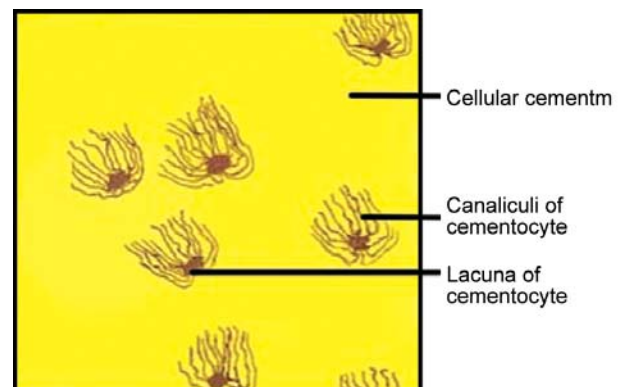


Figure 7.9: Lacunae and canaliculi of cementocytes in cellular cementum

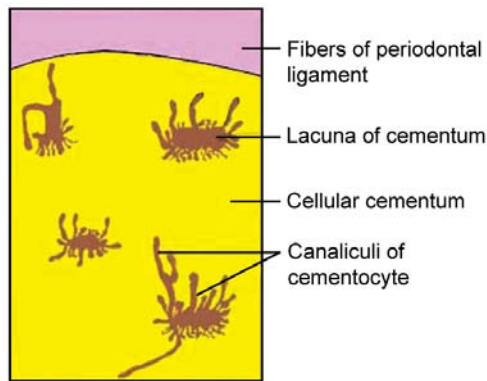


Figure 7.10: Mineralized ground section of premolar showing spider like cementocytes with numerous canaliculi

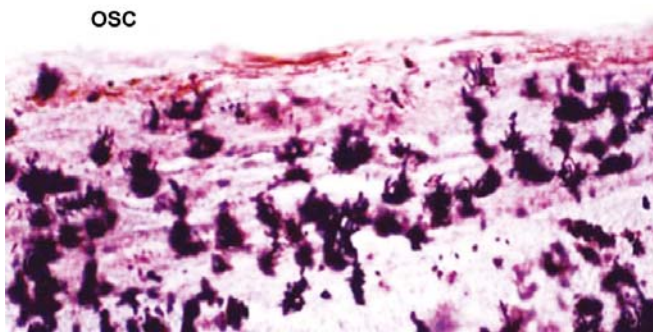


Figure 7.11: Cementum: Ground section of tooth: Cellular cementum showing cementocytes with thin cytoplasmic processes or canaliculi resembling spider legs and pointing towards outer surface of cementum (OSC) (x 150)

There is no definite pattern of distribution of these two types of cementum. These may be arranged in an alternate pattern. In classical presentation, acellular cementum predominates in the coronal half of the root and cellular cementum is present in the apical half of root.

Occasionally, acellular cementum may be found on the surface of the cellular cementum. Frequently, cellular cementum is formed on the surface of acellular cementum. It may also be present on the entire thickness of apical cementum (Fig. 7.14).

A cellular cementum is the first formed cementum and is therefore referred to as primary cementum (Fig. 7.15). It covers approximately the cervical third or half of the root. It does not contain cells. This cementum is formed before tooth reaches the occlusal plane and its thickness ranges from 30 to 230 microns. It is thinnest at the cemento-enamel junction and thickest towards root apex. By its continuous growth, it contributes to the length of root (Fig. 7.16). It has a lamellated structure like bone and shows resting lines of periods of inactivity (Fig. 7.17). These resting lines or incremental lines are more calcified than the interlamellar cementum. The resting or incremental lines are present in both acellular and cellular cementum between the layers. Sharpey's fibers make up most of the structure of acellular cementum.

It is observed by scanning electron microscope that resting cemental surfaces show low, rounded projections, which correspond, to the centers of Sharpey's fibers.

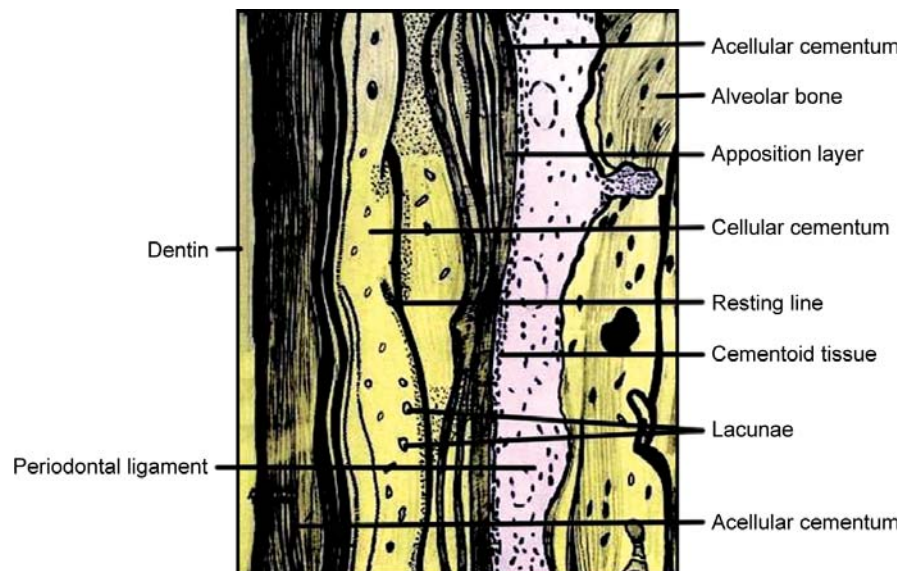


Figure 7.12: Alternate layers of cellular and acellular cementum. Degenerated cementocytes appear as empty lacunae in cellular cementum

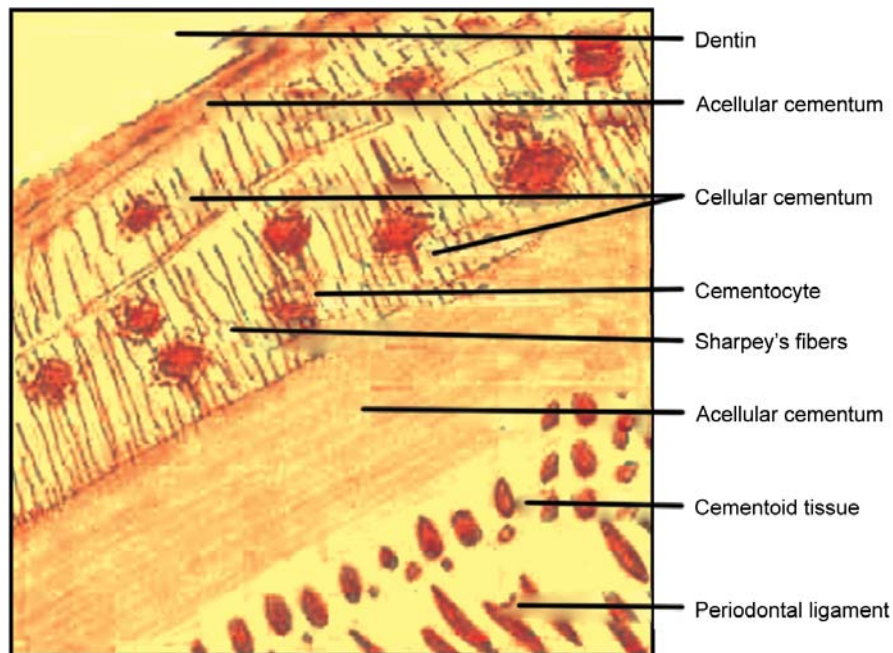


Figure 7.13: Sharpey's fibers and acellular cementum and cellular cementum as seen in ground section

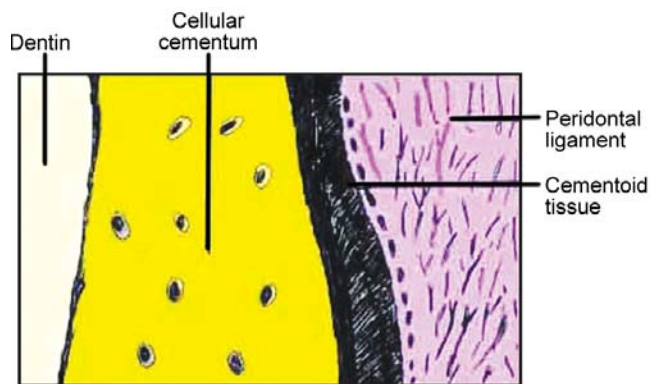


Figure 7.14: Cellular cementum comprises of the entire thickness of apical cementum

Cemental surfaces with actively mineralizing sites have many small openings, which are the sites where individual Sharpey's fiber enter the tooth.

Sharpey's fibers make up most of the structure of acellular cementum. These have a principal role of supporting the tooth within jaw. The Sharpey's fibers are inserted at right angles to root surface and penetrate deep into the cementum. They run at right angles to the collagen fibers of cementum. The number, size and distribution of Sharpey's fibers increases with function of teeth. The fibers which are derived from Sharpey's fibers are called

extrinsic fibers. The collagen fibers derived from cementoblasts are called intrinsic fibers. The intrinsic fibers are arranged parallel to root surface. The collagen fibers present in cementum are both extrinsic fibers and intrinsic fibers. The calcified structure of acellular cementum consists of collagenous fibers and ground substance. Sharpey's fibers are also completely calcified in the acellular cementum. The inorganic mineral crystals are oriented parallel to the collagen fibrils of cementum. A 10 to 50 micron wide zone of Sharpey's fibers near the

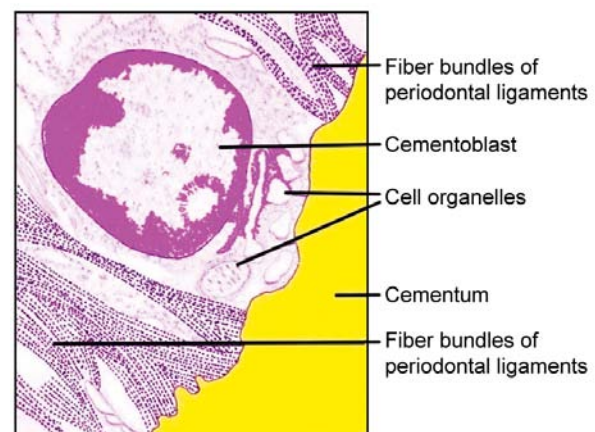


Figure 7.15: Formation of acellular cementum



Figure 7.16: Cementum is continuously deposited at the root apex which contributes to the length of root

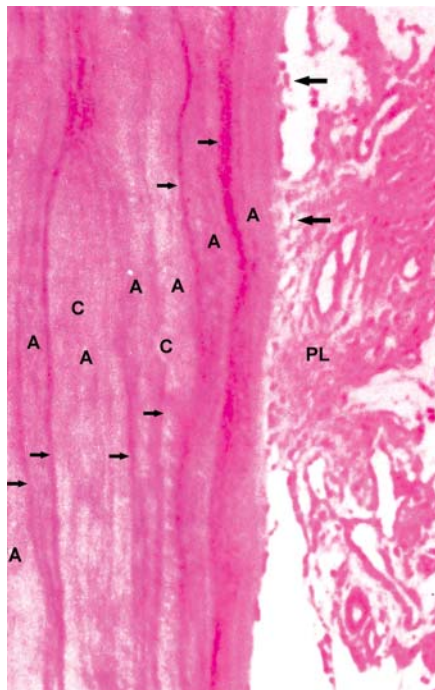


Figure 7.17: Cementum (C) showing resting lines and apposition in decalcified tooth. Resting lines have been indicated by short arrows. Periodontal ligament (PL) shows single layer of cuboidal cells on the surface of cementum called cementoblasts, indicated by arrows. (H & E x 200). Apposition layers have been shown by A

cementodentinal junction is partially calcified. According to evidence obtained by scanning electron microscopy, the peripheral portions of Sharpey's fibers are more calcified than the interior portions of Sharpey's fibers (Figs. 7.18).

Cellular cementum is formed after the tooth reaches occlusal plane. It is more irregular and contains cells within its matrix called cementocytes (Fig. 7.19). The

cementocytes are present in individual spaces called as lacunae and their processes lie in the canaliculi. The canaliculi are directed towards the periodontal ligament. Acellular cementum is more on the coronal half of the root and cellular cementum is more on the apical half of the root. Layers of acellular and cellular cementum are usually laid down in an alternate pattern.

Cellular cementum is less calcified than the acellular cementum. Sharpey's fibers make up a smaller portion of cellular cementum and are separated by collagen fibers. The collagen fibers are arranged parallel to the root surface.

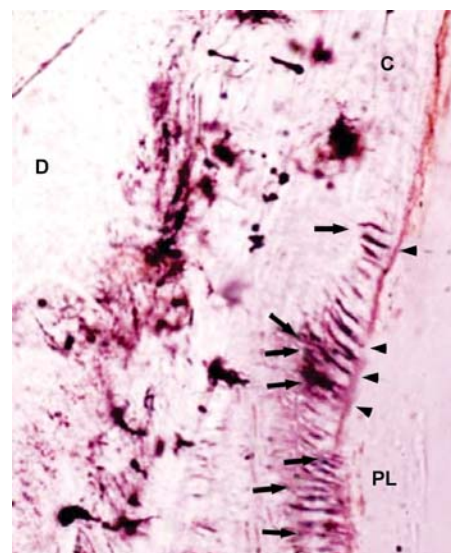


Figure 7.18: Sharpey's fibers indicated by the arrows are the collagen fibrils, which extend from periodontal ligament (PL) into cementum. In cementum they are shown by arrows (C) acting as an anchor. D = Dentin, Ground section of tooth. (x 100)

Cellular cementum is formed at a faster rate than the acellular cementum. The apical third of the root is usually covered by cellular cementum. Formation of cementum continues throughout the life. Usually it is cellular cementum, which continuously contributes to the length of root. The periodontal ligament alters or changes according to functional need of the tooth. Both acellular and cellular cementum are arranged in lamellae. These lamellae are separated by incremental lines, which are parallel to the long axis of root. These incremental lines represent rest periods during the formation of cementum. This indicates periodic formation of cementum. These lines are more mineralized than the adjacent cementum. These incremental lines have less collagen and more ground substance than other portions of the cementum (Fig. 7.20).

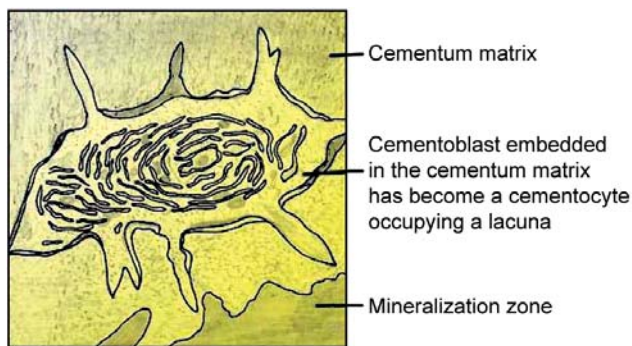


Figure 7.19: Formation of cellular cementum

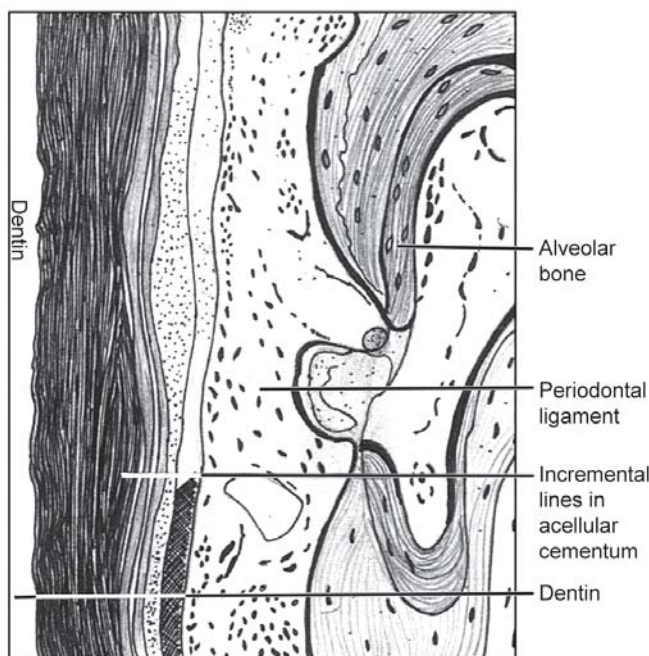


Figure 7.20: Incremental lines in acellular cementum

CLASSIFICATION

Based on the nature and origin of the organic fibrous matrix, Schroeder has classified cementum as follows.

1. Acellular Afibrillar Cementum (AAC)

Acellular afibrillar cementum neither contains cells, nor extrinsic or intrinsic collagen fibers. This type of cementum is a product of cementoblasts and is found as cervical cementum near the crown of tooth as spurs and patches over enamel and dentin along the cemento-enamel junction. Its function is not known.

2. Acellular Extrinsic Fiber Cementum (Primary Cementum) (AEFC)

Acellular extrinsic fiber type of cementum is composed of densely packed bundles of Sharpey's fibers that are derived from the periodontal ligament. It is found in the cervical and middle third of roots but sometimes it may extend further apically. The organic matrix of this type of cementum does not contain any type of cells. This is the only type of cementum observed in single rooted teeth. The space between the extrinsic fibers is filled with the noncollagenous proteins its function is anchorage (Table 7.2).

3. Mixed Stratified Cementum (MSC)

Mixed stratified cementum contains alternating layers of acellular and cellular cementum. Cellular mixed stratified cementum is composed of extrinsic (Sharpey's) and intrinsic (collagen) fibers and contains cells in its matrix. It is situated in apical portion and functions. Its function is adaptation.

4. Cellular Intrinsic Fiber Cementum (CIFC)

It is also called secondary cementum. It is situated at middle to apical third and furcations. Its function is adaptation. Cellular intrinsic fiber cementum has fibers derived almost entirely from cementoblasts. Thus, it is devoid of Sharpey's (extrinsic) fibers. In humans, it fills resorption lacunae. Intermediate cementum is an ill-defined zone near the cementodentinal junction of certain teeth. It contains remnants of Hertwig's epithelial root sheath embedded in calcified ground substance (Table 7.2).

Table 7.2: Differences between the cellular intrinsic fiber cementum (CIFC) and acellular extrinsic fiber cementum (AEFC)

S.No	Character	CIFC (Secondary cementum)	AEFC (Primary cementum)
1.	Location	Present in apical third and furcation areas. It is absent in single rooted teeth.	Present from cervical margin upto apical third and only type of cementum seen in single rooted teeth.
2.	Formation	Later, rapid and known as secondary cementum	Earlier, slow and known as primary cementum
3.	Cementoid	Present on the surface	Absent
4.	Cementoblast	Derived from dental follicle and express parathormone receptor	Derived from HERS and do not express parathormone receptor
5.	Cementocyte	Seen	Not seen
6.	Incremental lines.	Lines are apart	Lines are closer
7.	Fibers	Intrinsic fibers and produced by cementoblast.	Extrinsic fibers and produced by fibroblasts
8.	Noncollagenous protein	Tenascin, fibronectin and osteocalcin are present.	These proteins are absent
9.	Proteoglycans	Versican, decorin, biglycan and lumican are present in matrix.	These proteoglycans are absent
10.	Growth factors	TGF β and IGF are seen.	These factors are not seen.
11.	Main function	Adaptation and repair	Anchorage
12.	Response to local factors	More profoundly	Less profoundly
13.	Matrix vesicles	Involved the mineralization	Not involved in mineralization
14.	Composition	Mg/Ca ratio was higher where Ca/P ratio was lower	Mg/Ca ratio was lower than Ca/P ratio.

CEMENTODENTINAL JUNCTION AND CEMENTOENAMEL JUNCTION

Cementodentinal Junction

The cementodentinal junction is interface between the dentin and cementum. The cementum is attached to dentin firmly (Fig. 7.21).

The cementodentinal junction is sometimes scalloped in deciduous teeth. It is smooth in the permanent teeth. With the electron microscope, a narrow interface area present between the two tissues can be detected. The collagen fibrils of cementum and dentin intertwine at their interface in a very complex manner. Proteoglycans are important for the attachment. It is not possible to determine which fibrils are cemental in origin and which fibrils are of dentinal origin. The cementodentinal junction contains large quantity of collagen associated with glycosaminoglycans like dermatan sulfate and chondroitin sulfate and

help in redistribution of occlusal loads to the alveolar bone. Area of dentin adjacent to the cementodentinal junction appears granular in ground section; possibly due to

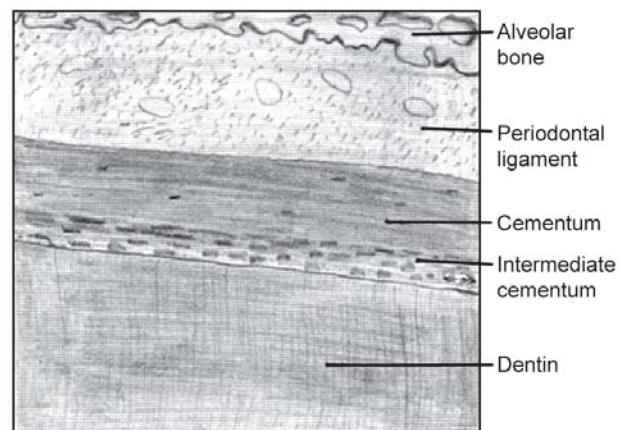


Figure 7.21: Intermediate cementum is present between acellular cementum and dentin

coalescing and looping of terminal portions of dentinal tubules and is called Tome's Granular layer. In decalcified and stained Histologic sections, cementum stains more intensely than dentin. In some teeth, dentin is separated from cementum by a tissue called intermediate cementum. This intermediate cementum layer does not exhibit features either like dentin or cementum. (Fig. 7.22). This layer is predominantly seen in the apical region of the posterior teeth and is rarely seen in deciduous or anterior teeth. It is believed that this layer is formed due to entrapment of cells of Hertwig's epithelial sheath during deposition of dentin or cementum matrix. The area of intermediate cementum can be found as a continuous layer or sometimes it is found only in isolated areas, characterized by wide irregular branching spaces. This layer appears structureless and known as Hyaline layer.

Cementoenamel Junction

The interface between the cementum and enamel at the cervical region of a tooth is known as cementoenamel junction (Figs 7.23 and 7.24 A to C). The relation between the enamel and cementum at the cervical region of teeth can be of three types (Figs 7.25 to 7.27).

Pattern I

The cementum may overlap the cervical end of enamel for a short distance (Figs 7.24 A and 7.25). This type of cementoenamel junction is seen in 60 percent of all teeth.

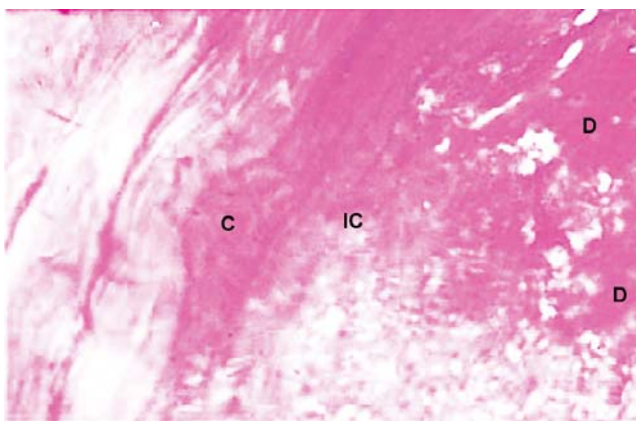


Figure 7.22: Intermediate cementum: Sometimes near the apex of the root, junction between cementum (C) and dentin (D) is occupied by a material having structural characteristics neither of cementum nor of dentin and is called intermediate cementum (IC). As seen in a decalcified tooth (H & E x 100)

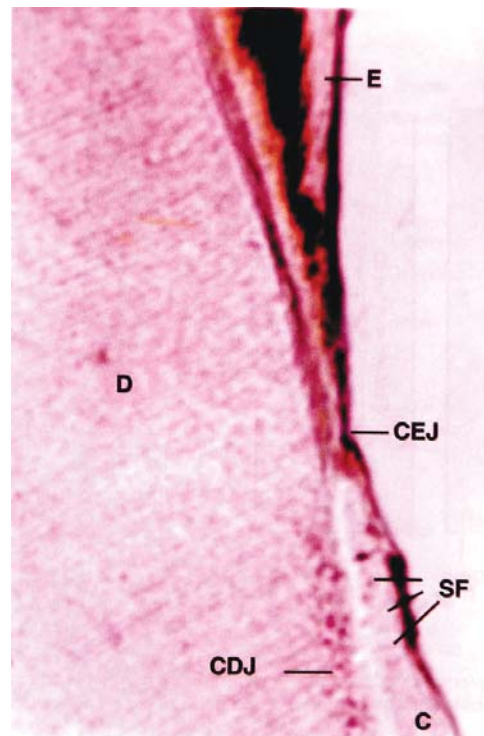
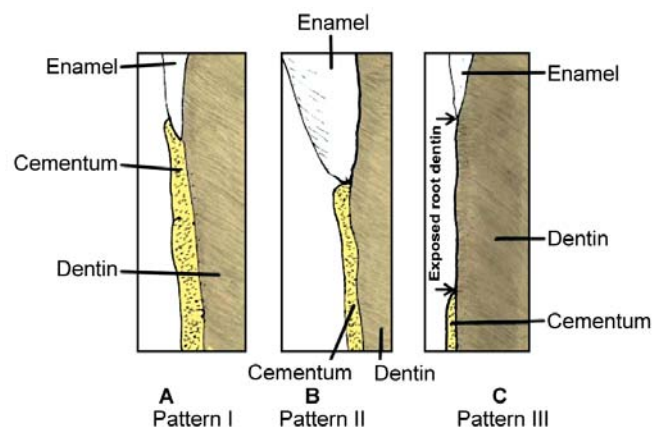


Figure 7.23: Cementoenamel junction (CEJ): Ground section showing slight overlapping of cementum (C) over the Enamel (E). Cementum towards proximal part of root is more acellular in nature. Sharpey's fibers (SF) perpendicular to the cementodentinal junction (CDJ) can be seen (x 150). D = Dentin



Figures 7.24 A to C: Types of cementoenamel junction (A) Cementum overlaps the enamel. (Pattern I). (B) Cementum and enamel meet as sharp line (as a butt joint). (Pattern II). (C) A gap is present between the cementum and enamel exposing root dentin. (Pattern III)

This overlapping occurs due to degeneration of enamel epithelium at their cervical ends. This allows the connective tissue to come in direct contact with enamel surface and differentiate into cementoblasts. The

cementoblasts produce a laminated, electron dense reticular material called afibrillar cementum. Afibrillar cementum is so named because it does not contain collagen fibrils with 64-nanometer periodicity.

Pattern II

In about 30 percent of all the teeth, the cementum and enamel meet as a sharp line (as butt joint) (Figs 7.24B and 7.26).

Pattern III

In about 10 percent of all the teeth, cementum and enamel fails to meet (Figs 7.24 C and 7.27). A small segment of dentin is exposed between the enamel and cementum or it may be covered by reduced enamel epithelium. This

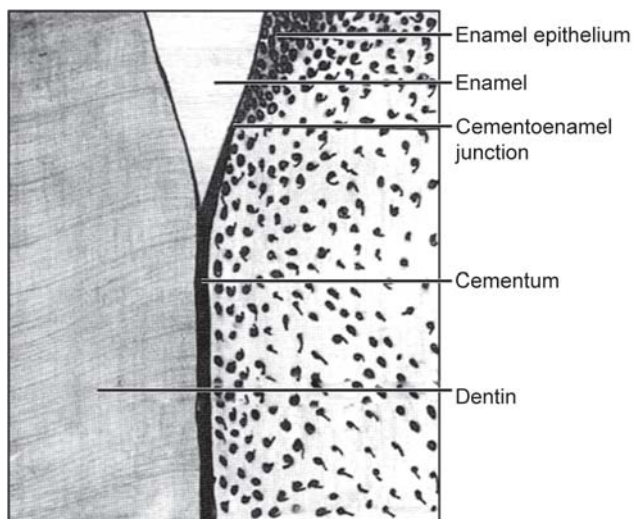


Figure 7.25: Enamel is overlapped by cementum (Pattern I)

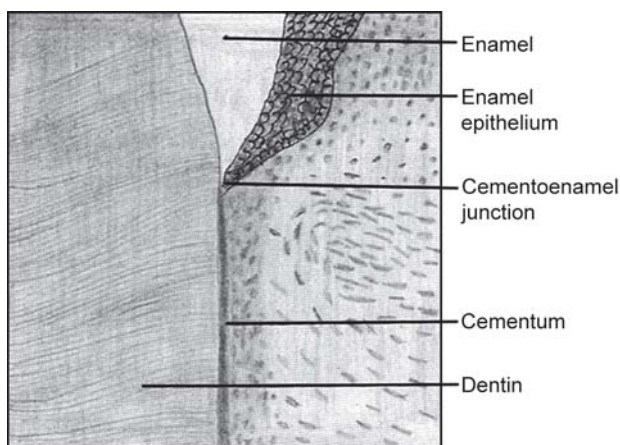


Figure 7.26: Enamel and cementum meeting at sharp line (Pattern II)

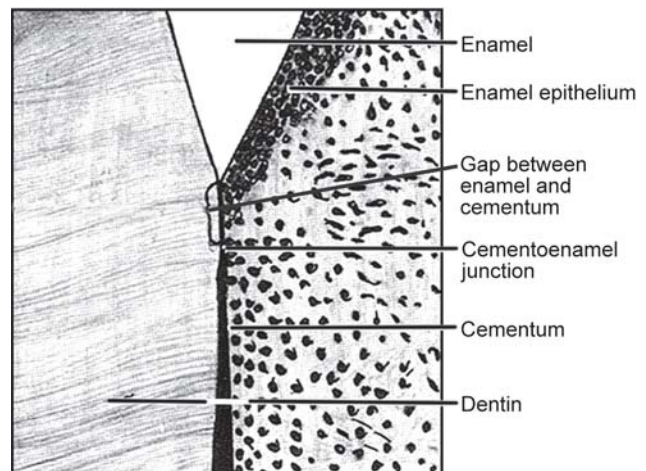


Figure 7.27: There is a gap between enamel and cementum (Pattern III)

condition occurs due to delay in separation of enamel epithelium at the cervical portion of root. In this case cementoenamel junction is not formed.

FUNCTIONS OF CEMENTUM

The following are the functions of cementum:

1. The most important function of cementum is to provide a medium for the attachment to the collagen fibers of periodontal ligament. In hypophosphatasia, which is a hereditary disease, there is loosening and premature loss of anterior deciduous teeth. This is because of the almost total absence of cementum.
2. Cementum is harder than alveolar bone and has no blood supply, and does not show resorption under masticatory or orthodontic forces. Thus, during heavy orthodontic forces, tooth integrity is maintained and alveolar bone being elastic in nature changes its shape, fulfilling the orthodontic requirement.
3. Cementum has property of continuous deposition and does the patchwork or repairs for the damages such as fracture or resorption of root surface.

Regular cementum deposition at the apex of the root helps to replenish the lost tooth height due to occlusal wear or helps in passive eruption of teeth.

CEMENTAL ANOMALIES

Hypercementosis

Hypercementosis means abnormally prominent thickness of the cementum on root surface. This abnormality may affect all the teeth of dentition together or may occur on a single tooth of dentition (Figs 7.28 and 7.29).

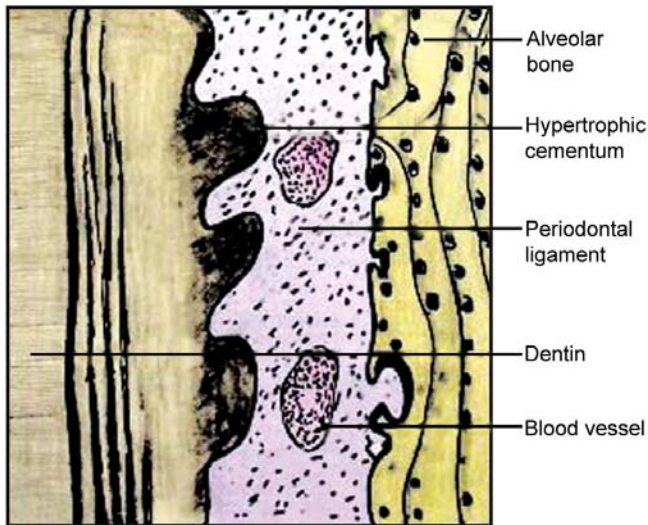


Figure 7.28: Prong-like excementosis

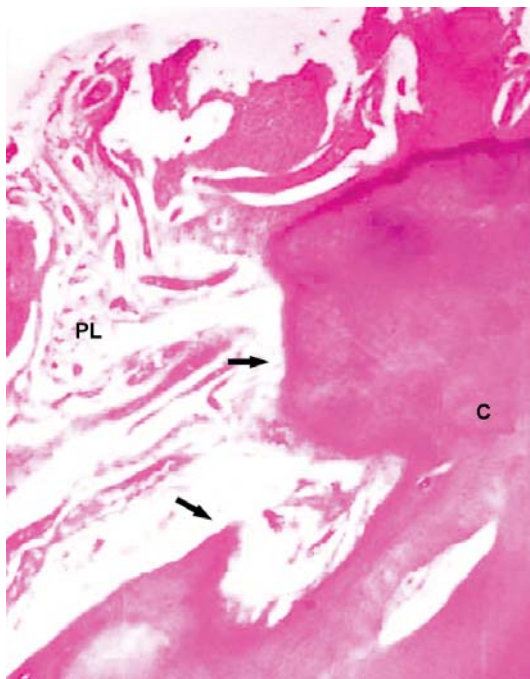


Figure 7.29: Excementosis: Spurs or tongues (indicated by arrows) of cementum (C) project into the periodontal ligament (PL). These are generally seen as a reaction to injury to the tooth. (H & E x 200)

Local and Generalized Hypercementosis

- Localized Hypercementosis is found at a single location anywhere on the root surface. The examples of localized hypercementosis are cemental spikes, and excementosis (Figs 7.30 and 7.31), which is a rounded

projection that develops by deposition of cementum over degenerated epithelial rests on the root surface. Sometimes the embedded calcified round bodies are found in localized areas of hyperplastic cementum. These are also called excementosis.

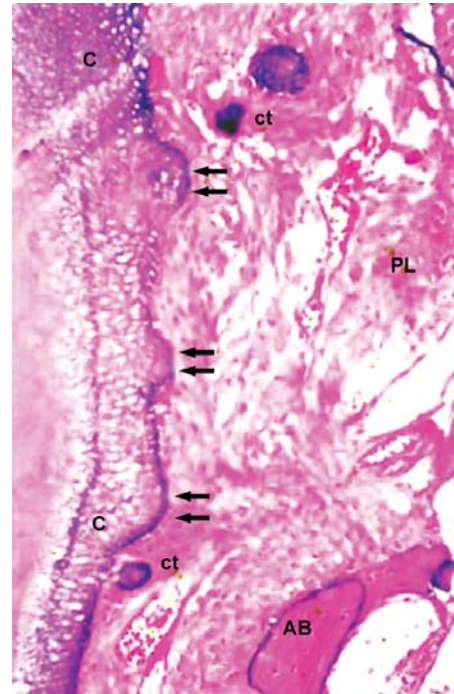


Figure 7.30A: Excementosis: Spurs or protuberances of cementum (C) project into the periodontal ligament (PL). These extensions indicated by double arrows are seen as a reaction to injury to the tooth. Many round calcified cementicles (ct) are lying free in the ligament, AB = Alveolar bone. (H & E x 100)

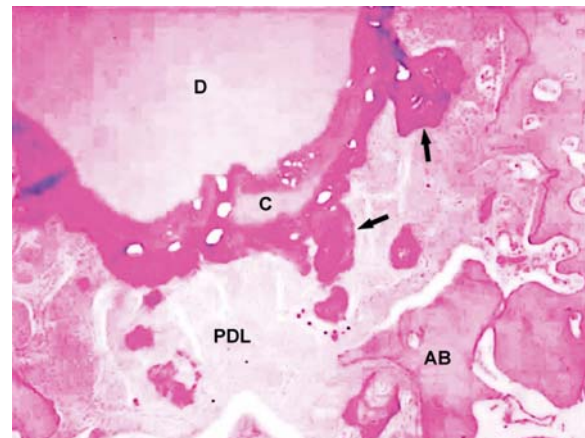


Figure 7.30B: Excementosis: Spurs or protuberances of cementum (C) indicated by arrows projecting into the periodontal ligament (PDL), dentin (D) and alveolar bone (AB) x400

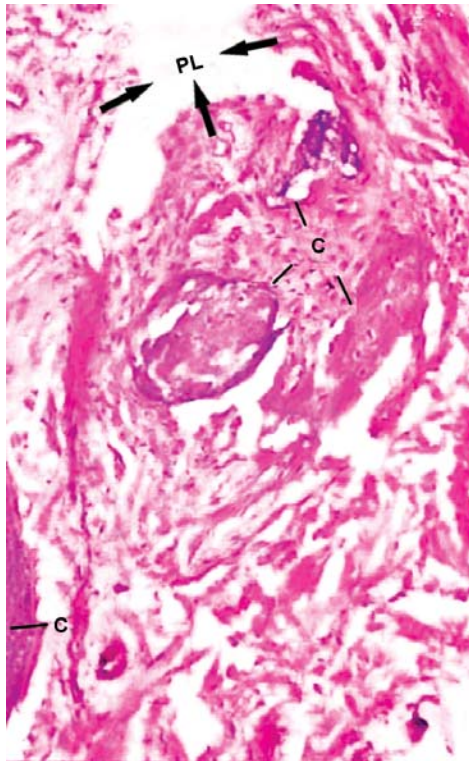


Figure 7.31A: Excementosis: Cross sections of cemental extensions or cementicles present in the periodontal ligament (PL) C = Cementum extensions or cementicles (x 150)

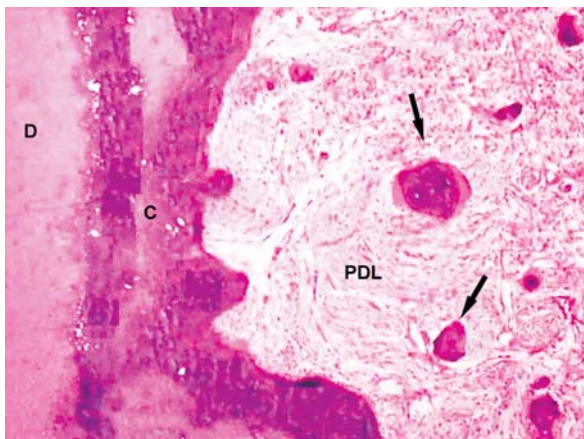


Figure 7.31B: Excementosis: Cemental extensions or cementicles (shown by arrow) present in the periodontal ligament (PDL). Cementum (C) and Dentin (D) x 400

- b. Generalized hypercementosis that involves roots of all the teeth of a dentition is present in certain conditions such as:
- i. Paget's disease of bone [Hereditary]
 - ii. Chronic periapical infection.
 - iii. Non-functional teeth without any antagonist.

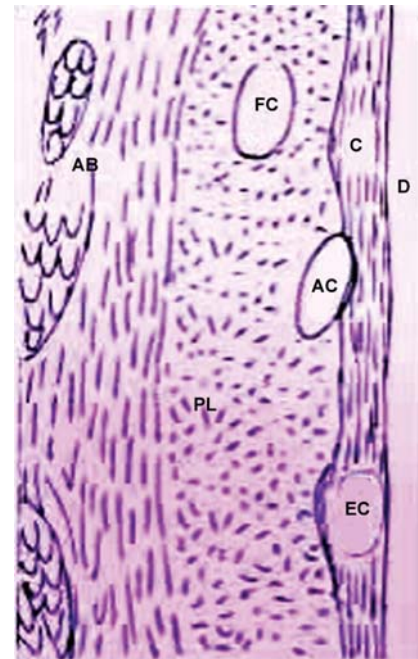


Figure 7.31C: Various types of cementicles in periodontal ligament. FC = Free cementicle. AC = Attached cementicle. EC = Embedded cementicle. AB = Alveolar bone. PL = Periodontal ligament. C = Cementum. D = Dentin

In case of periapical infection, cementum is deposited adjacent to inflamed periapical tissue.

Difference between Cemental Hyperplasia and Hypertrophy

Cemental Hyperplasia Hypercementosis is also called as cemental hyperplasia when cemental growth does not help in increasing functioning of the tooth or it occurs in non-functioning teeth, like hypercementosis due to periapical infection. (Figs 7.32 and 7.33). In non-functioning teeth, hyperplasia is characterized by reduction in the number of Sharpey's fibers embedded in the root.

Cemental Hypertrophy If cemental overgrowth improves or helps in the functioning of teeth, this is referred as cemental hypertrophy. Cemental spike generally develops from extensive occlusal forces or tensional orthodontic force. This provides a greater surface area for the attachment of periodontal fibers (Figs 7.34 and 7.35).

In localized hypertrophy, a spur or prong-like extension of cementum may be formed. This is generally seen in teeth, exposed to great stress. Prong provides a larger surface area for the attaching fibers. As a result, a firmer anchorage of the tooth to the surrounding alveolar bone is formed (Figs 7.28 and 7.29).

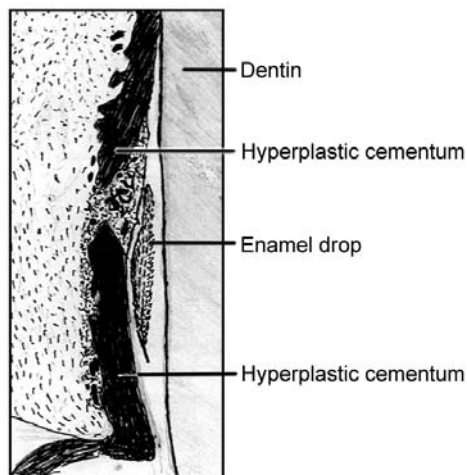


Figure 7.32: Irregular hyperplastic cementum covering the surface of enamel drop

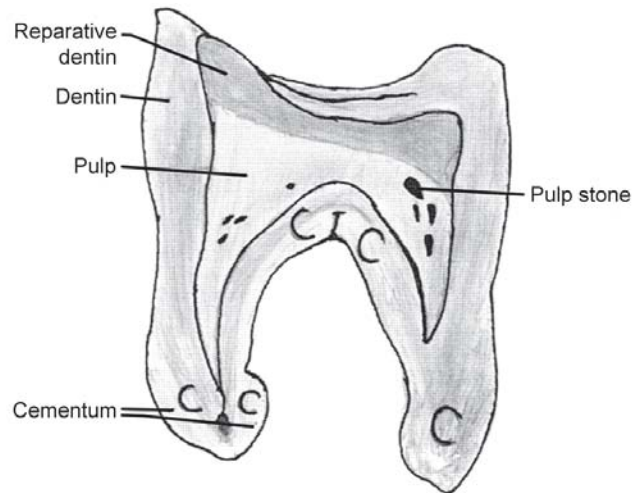


Figure 7.34: Longitudinal section of a demineralized mandibular molar showing hypertrophy of cementum at the furcation area and root apices

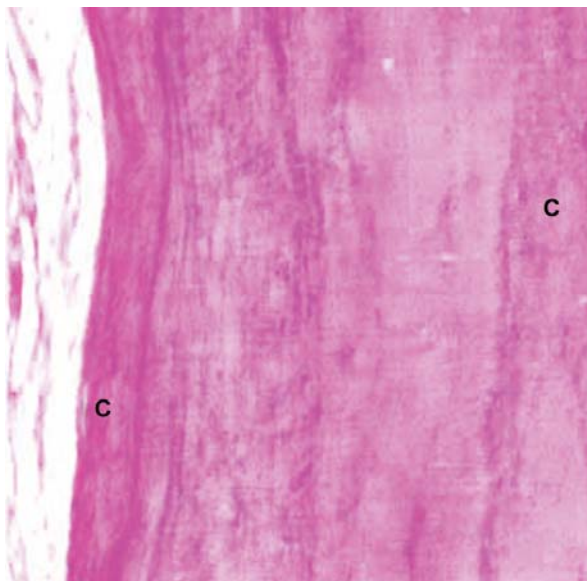


Figure 7.33: Hypercementosis: Extremely thick layer of cementum (C) is found near the root or in non-functional teeth or in tooth under repair (H & E x 100)

Cementicles

Cementicles are round lamellated cemental bodies that lie free in the periodontal ligament space (Figs 7.30 A and B, 7.31A and B, and 7.36) or are attached to the root surface. Cementicles develop around a central nidus, which may be a spicule of bone or cementum or calcified epithelial rests. Mostly they are found in an aging person along the root. They may be found at the site of trauma.

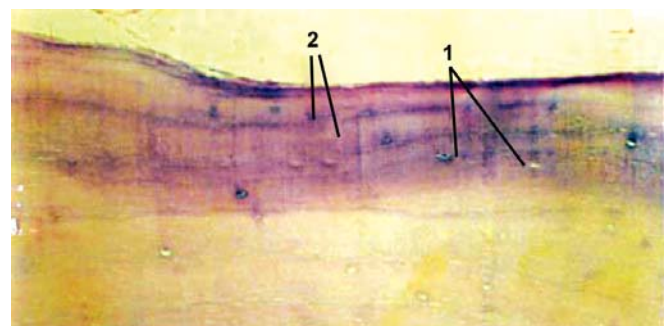


Figure 7.35: Cementum: Decalcified section of tooth: Tooth with hypercementosis showing distinct lines of apposition (2). Lacunar spaces (1) in which cementocytes are located are clearly seen but cell bodies or canaliculi can not be seen in contrast to the ground section

Cementoma

Cementoma is also called benign cementoblastoma or cemental dysplasia. (Figs 7.37 and 7.38). These are cemental masses situated at the apex of the root. These are included in the category of a slowly growing odontogenic neoplasm and may cause expansion of jaw. These are actually true neoplasms of functional cementoblasts.

These occur more in females than males under the age of 25 years and the mandibular first molar is the most common site of development. The surface of cementoma is lined by newly formed cementoid, which is lined by cementoblasts and surrounded by connective tissue capsule.



Figure 7.36: Cementicles lying free in the periodontal ligament and adherent to the root surface

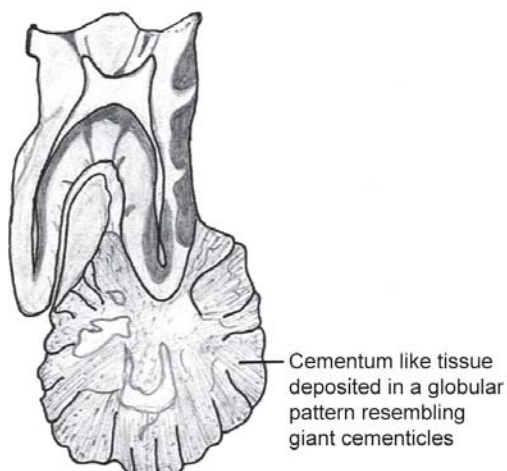


Figure 7.37: Benign cementoblastoma (As seen in a Photomicrograph)

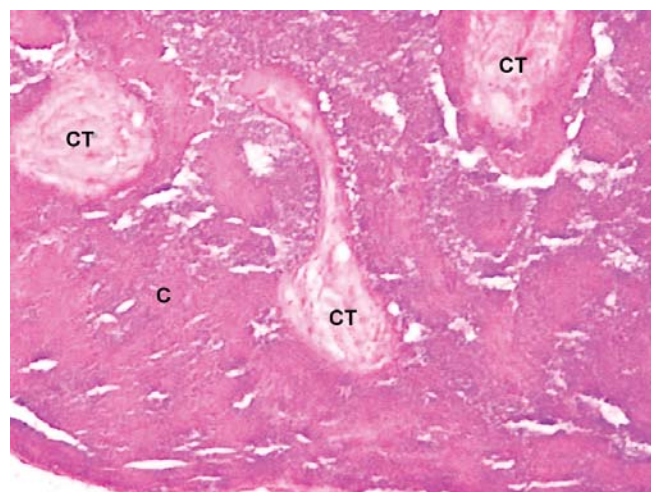


Figure 7.38B: Cemental Dysplasia: Irregular masses of cementum (C) are rich in cementocytes but lack in incremental pattern of growth. Intervening are seen islands of connective tissue (CT) x 100

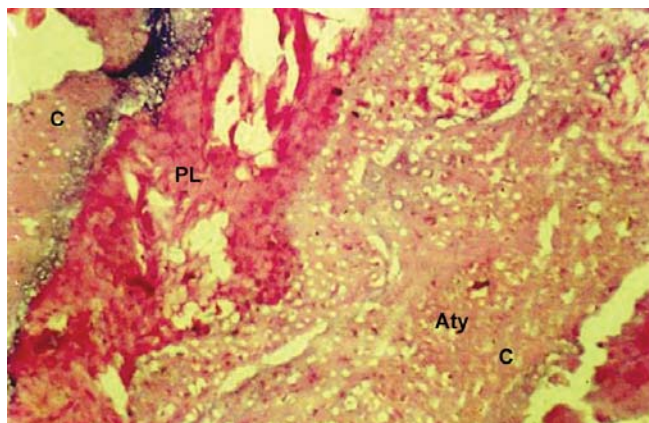


Figure 7.38 A: Cemental dysplasia cementoma / Cementoblastoma: Irregular mass of atypical cementum (Aty) very rich in cementocytes but lacks in incremental pattern of growth is present around the root. C = Cementum, PL = Periodontal ligament x 40

REPAIR AND RESORPTION OF CEMENTUM

Cementum on the root surface undergoes resorption and repair alternately according to the change in environment faced by it. The resorption can be of a very small degree that can be detected only microscopically (Fig. 7.39) or it can be a large concavity that can even be detected radiographically.

Causes of Cemental Resorption

There may be local or systemic causes of cemental resorption. These are as follows:



Figure 7.39: Resorption of cementum: Ground section showing irregular resorption of cementum (C) as irregular concavities. D= Dentin (x 100)

Local Causes

Local conditions which give rise to cemental resorption are as follows:

1. Trauma from occlusion
2. Cysts and tumors
3. Periapical pathology
4. Excessive orthodontic force
5. Embedded teeth
6. Replanted and transplanted teeth.

Systemic Causes

Systemic conditions causing or inducing cemental resorption are as follows:

1. Deficiency of calcium
2. Deficiency of vitamin A and D.
3. Hypothyroidism

Types of Cemental Repair

Repair of cementum is a process to heal the damage caused by resorption or cemental fracture (Fig. 7.40). Repair may be anatomic or functional.

Anatomic Repair

In anatomic repair the root outline is re-established as it was before cemental resorption. It generally occurs when the degree of destruction is low (Figs 7.41 A to C).

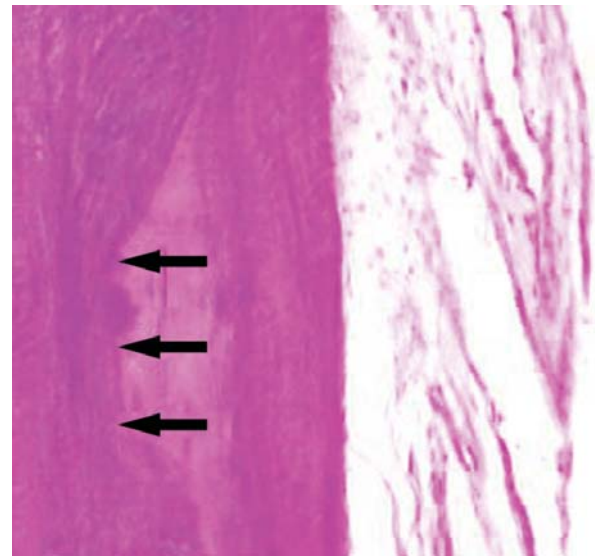


Figure 7.40: Repair of the resorbed cementum: Resorption of cementum leads to depressed or scalloped cemental margin indicated by arrows. The defect is filled with newly laid down layers of cementum (H & E x 200)

Functional Repair

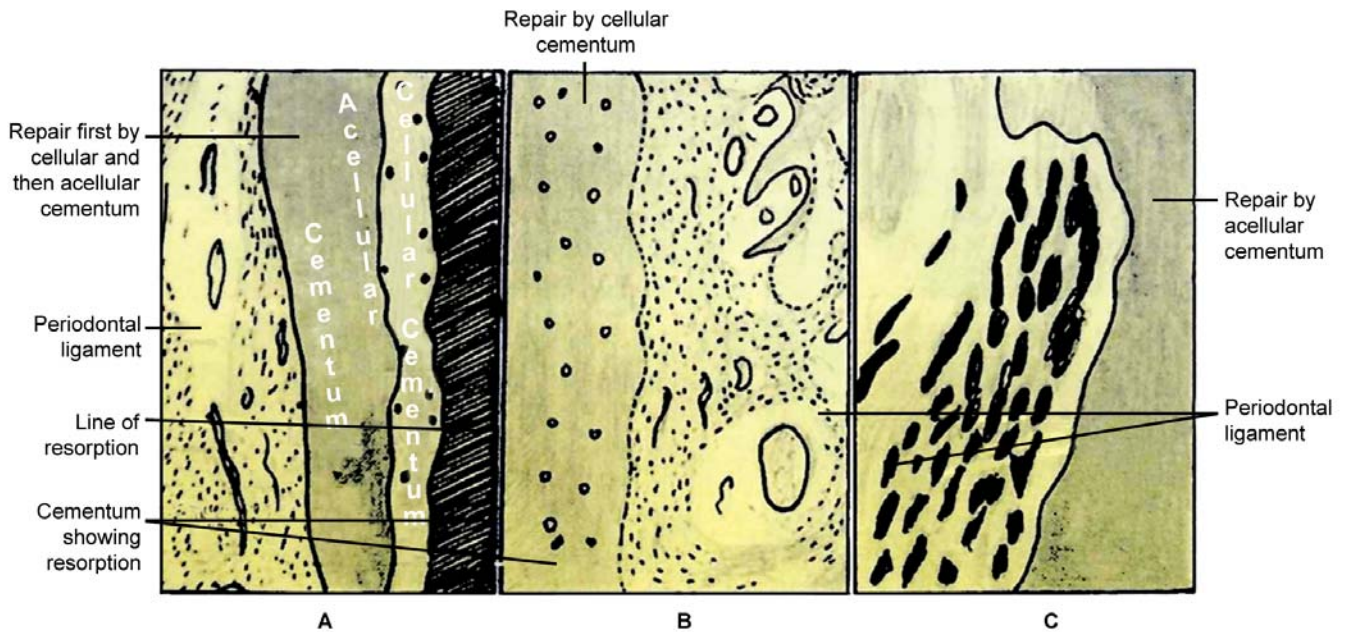
In the cases of large cemental resorption or destruction, repair does not re-establish the same anatomic contour as before, because only thin layers of acellular and cellular cementum are deposited over the concavity created by cemental resorption. To maintain the width of periodontal ligament, the adjacent alveolar bone grows and takes the shape of defect following the root surface (Figs 7.42 and 7.43). This is done to improve the function of tooth, thus called functional repair.

Histology of Repair and Resorption

The microscopic appearance of the area showing root resorption appears as bay-like cavity on the root surface (Fig. 7.44) and is surrounded by large mononuclear macrophages.

Periods of repair and deposition of new cementum alternate with the periods of resorption. Thus, resorption is not a continuous process. Reversal lines separate newly deposited cementum from old cementum and it lines the border of the resorption concavity. Periodontal fibers get attached to the newly laid cementum and maintain the functional relation.

It is important to note that the presence of viable periodontal ligament is necessary for cementum deposition and both vital and nonvital teeth can undergo cemental resorption.



Figures 7.41 A to C: Cementum resorption is repaired by; A. First by cellular and then acellular cementum, B. Cellular cementum, C. Acellular cementum

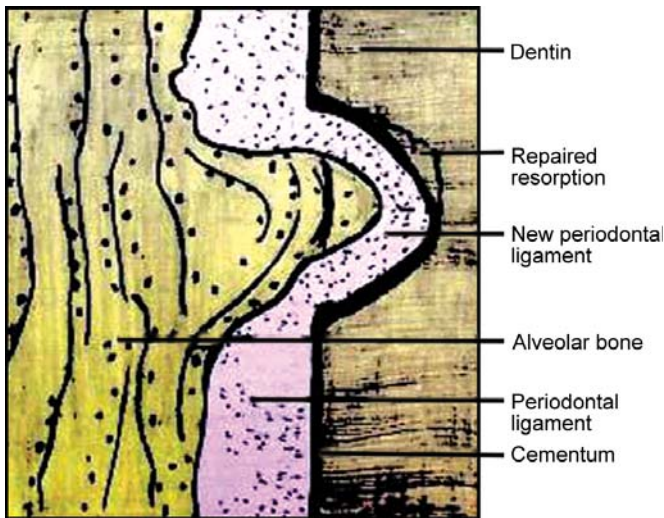


Figure 7.42: Normal width of periodontal ligament is re-established after functional repair of cementum resorption by bone deposition

AGING OF CEMENTUM

Due to aging, the smooth surface of the cementum becomes rough, due to calcification of some bundles of ligament fibers, at the place of their attachment to cementum. This happens on almost all cemental surfaces except in apical area. In apical area, due to aging, continuous cementum

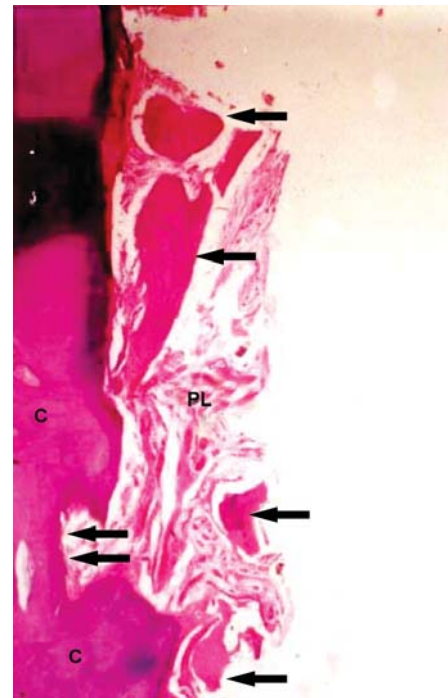


Figure 7.43: Functional repair of cementum (C) by proliferation of ossified trabeculae of alveolar bone indicated by single arrows. Defect in cementum is shown by double arrows. PL = Periodontal ligament (H & E x 100)

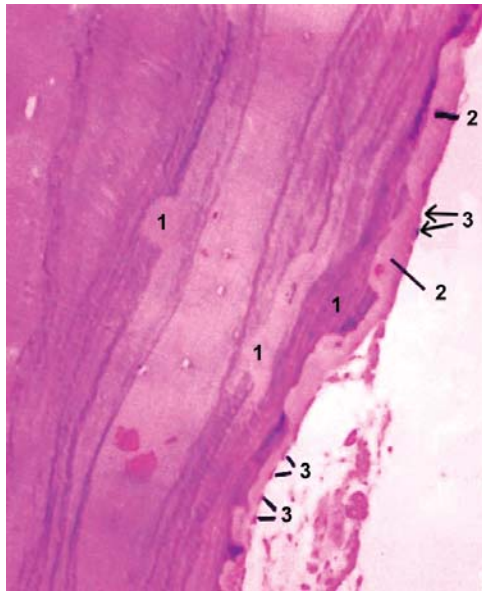


Figure 7.44: Erosion of the cementum is seen as irregularly scalloped or eaten up appearance of the normal mineralized cementum (1) repaired by pinkish cementoid (2). Thin layer of cementoblasts (3) can be seen on the surface of cementoid (As seen in decalcified tooth). (H & E x 100)

deposition takes place, which sometimes may lead to closure of the apical foramen (Fig. 7.45). Due to aging, cementum can also resorb for some time and deposition can also take place for some time creating reversal lines.

CLINICAL CONSIDERATIONS

Ankylosis

When there is fusion of cementum and alveolar bone with no periodontal ligament in between, it is known as ankylosis.

Causes of Ankylosis

1. Replantation and transplantation of teeth in which periodontal ligament is damaged.
2. Embedded teeth
3. Chronic periapical infection
4. Trauma to deciduous teeth - leading to destruction of dental follicle of underlying developing permanent tooth germ, causing its ankylosis. [Dental follicle is responsible for the formation of periodontium]

In ankylosis, there is gradual progressive resorption of root and its replacement by bone.

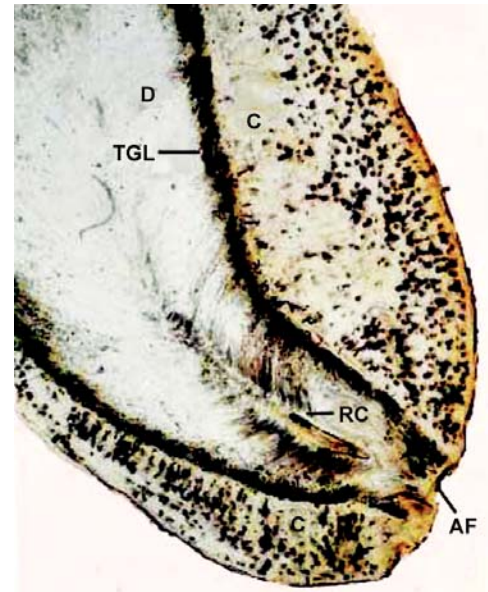


Figure 7.45: Ground section of tooth apex of root showing apical foramen: part of root canal (RC) closed by layer of cementum (C) at the apical foramen (AF). Cementum at the apex is thick and cellular. Tomes' granular layer (TGL) between dentin (D) and cementum is clearly visible

ORTHODONTIC TOOTH MOVEMENT

In proper orthodontic treatment, bone resorption and formation result in tooth movement while cementum resorption is much less or absent. The bone is more vascular and generative degenerative processes are much more rapid in bone while cementum is avascular and generative and degenerative processes are much slower in cementum.

Extraction of Teeth

Before performing extraction of any firm tooth, its radiograph should be taken. This is done to examine if there is ankylosis of tooth, hyperplasia of cementum, presence of cementoma or excementosis. Presence of any of the above abnormalities attaches the tooth very tightly to bone and while attempting extraction, may cause fracture of the tooth and or bone.

Fracture of Root

When tooth receives an accidental blow or accidentally bites on very hard object, root may fracture. This fracture may be horizontal or vertical. The tooth with vertical fracture

has poor prognosis and usually it cannot be repaired by cementum easily. It should be extracted or stabilized by intracoronal splinting or banding.

The tooth with horizontal fracture, depending upon the location of fracture line and age of patient, has variable prognosis. If fracture is at the apical and middle third of root in young patient, then it can be repaired by cementum and prognosis of the vitality of pulp of the tooth for survival is fair. But if the horizontal fracture is at the coronal third, then prognosis for the continued vitality of the pulp of the tooth is poor.

Importance of Root Planing in Periodontal Treatment

Root planing is a treatment procedure to remove calculus and necrotic cementum and smoothening the root surface in order to reduce the pocket depth. In the case of deep pockets, surface of cementum exposed in pockets becomes hypermineralized and endotoxins produced by plaque bacteria are incorporated into cementum. These endotoxins cause structural changes in cementum and may interfere in healing during periodontal treatment. Root planing removes hypermineralized necrotic cementum producing a clean and fresh surface for healing of pocket.

Cellular cementum is similar to bone but it does not contain any nerves. Hence, cementum is not sensitive. If the cementum is removed the underlying dentin is exposed, this may result in sensitivity. Repair of cementum is a very important defensive mechanism. Cementum is resistant to resorption under mild pressure. Resorption of alveolar bone and not of cementum makes orthodontic tooth movement possible.

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Periodontal Ligament

- Introduction
- Evolution
- Development
- Cells
- Extracellular substance
- Structures present in the connective tissue
 - Blood vessels
 - Lymphatics
 - Nerves

- Functions
 - Supportive function
 - Sensory function
 - Nutritive function
 - Formative function
 - Homeostatic function
- Aging of ligament
- Clinical considerations

INTRODUCTION

Periodontal ligament is the integral part of the periodontium. The periodontium is an attachment apparatus of the teeth. It is a connective tissue organ, which is covered by epithelium on top surface. Teeth are attached to the bones of the jaws by periodontium. It consists of four connective tissues: (a) cementum, (b) periodontal ligament, (c) the bone that lines the alveolus, and (d) the deeper part of the gingiva. Two of these tissues are the mineralized and two are fibrous. The cementum and the alveolar bone are the mineralized tissues while the periodontal ligament and the gingiva are fibrous tissues. On one side, the periodontium is attached to the dentin of the root of the tooth by cementum while on the other side it is attached to the bone of the jaws by the alveolar bone (Figs 8.1 and 8.2).

The periodontal ligament is soft, fibrous specialized connective tissue. It is present in the periodontal space, which is situated between the cementum of the root of the tooth and the bone forming the socket wall. The periodontal ligament extends coronally up to the most apical part of the connective tissue of the gingiva. Because the collagen fibers are attached to cementum and alveolar bone, the ligament provides a soft tissue continuity between the mineralized connective tissues of the periodontium. The average thickness of periodontal ligament is 0.15 to 0.38 mm. Its appearance is an hour glass appearance because ligament is thinnest at the middle third of the root.

The periodontal ligament is a cellular connective tissue. It consists of the blood vessels, various cells and

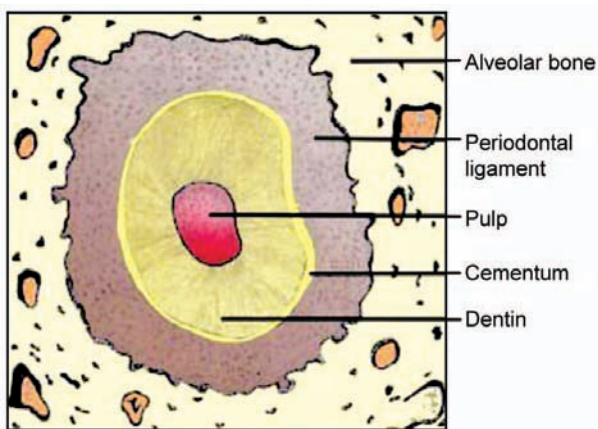


Figure 8.1: Transverse section of root showing relationship between pulp, dentin, cementum, periodontal ligament and alveolar bone

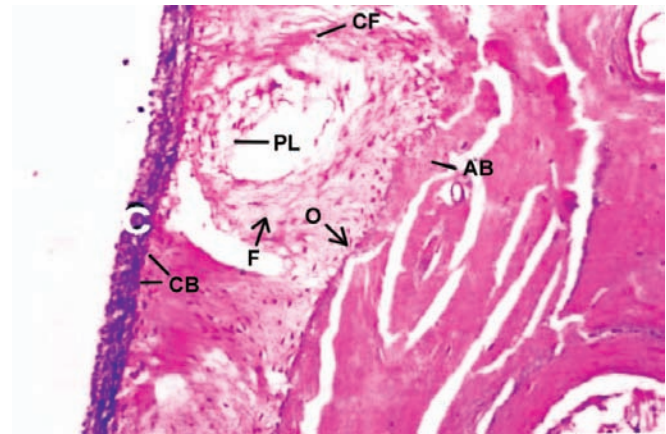


Figure 8.2: Periodontal ligament (PL): Periodontal tissue occupies periodontal space which is in between cementum (C) and alveolar bone (AB) (x 150). CB = Cementoblasts, O = Osteoblasts, CF = Collagen Fibers, F = Fibroblasts

extracellular matrix (Fig. 8.2). Extracellular matrix consists of fibers and the ground substance. The majority of the fibers are collagen and the matrix consists of macromolecules, which are proteins and polysaccharides. The average thickness of periodontal ligament in young adult is 0.21 mm, in mature adult is 0.18 mm and in older adult is 0.15 mm.

Other terms which were previously used for periodontal ligament were (a) desmodont, (b) gomphosis, (c) pericementum, (d) dental periosteum, (e) alveolodental ligament, and (f) periodontal membrane. On radiograph, periodontal ligament appears as a radiolucent area of 0.4 to 1.5 mm, thickness between the radiopaque cementum and radiopaque lamina dura of the alveolar bone.

EVOLUTION

A series of coordinated changes in the jaws have occurred during an evolutionary step from reptiles to mammals. In reptiles, the teeth are ankylosed to the bone whereas in mammals, the teeth are suspended by the ligaments in their sockets. So there is a gradual change seen in the mode of attachment of the tooth, from the bone to periodontal ligament.

There is a radical reconstruction of the mandible from reptiles to the mammals. In reptiles, the bones of the mandible are joined by the sutures. The dentary, which is the upper bone, carries the ankylosed teeth. The mandibular articulation with the cranium takes place by the articulare, which is the separate bone of the mandible and the quadratum, which is a separate bone of the cranium.

During evolution, when advanced type of reptiles changed to mammals, the dentary occupied larger part and other mandibular bones were reduced in size. At last, mammalian mandible was formed only by the dentary. The other bony components of the reptilian mandible were either lost or changed into two ossicles of the middle ear. The articular changed into the malleus and the quadratum changed into the incus. Along with this, the dentary formed a new temporomandibular articulation.

There also occurred a change in the pattern of growth. In reptiles, growth of the mandible is sutural while in mammals; the cartilage of condyle becomes the growth site of mandible. The growth of the mandibular height in reptiles, occurs in mandibular sutures while in mammals, the growth occurs at the free margin of the alveolar process. In the reptiles, the teeth move with the bone while in mammals, the teeth move as a unit. This is because of the remodeling of the periodontium.

Thus, during the evolution from the reptiles to the mammals, there is a replacement of the ankylosis of the tooth and bone to a ligamentous suspension of the tooth. Because of this, movement of the mammalian teeth is made possible, resulting in continual repositioning as required by the jaw growth and also tooth wear.

DEVELOPMENT

Shortly after the beginning of root formation and the formation of the outer dentinal layer of root, the periodontal ligament is formed. The external and internal dental epithelia proliferate from the cervical loop of dental organ to form a Hertwig's epithelial root sheath. This sheath is double layered (Fig. 8.3). Because of the growth changes, the root sheath is stretched and it fragments to form the discrete clusters of the epithelial cells called as 'epithelial cell rests of Malassez' 'or' 'epithelial rests of Malassez'. Now the periodontal ligament formation occurs (Fig. 8.4). The enamel organ and Hertwig's epithelial root sheath are surrounded by a dental sac that is formed by condensed cells. A thin layer of these cells lies adjacent to the dental (enamel) organ. This is known as dental follicle. The cells of the dental follicle divide and differentiate into the cementoblasts, fibroblasts and osteoblasts (Fig. 8.2). The fibroblasts synthesize the fibers and ground substance of the periodontal ligament. These fibers of periodontal ligament then get embedded at one end into the newly formed cementum laid by cementocytes and at the other end into the bone laid by osteoblasts (Fig. 6.6).

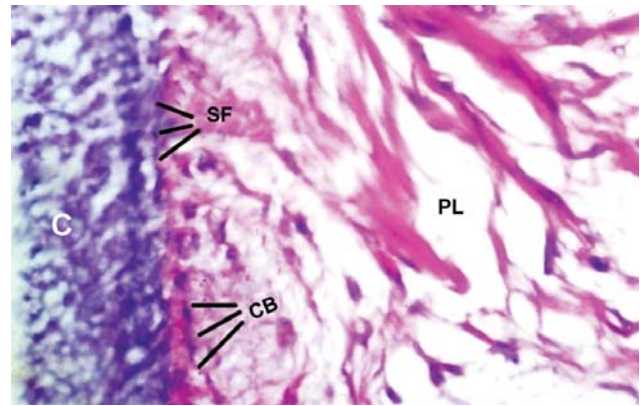


Figure 8.3: Tooth germ showing tooth follicle which is continuous with dental papilla around the cervical loop

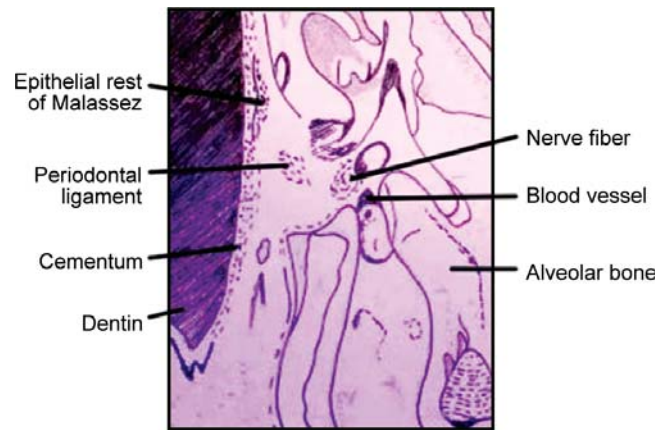


Figure 8.4: Apical region of a developing root of the tooth

When tooth erupts in the oral cavity only the alveolar crest fibers of periodontal ligament are histologically distinguishable. Later fine brush like fibers extending from the cementum and alveolar process, elongate towards each other, ultimately fuse and crosslinking of individual collagen molecular units occur. When tooth erupts in oral cavity, these fibers get oriented in a characteristic manner. The fiber bundles of the periodontal ligament gradually thicken after the teeth have been in function for some time. At the time of first occlusal contact of the tooth, with its antagonist, horizontal fibers are completely developed and when definite occlusion is established, oblique fiber bundles are matured followed by the formation of apical fiber group.

The damaged periodontal fibers are replaced and remodeled by newly formed fibers. The renewal capability is an important characteristic of periodontal ligament. The fibroblasts of the periodontal ligament provide maintenance of the system when repair is required.

CELLS

Various types of cell are found in a functioning periodontal ligament. This helps in the synthesis and resorption of the alveolar bone and fibrous connective tissue of the ligament and the cementum (Table 8.1). The cells of the periodontal ligament may be divided into following types.

Table 8.1: The proportion of the cells of periodontal ligament in mouse molar teeth

Element (Cells)	Proportion (%)
Fibroblasts	35
Collagen	51
Vessels	10
Nerves	1 (95% unmyelinated)
Oxytalan fibers	0.45
Other cells	2.55

Synthetic Cells

In the periodontal ligament, synthetic cells are present in all stages of activity. These synthetic cells produce proteins. It means that they transcribe ribonucleic acid (RNA), synthesize ribosomes in the nucleolus and transport them to cytoplasm. The rough endoplasmic reticulum (RER) and Golgi apparatus increase translation and transport of the proteins.

These functional activities are reflected morphologically under the light and electron microscope. A large open faced or vesicular nucleus which contains prominent nucleoli reflects increased transcription of RNA and production of the ribosomes.

Large quantities of RER (rough endoplasmic reticulum) are recognized in the electron microscope. Under light microscope, these are reflected after staining with hematoxylin and eosin. Golgi bodies and vesicles are seen in electron microscope. Under light microscope they are seen as clear, unstained area in hematoxylin stained cytoplasm.

Large numbers of mitochondria under electron microscope reflect increased requirement of energy. To accommodate all these organelles, a large amount of cytoplasm is present. Thus the synthetic cells of periodontal ligament under light microscope are seen to contain a large, open-faced nucleus, with prominent nucleoli and an abundant hematoxylophilic cytoplasm. The cells of

periodontal ligament synthesize the alveolar bone, cementum and fibrous connective tissue. The cells are as follows:

Osteoblasts

Osteoblasts are derived from the multipotent mesenchymal cells. They cover the periodontal surface of the alveolar bone.

These constitute the modified endosteum. The surface of the bone lining a socket is regarded as an interior surface of the bone. This surface of the bone is covered by the osteoblasts in various stages. The collagen fibers of ligament that penetrate the alveolar bone are present between the cells. Osteoblasts are basophilic cuboidal cells, with abundant rough endoplasmic reticulum, mitochondria and vesicles. These cells contact one another through tight junctions and desmosomes. Round prominent nucleus is placed at the basal end of the cell and microfilaments are prominent at the secreting surface.

Fibroblasts

Fibroblasts are found in the periodontal ligament surrounded by fibers and ground substance (Fig. 8.2). The fibroblasts are arranged parallel to the collagen fibers. Fibroblasts are the principal cells of the periodontal ligaments. Fibroblasts near the alveolar bone are originated from the perivascular mesenchyme and fibroblasts near cementum are originated from ectomeschymal cells of dental papilla. Fibroblasts synthesize and shape the proteins of the extracellular matrix and attach to the substratum of the matrix through surface receptors for fibronectin and collagen. Fibroblast are characterized by very fast turnover of collagen in the periodontal ligament.

Fibroblasts are large fusiform cells with well developed cytoskeleton, extensive cytoplasm, abundant organelles, large nucleus, one or more prominent nucleoli and long, thin cytoplasmic extension. Fibroblasts show adherens and gap junction.

The functions of fibroblasts are as follows:

1. Formation and remodeling of periodontal ligament fibers,
2. Maintain the width of the periodontal ligament,
3. Production of collagen, elastin, structural connective tissue proteins, glycoproteins and glycosaminoglycans,

4. Secretion of active collagenase and enzyme matrix metalloproteinases.

Cementoblasts

Cementoblasts are found on the surface of the cementum. They are distributed in the same manner as the osteoblasts are on bone surface (Figs 8.2 to 8.6). Cementoblasts are cuboidal cells with abundant cytoplasm, large vesicular nucleus, one or more nucleoli and abundant mitochondria. All the organelles related with synthesis and secretion of protein are also present in the cementoblast. Receptors for growth hormone and epidermal growth factor are present.

Prominent cytoplasmic processes are present in cementoblast depositing cellular cementum and absent in cementoblast depositing acellular cementum. Cells contact one another through desmosomes and gap junctions.

Resorptive cells

Osteoclasts

Multinucleated osteoclasts are the cells which resorb the bone. Under light microscope, the osteoclasts are seen to be present in the shallow depression known as Howship's lacunae (Fig. 8.7). When viewed under electron microscope, these cells exhibit numerous mitochondria, lysosomes, abundant Golgi saccules and free ribosomes, but little rough endoplasmic reticulum. The surface of an osteoclast which is in contact with the bone has a ruffled border. The presence of osteoclasts indicates resorption of the alveolar bone. Histochemical test shows the presence of acid phosphatase in lysosomes.

A zone of specialized membrane, which is closely applied to the bone, is present between the ruffled border and the plasma membrane. The cytoplasm below this membrane lacks the organelles. This is known as 'clear zone'.

Resorption occurs in two stages. At first the mineral is removed at bone margins, and then exposed organic matrix disintegrates. The osteoclasts demineralize the inorganic part as well as disintegrate the organic matrix.

In an inactive osteoclast, the ruffled border is not present. Osteoclasts are normally seen in a functioning periodontal ligament, as it helps in remodeling. The remodeling process allows functional changes in the position of tooth.

Fibroblasts

Fibroblast cells are capable of both synthesis and resorption. The collagen fibers of periodontal ligament can be resorbed by the mononuclear fibroblasts. These fibroblasts are present in the periodontal ligament for the remodeling process. They exhibit lysosomes, which contain collagen fragments undergoing digestion (Fig. 8.8). The presence of collagen resorbing fibroblast in a normal functioning periodontal ligament indicates resorption of fibers occurring during remodeling of periodontal ligament. Collagen degradation is both extracellular and intracellular event.

- I. *Extracellular degradation of collagen* Extracellular degradation of collagen involves the collagenase enzyme. Stromelysin (MMP-III) remove the fibronectin and proteoglycans present on surface of the fibrils which mask the binding site of the collagenase. Collagenase enzyme breaks the triple helical portion of molecules within the fibrils. Collagenase and MMP- IV denature the collagen under physiological condition. MMP-II and MMP-V degrade the remaining molecule by proteolysis.
- II. *Intracellular degradation of collagen* Lysosomal enzymes are involved in the intracellular degradation of collagen. The sequence of intracellular degradation of collagen is as follows:
 1. Phagocytosis of collagen fibrils by the fibroblast.
 2. Banded fibrils are surrounded by electron-dense zone.
 3. Phagolysosome is formed by the fusion of phagosome with primary lysosomes.
 4. Indistinct banding of the fibrils.
 5. Enzymatic degradation of the fibrils.
 6. Fibrilloses its characteristic structure.

Cementoclasts

Cementoclasts are found in the periodontal ligament but the cementum is not remodeled like alveolar bone and periodontal ligament. Under certain conditions, the resorption of cementum can occur. The mononuclear cementoclasts or multinucleated giant cells are located in Howship's lacunae. These are found on surface of the cementum.

Progenitor Cells

These are the undifferentiated mesenchymal cells. They constitute an important cellular component of the

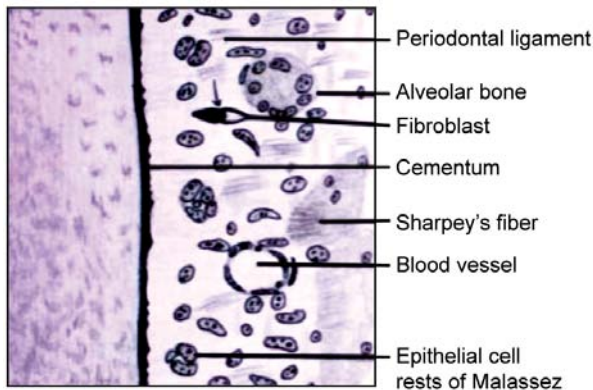


Figure 8.5: Section of periodontal ligament

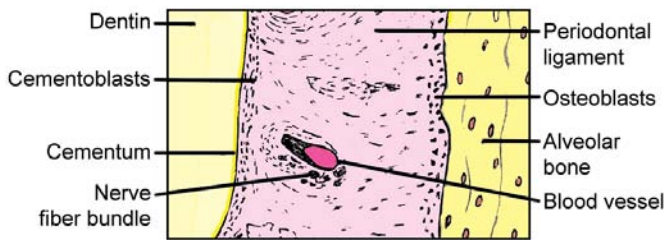


Figure 8.6: Midroot region in a transverse section of tooth

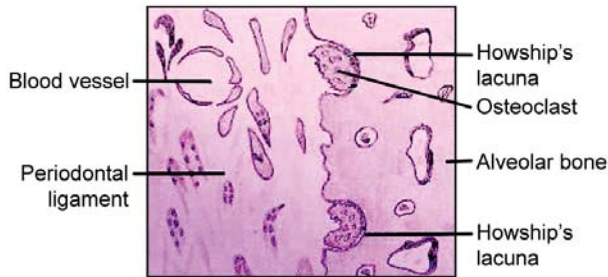


Figure 8.7: Periodontal ligament showing osteoclasts located on the surface of alveolar bone in Howship's lacunae

periodontal ligament. They have the capacity to undergo mitotic division and replace the differentiated cells dying at the end of their life span. The progenitor cells are located in perivascular region. These cells have a small close-faced nucleus and a very little cytoplasm. When cell division occurs, one of the daughter cells differentiates into functional type of connective tissue cells. The other remaining cells retain their capacity to divide. Progenitor cells of fibroblast are small, less polarized and contain less rough endoplasmic reticulum and Golgi saccules. Cells of osteoblast subtype contain high levels of alkaline phosphatase.

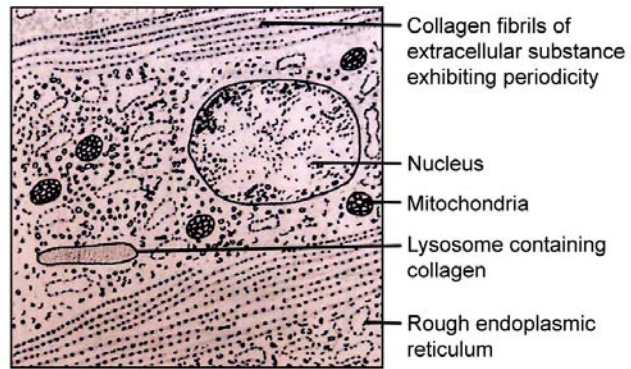


Figure 8.8: Periodontal ligament exhibits numerous profiles of rough endoplasmic reticulum, Golgi membrane and collagen containing lysosome (as seen under Electron Microscope)

In orthodontic therapy, after application of force to a tooth or after wounding, burst of mitosis occurs in the progenitor cells. This stimulates the proliferation and differentiation of cells of periodontal ligament. From the perivascular area these cells enter the periodontal ligament.

Mesenchymal Stem Cells

Mesenchymal stem cells in the periodontal ligament perform the following functions.

1. Tissue homeostasis.
2. Source of renewable progenitor cells. These cells can generate the cementoblasts, osteoblasts and fibroblasts.

Epithelial Rests of Malassez (Epithelial Cell Rests of Malassez)

Epithelial rests of Malassez were first described by Malassez in 1884 and are found close to the cementum. These cells are remnants of the epithelium of Hertwig's epithelial root sheath. Epithelial rests of Malassez are arranged parallel to the root surface and persist as a network, strands, islands or tubule-like structures. These epithelial rests are formed at the time of cementum formation. The continuous layer of root sheath breaks into lace-like strands. The electron microscopic observations revealed that these cells exhibit the tonofilaments and are attached to each other by desmosomes. These cells are abundant in the furcation areas. Cell rests are cuboidal with prominent nucleus, scanty cytoplasm, well distributed mitochondria and poorly developed rough endoplasmic reticulum and Golgi apparatus.

These cells may form cementicles by calcification or proliferate to form cysts or tumors. More cells are observed in the children and up to second decade of life are commonly located in the apical region. Later on after second decade they are found in the gingiva above the alveolar crest.

When a longitudinal or transverse section of a tooth is seen, the strands of the network appear as the isolated islands. The epithelial cells are isolated from the connective tissue cells by a basal lamina. Cells of the epithelial rests can undergo rapid proliferation and can produce a variety of cysts and tumors when certain pathologic conditions are present (Figs 8.9 to 8.14).

CELL BIOLOGY OF NORMAL PERIODONTIUM

Growth factors and cytokines play important role in tissue homeostasis and pathogenesis of periodontal disease. Important factors are as follows.

Transforming Growth Factor (TGF)

Transforming growth factor causes change in the normal cell growth TGF is of two types.

- A. TGF- α - synthesized primarily by malignant cells and effect is similar to the biological effects.
- B. TGF- β - synthesized by normal cells and its functions are as follows:
 - a. Reduce the level of collagenase expression.

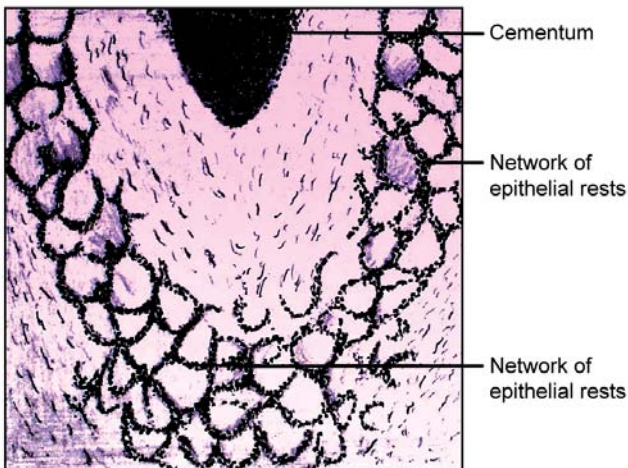


Figure 8.9: Periodontal ligament with network of epithelial rests of Malassez (Tangential section parallel to root surface)

- b. Stimulate the synthesis of connective tissue matrix.
- c. Enhance the induction of tissue inhibitors of matrix metalloproteinases.

Fibroblast Growth Factor (FGF)

FGF is a potent stimulator of mitogenesis and cell migration of periodontium. FGF can be acidic or basic in nature.

- A. Functions of acidic fibroblast growth factor (AFGF)
 - a. In bone tissue culture, AFGF stimulates DNA synthesis and cell replication.
 - b. Effect the neovascularization.
- B. Functions of basic fibroblast growth factor
 - a. Increase cells of osteoblastic lineage
 - b. Stimulate replication of bone cells.

Matrix Metalloproteinases (MMPs)

MMPs are proteolytic enzymes, important for remodeling and degradation of extracellular matrix components. MMPs are found in macrophages, neutrophils, osteoblast, osteoclasts, epithelial cells, fibroblast and also produced by periodontal pathogens.

Platelet Derived Growth Factor (PDGF)

PDGF is secreted from the α granules of platelets. Its receptors are located on surface of various cells like fibroblasts, glial cells and vascular smooth muscle cells. Functions of PDGF are as follows.

- A. Potent mitogen for various cells
- B. Promote the migration of cells
- C. Stimulate the formation of type V collagen
- D. In bone culture, increases the proliferation of cells
- E. Increases the bone resorption.

Interferon- γ

Interferon- γ is important for immunomodulatory effect. Its production is modulated by cytokines.

Interferon- γ affects the production of collagen by turning off the collagen gene expression.

Interferon- γ is important for the activity of various cells like T lymphocyte, B lymphocyte, Natural killer cells, macrophages, etc.

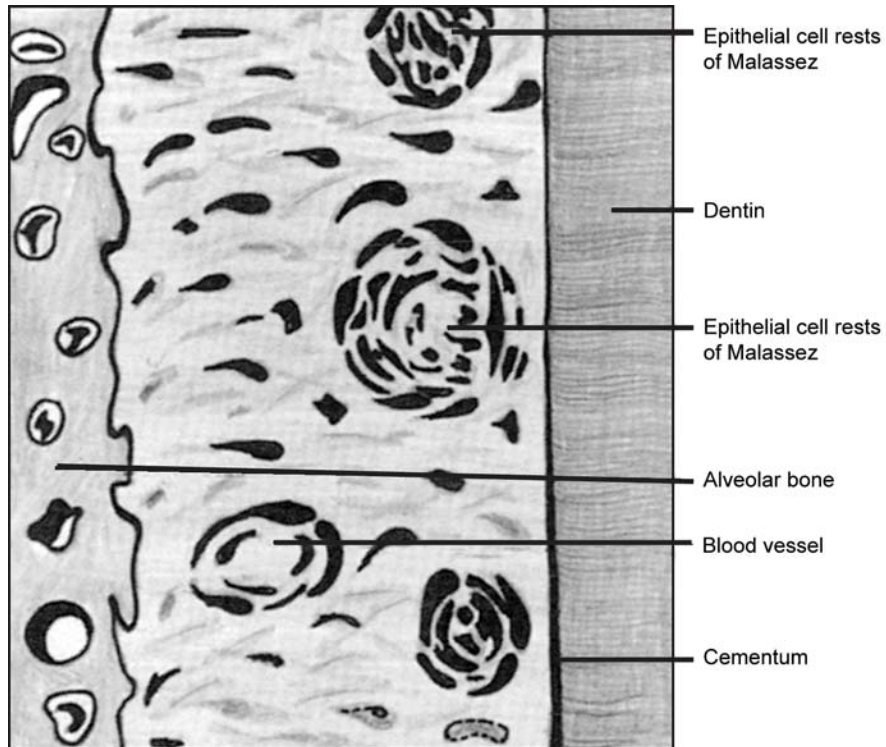


Figure 8.10: Islands of epithelial cell rests of Malassez in periodontal ligament

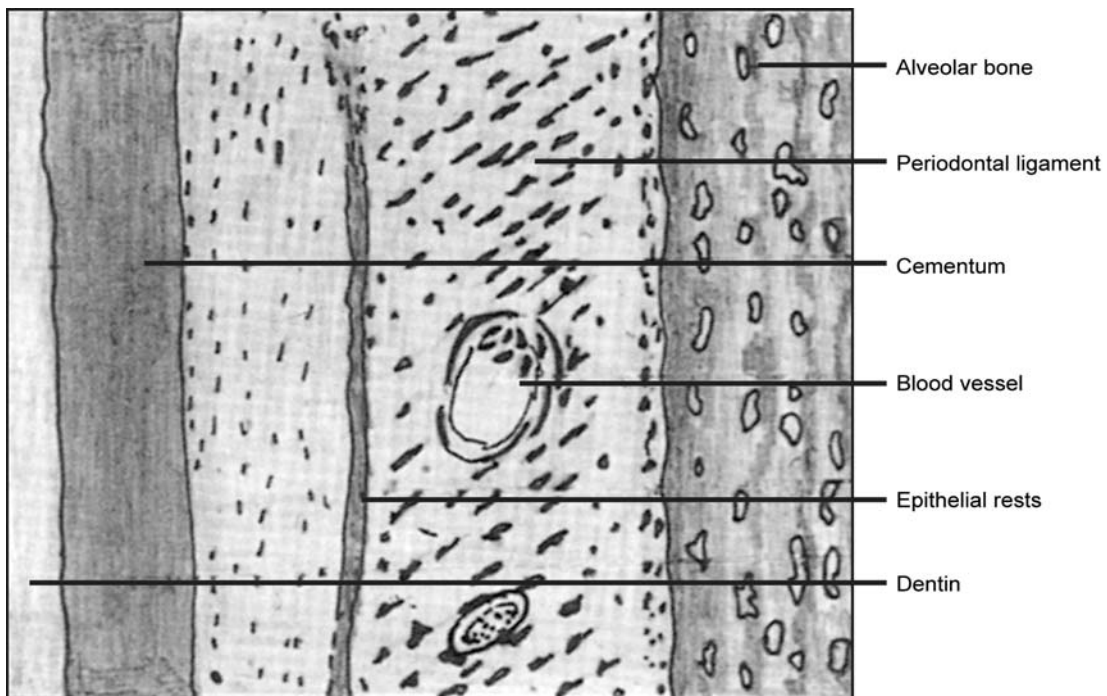


Figure 8.11: Long strand of epithelium in periodontal ligament (as seen under Electron Microscope)

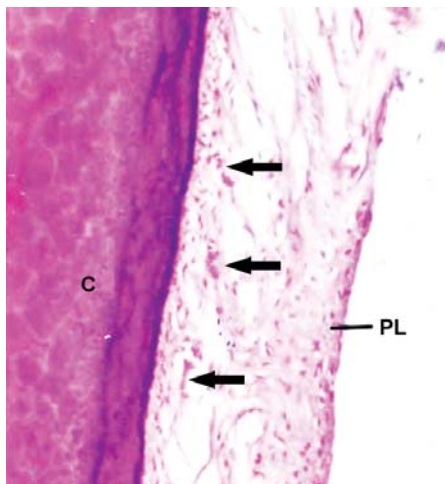


Figure 8.12: Epithelial cell rests of Malassez: Periodontal ligament (PL) showing small groups of round cells indicated by arrows. These cell rests lie close to cementum (C) and represent remnants of epithelial root sheath (H & E x 100)

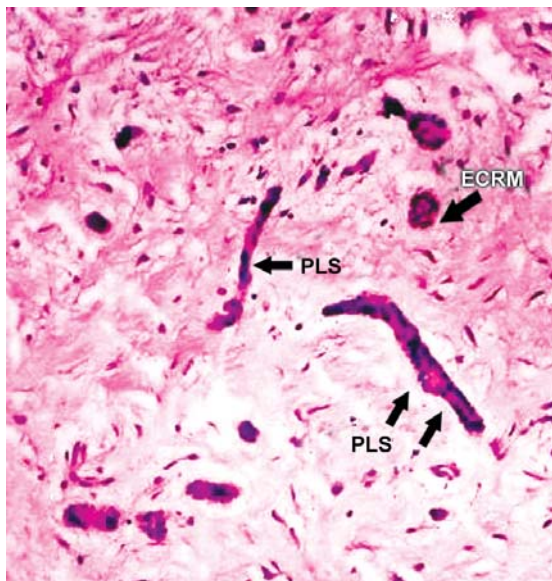


Figure 8.13: Many periodontal ligament strands (PLS) and groups of epithelial cell rests of Malassez (ECRM). (H & E x 100)

RELATIONSHIP BETWEEN THE CELLS

Cells of periodontal ligament are associated with the cementum, bone and fibrous connective tissue and form a three-dimensional network. Cells are in contact with each other through their processes. The nature of junction is gap junction, which establish a direct communication between the cells.

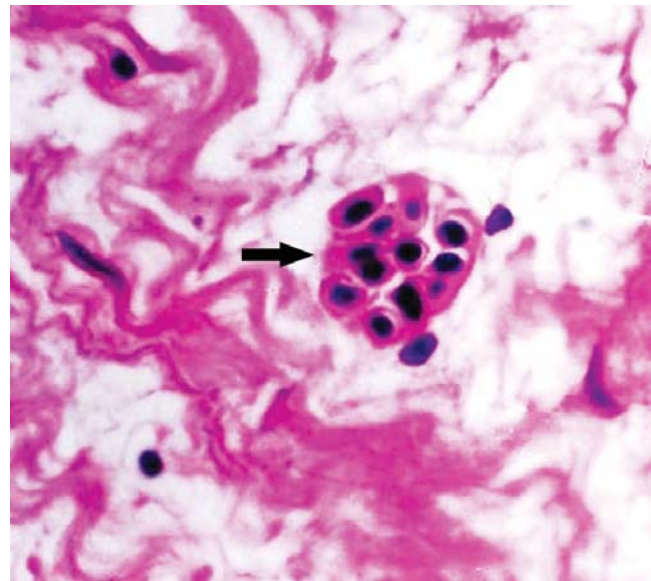


Figure 8.14: High magnification (Fig. 8.13) of showing round or cuboidal nature of epithelial cell rests of Malassez in cross-section indicated by arrow. Cell boundaries are distinct (H & E x 400)

Mast Cells

Mast cells are the small, round or oval cells. These cells are characterized by the numerous cytoplasmic granules, which mask its small, indistinct round nucleus. The diameter of the mast cells is about 12 to 15 microns. The granules have been shown to contain heparin and histamine, but the role of heparin in the mast cells is not clear. The release of the histamine into the extracellular compartment causes proliferation of the endothelial and mesenchymal cells.

These granules stain with basic dyes. These are also positively stained by periodic acid Schiff reagent. Under electron microscope, the mast cell cytoplasm shows to contain free ribosomes, granular endoplasmic reticulum, few mitochondria and prominent Golgi apparatus. The diameter of granules averages about 0.5 to 1 microns and the granules are membrane bound. Mast cells play an important role in regulating the endothelial and fibroblast cell populations. These cells degranulate in response to antigen - antibody formation on their surface.

Macrophages

Macrophages are derived from blood monocytes. These are present near the blood vessels. These can be differentiated from fibroblasts by the phagocytosed material which is

present in the cytoplasm of macrophages. These cells have a horseshoe-shaped or kidney-shaped nucleus with peripheral chromatin. Under electron microscope, microvilli are seen on the surface and cytoplasm contains numerous free ribosomes. Rough endoplasmic reticulum is less prominent and numerous lysosomes are present. Macrophages help in phagocytosing dead cells and secreting growth factor, which helps to regulate the proliferation of adjacent fibroblasts. Important molecules synthesized by the macrophages are interferon, prostaglandin and growth factors of fibroblast and endothelial cells.

Eosinophils

Eosinophils are rarely seen in periodontal ligament. These cells possess various granules. Granules contain one or more crystalloid structures. The important function of eosinophil is phagocytosis.

EXTRACELLULAR SUBSTANCE

Extracellular substance comprises the following.

1. Fibers
 - a. Collagen
 - b. Oxytalan
2. Ground substance
 - a. Proteoglycans
 - b. Glycoproteins

Fibers

The fibers of the periodontal ligament are made up of collagen and oxytalan.

Collagen

Collagen fibers are the principal fibers of the periodontal ligament. The periodontal ligament is basically made up of a type I and type III collagen. Collagen is a high molecular weight protein. To the collagen are attached small number of sugars and the small glycoproteins. There are approximately twelve types of collagen, which are similar in chemical structure. Each collagen is produced by a different gene. Collagen macromolecules are rod-like and are arranged in the form of fibrils. The fibrils show an ordered periodic banding pattern. These fibrils are of very

small diameter so these cannot be seen by light microscope. Fibrils are packed side by side to form bundles or fibers. These fibers are the smallest order of collagen that can be seen by light microscope. Many collagen fibers are arranged in larger bundles and are termed principal fibers. The collagen fibers show deep green color by Masson's trichrome stain (Figs 8.15 to 8.17). Vitamin C helps in the formation and repair of collagen.

Type I collagen contains two identical $\alpha 1$ (I) chains. This collagen is uniformly distributed and contains less hydroxylysine and glycosylated hydroxylysine. Type I is covalently linked to type III.

Type III collagen contains three identical $\alpha 1$ (III) chains. This collagen accounts for 20 percent of collagen fibers and is high in hydroxyproline, low in hydroxylysine and also contains cysteine. This collagen is important for collagen turnover, and tooth mobility and is found in the periphery of Sharpey's fiber attachment into alveolar bone.

Type IV collagen is associated with epithelial cell rests and blood vessels. This collagen is important in maintaining the integrity of the periodontal ligament by anchoring the vasculature to the elastic system.

Type VI collagen is a short chain molecule and is important in maintaining the elasticity and integrity of the extracellular matrix.

Traces of Type VII are also found in ligament and are associated with epithelium cell rests and blood vessels.

Type VIII is found when the ligament is fully functional.

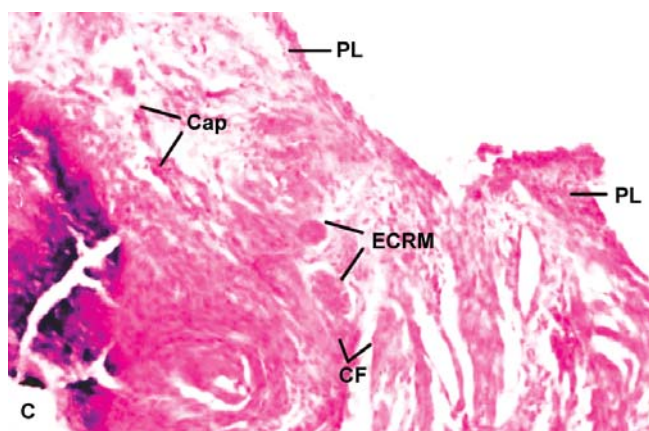


Figure 8.15: Periodontal ligament (PL) showing bundles of collagen fibers (CF), epithelial cell rests of Malassez (ECRM), capillaries (Cap) and other cellular components (H and E x 150) C= Cementum

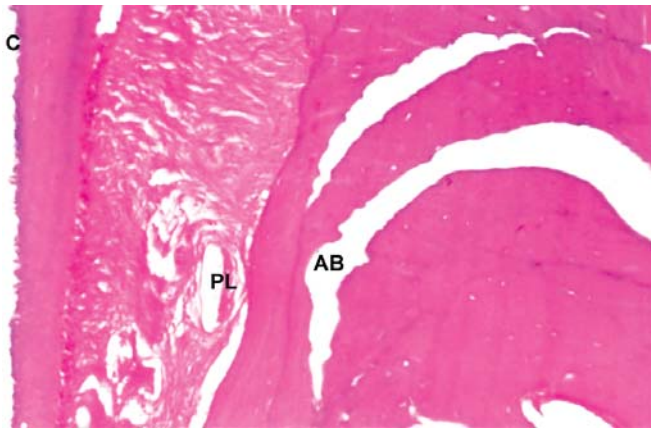


Figure 8.16: Periodontal ligament (PL) consisting of dense collagen bundles arranged horizontally attached to the cementum (C) on left side and to the alveolar bone (AB) on right side anchoring the tooth to the bone. As seen in decalcified section. (H & E x 200)

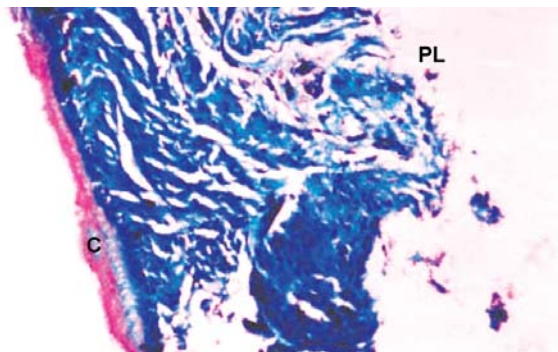


Figure 8.17: Masson's Trichrome stain showing deep green color of the collagen fibers of periodontal ligament (PL). The layer of cementum (C) stain red x 100

PRINCIPAL FIBERS

The principal fibers are arranged in five particular groups. These are as follows .

- i. *Alveolar crest group:* The fibers originate from the crest of alveolar process to the cervical part of the cementum. The fibers of this group resist the various forces like tilting, extrusive, intrusive and rotational forces.
- ii. *Horizontal group:* These fibers run at right angles to the long axis of the tooth from the cementum to the alveolar bone. These fibers are mostly parallel to the occlusal plane of the arch and located in the coronal one-fourth of the periodontal space. The fibers of this group resist the horizontal and tipping forces.

- iii. *Oblique group:* The bundles of oblique group constitute the main attachment of the tooth. These fiber bundles are most numerous. They run obliquely from cementum to the bone. The half life of collagen fibers is between three to twenty-three days. The fibers of this group resist the vertical and intrusive forces.
- iv. *Apical group:* The fibers radiate from the apical region of the root to the surrounding bone. These bundles are irregularly arranged. These fibers are observed when the root is completely formed. The fibers of this group resist the luxation force, prevent tipping movement and protect the blood vessels, lymph and nerves.
- v. *Inter-radicular group:* The bundles originate from the furcation area of multi rooted teeth to the crest of inter radicular septum. On one side of the periodontal space, the collagen fibers are embedded into cementum, while on other side they are attached to the alveolar bone. These embedded fibers are the Sharpey's fibers. These fibers get entrapped in alveolar bone either during development of the interdental septum or by bone deposition at the alveolar crest. The fibers of this group resist the luxation, torquing and tipping forces. These fibers are lost when furcation area is exposed (Figs 8.18 to 8.25).

Intermediate Plexus

The principal fibers course from cementum to bone. Under light microscope, it is seen that some fibers arising from cementum and bone are joined in the mid-region of the periodontal space. This gives rise to a distinct zone called as intermediate plexus. The remodeling of fibers takes place in intermediate plexus. This allows adjustments in the ligament, which accommodate small movements of the tooth. But there is no support for this belief and it is thought that intermediate plexus is an artifact that arises out of the plane of section. Fibers form a three dimensional complex network by joining the neighboring fibers. Crimping is a specific type of waviness in collagenous tissue of the periodontal ligament. This feature is best observed under polarizing microscope. The group of fibers which maintain the functional integrity of the periodontum are found in lamina propria of the gingiva and form gingival ligament. They have been described in the chapter of oral mucosa membrane.

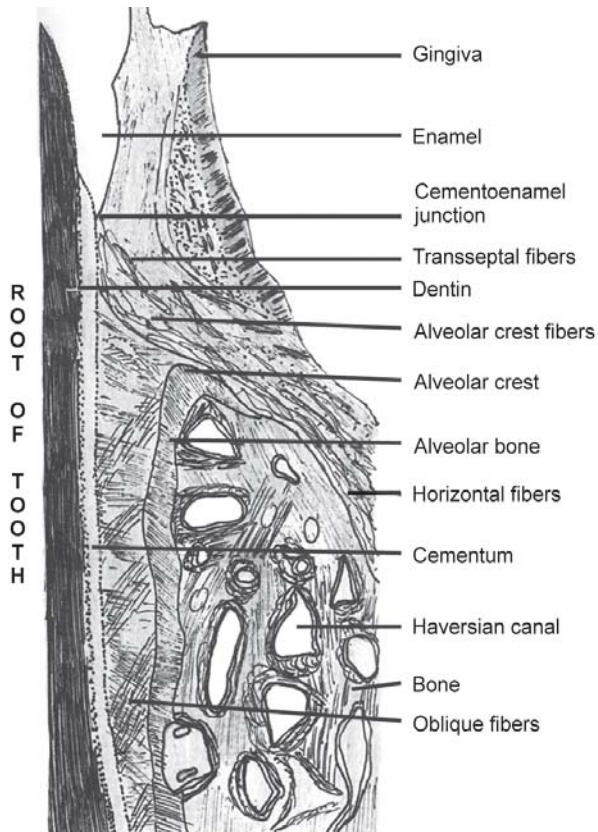


Figure 8.18: Periodontal structures (as seen under high magnification)

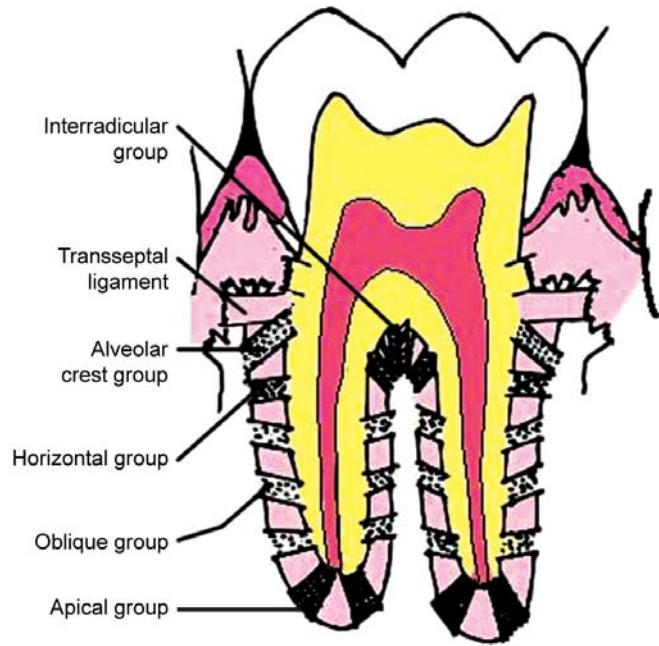


Figure 8.20: Classical arrangement of principal fibers of periodontal ligament

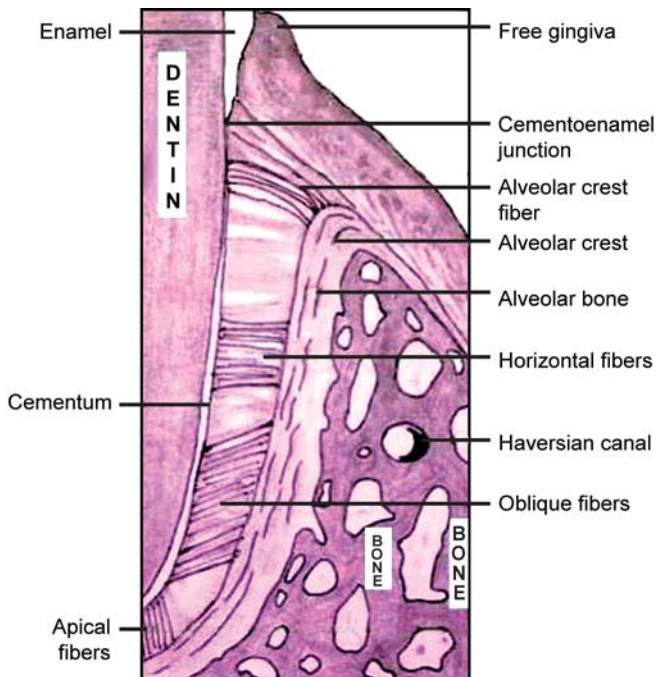


Figure 8.19: Various types of fibers of periodontal ligament

Elastic Fibers

Elastic fibers are of the following types.

1. Mature elastic fibers
2. Immature elastic fibers

Mature Elastic Fibers

Mature elastic fibers are also known as elastin fibers. These fibers consist of microfibrillar component, which is present around the periphery and scattered throughout the amorphous core of elastin protein. These fibers are mostly observed in the walls of afferent blood vessels.

Immature Elastic Fibers

Immature elastic fibers are of following types.

Elaunin Fibers: These fibers consist of microfibrillar component, embedded in a relatively small amount of amorphous elastin. These fibers are found in gingival ligament and peripheral ligament.

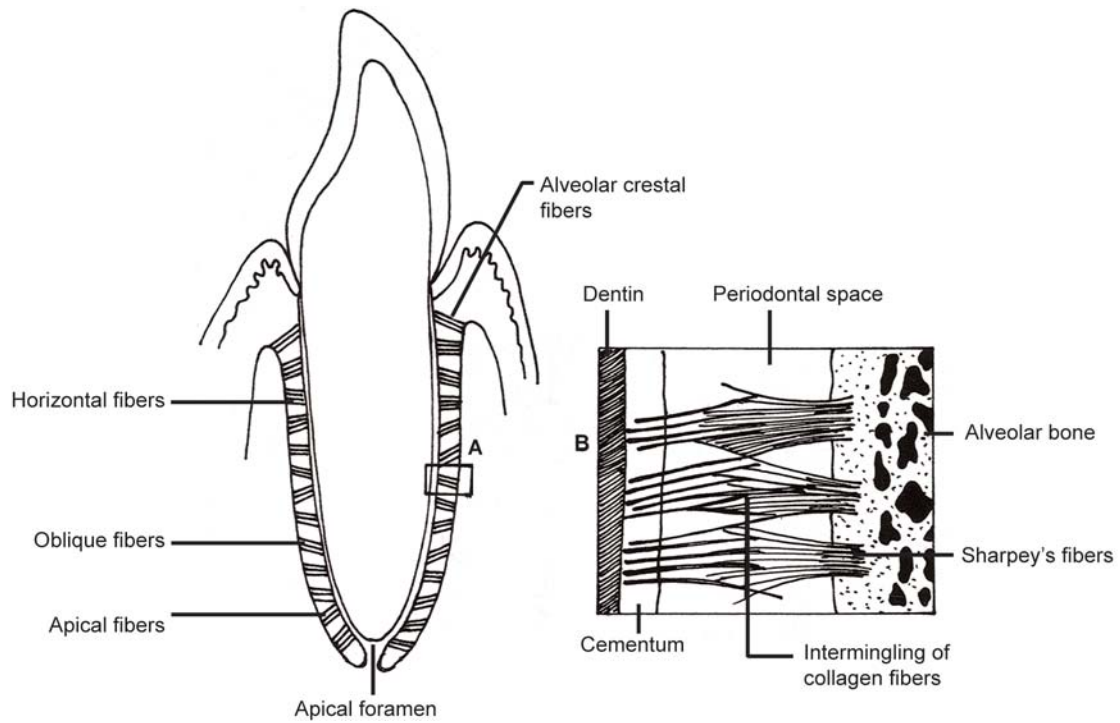


Figure 8.21: Principal fibers of periodontal ligament and their terminal parts. Sharpey's fibers and alveolar bone. 'B' is the enlarged view of section 'A'

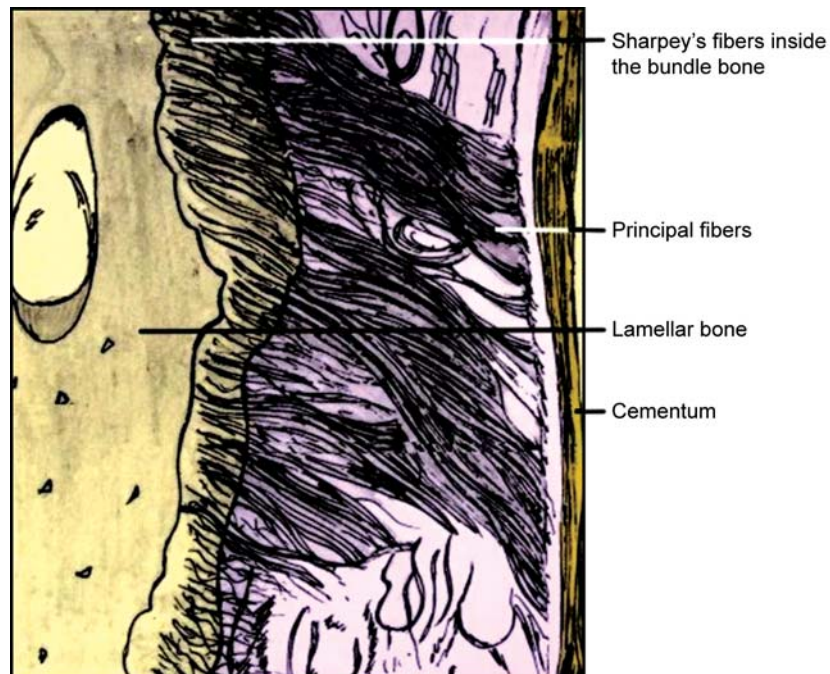


Figure 8.22: Principal fibers (continuous collagen fibers) embedded in the cementum (right) and bundle bone (left). Sharpey's fibers are within the bundle bone and overlying lamellar bone

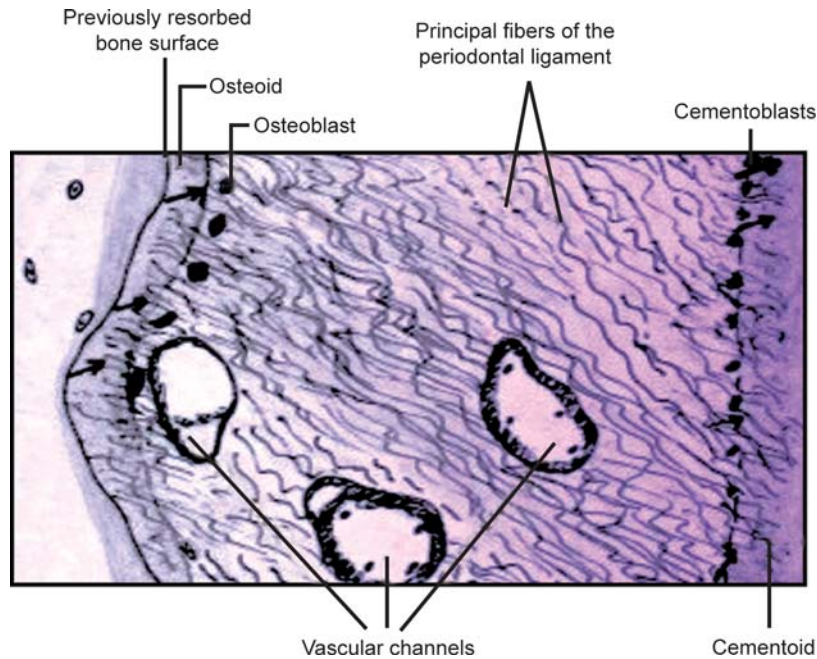


Figure 8.23: In longitudinal section principal fibers of the periodontal ligament follow a wavy course. Their ends are embedded (shown by small arrows) in forming calcified tissues osteoid and cementoid to hold the tooth in the alveolar bone

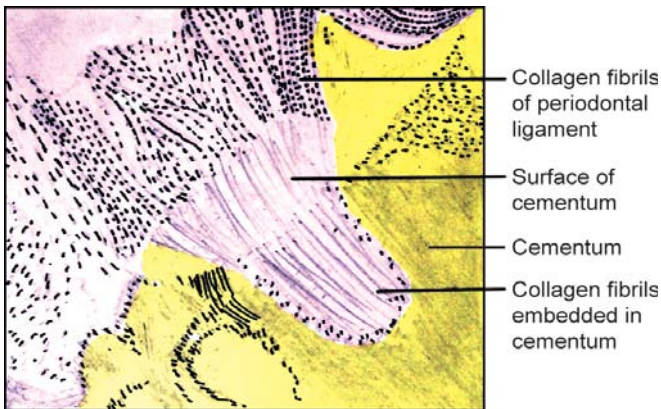


Figure 8.24: Collagen fibrils passing from the periodontal ligament into cementum. Their embedded portions in cementum are collectively known as Sharpey's fibers

Oxytalan Fibers: Some elastic fibers found in the periodontal ligament known as oxytalan are largely restricted to the walls of blood vessels (Fig. 8.26). The fibers originate from cementum or possibly bone and are embedded into the walls of blood vessels. The function of oxytalan is to support the blood vessels in the periodontal ligament. Oxytalan fibers are immature elastic fibers. If the tissues are oxidised and then stained to color elastic

fibers, oxytalan fibers can be demonstrated under light microscope. The orientation of oxytalan fibers is in an axial direction. Near the apex, the oxytalan fibers form a complex network. These fibers consist of microfibrillar component and are 0.5 to 2.5 micron in diameter.

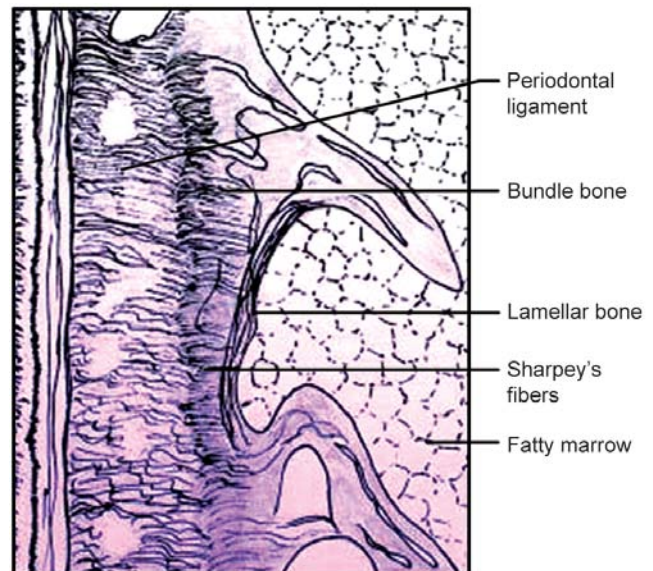


Figure 8.25: Penetration of Sharpey's fibers into bundle bone

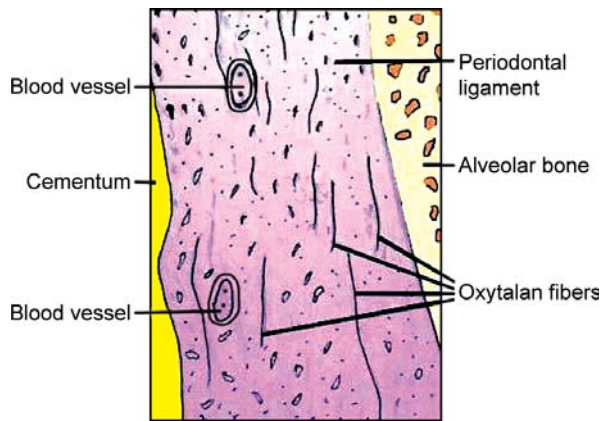


Figure 8.26: Orientation of oxytalan fibers in periodontal ligament

Reticular Fibers: Reticular fibers are fine immature collagen fibers. These fibers are related to the basement membrane of blood vessels and epithelial cells of periodontal ligament.

Secondary Fibers

Secondary fibers are non-directional and randomly oriented fibers. They represent the newly formed collagenous elements which are not incorporated into the principal fibers. These fibers are mostly associated with the path of vasculature and nerves elements.

Indifferent Fiber Plexus

The small collagen fibers run in all directions, associated with large principal collagen fibers and form a plexus known as indifferent fiber plexus. This plexus is thought to be an artifact because it is not examined under light microscope or transmission electron microscope.

Ground Substance

The ground substance is present in between the cells and fibers of the periodontal ligament (Fig. 8.27). All the anabolic and catabolic substances pass through ground substance. Hence, for proper functioning of the periodontal ligament its integrity should be maintained. Ground substance is a gel-like matrix and consists of hyaluronate, glycoprotein, proteoglycans glycosaminoglycans and substrate adhesion molecules. Two major groups of substances are, proteoglycans and glycoproteins. These groups are composed of proteins and polysaccharides. A glycoprotein, fibronectin occurs in filamentous form in the periodontal ligament. This glycoprotein contains a chemical group that gets attached to the surface of the fibroblast, to the collagen, proteoglycans and fibrin. The orientation of these proteins is related to microfilaments in cytoplasm of fibroblast. The ground substance has 70% water in periodontal ligament. Proteoglycans and glycoproteins are

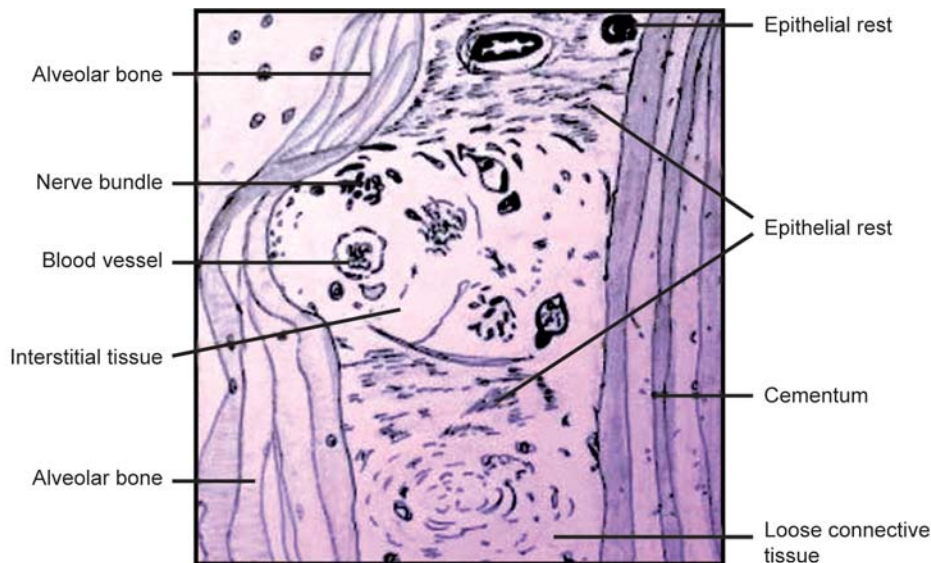


Figure 8.27: In periodontal ligament, the spaces between the fibers contain loose connective tissue, blood vessels and nerves

demonstrated histochemically by a dye or as an electron dense material under electron microscope. Glycosaminoglycans contain hexosamine, heparin sulfate and hexuronic acid.

Substrate adhesion molecules in the periodontal ligament are osteonectin, laminin, fibronectin, tenascin and undulin. Proteoglycans like fibromodulin, perlecan, Cd 44 and Syndecan 1 and 2 are also present in periodontal ligament. The function of ground substance is transporting the food to cells and the waste products from cells to blood vessels.

Interstitial tissue: Blood vessels, lymphatics and nerves are surrounded by a loose connective tissue and these areas are known as Interstitial tissue. They can be recognized under light microscope. Their biologic significance is not known.

STRUCTURES PRESENT IN THE CONNECTIVE TISSUE

Following structures are present in the connective tissue.

Blood Vessels

Periodontal ligament has an exceptionally rich vascular supply (Figs 8.5, 8.6, 8.10, 8.11, 8.26 to 8.31). The main blood supply to periodontal ligament is from superior and inferior alveolar arteries. Major arteriovenous anastomoses occurs in the ligament. The blood vessels are derived from the following:

- *Branches from apical vessels* - vessels supplying the pulp
- *Branches from intra-alveolar vessels*- run horizontally and penetrate the alveolar bone to enter into periodontal ligament.
- *Branches from gingival vessels.*

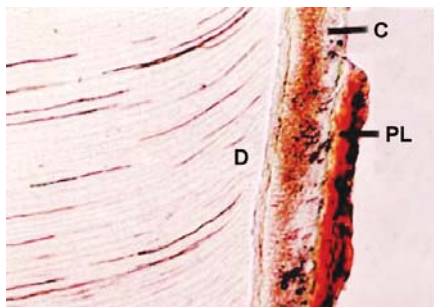


Figure 8.28: Periodontal ligament in ground section is seen as reddish amorphous structure due to high vascularity of the ligament (x 100). D= Dentin, C= Cementum, PL= Periodontal ligament

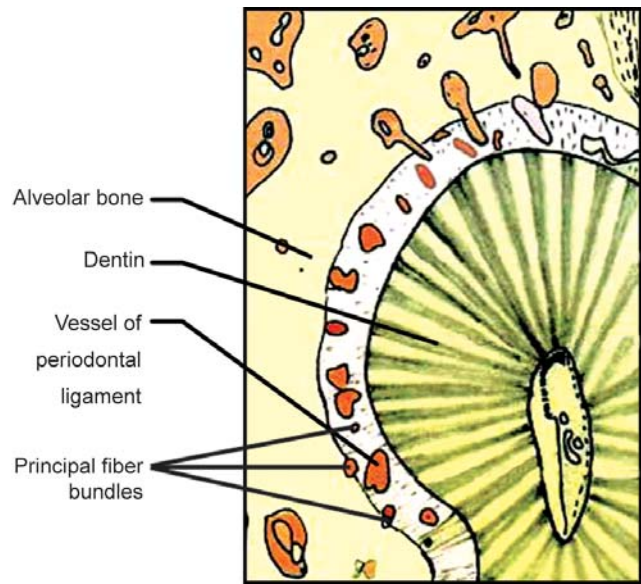


Figure 8.29: Location of major vessels of periodontal ligament is in between principal fiber bundles, close to alveolar wall. The branching and anastomoses of vessels form a capillary plexus around the root as seen in transference section

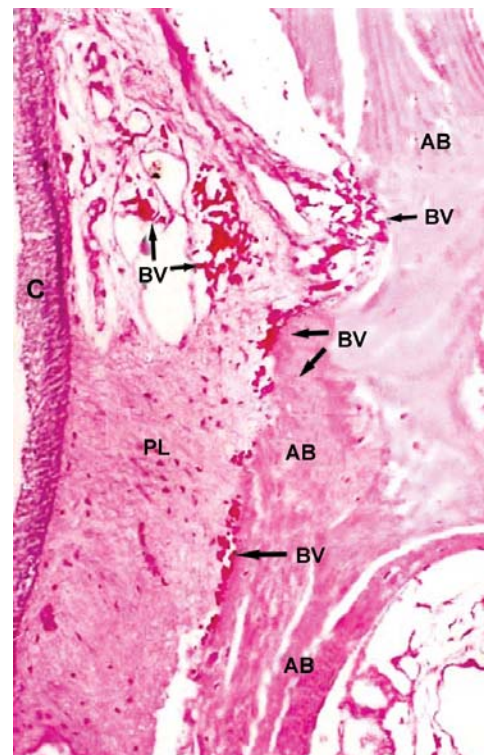


Figure 8.30: Blood supply of periodontal ligament (PL) is derived from the alveolar bone (AB). C = Cementum, BV = Blood vessel. (H & E x 100)

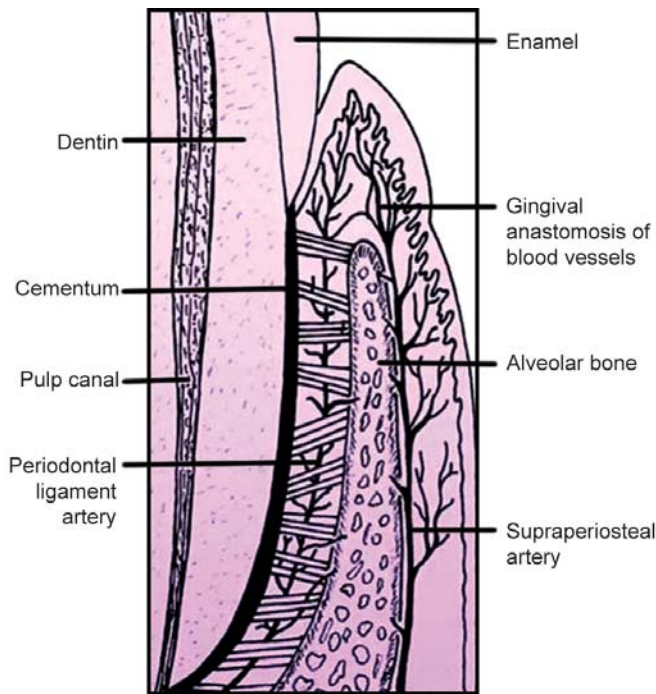


Figure 8.31: Blood supply to periodontal ligament showing branches from gingival vessels

The arterioles and capillaries ramify and form a rich network, which is mostly found in the periodontal space, nearer to bone than cementum. Rich vascular plexus is found at the apex and in the cervical part of the ligament. The average diameter of the arterioles in periodontal ligament is 20 micron. Fenestrations in the capillaries and cervical plexus of capillary loops are the specific features of periodontal ligament vasculature. These features increase the capacity for filtration and diffusion which might be related to high rate of turnover.

Venous vessels run axially and drain to apex. Between two sides of microcirculation there is the presence of many arteriovenous anastomoses. Mean average diameter of venous channels is 28 micron. Dense venous network is seen around the apex.

Lymphatics

Lymphatic vessels are also found in the periodontal ligaments. The lymphatic drainage is from the ligament to the alveolar bone. Lymphatic vessel network follows the path of blood vessels.

Nerves

On the basis of function, nerve fibers of periodontal ligament are of the following types.

- A. *Sensory fibers:* Sensory are related with touch, pain, pressure and proprioceptive sensation.
- B. *Autonomic fibers:* Autonomic fibers are related with vessels of periodontal ligament.

Nerves found in ligament passes through foramina in the alveolar bone. These nerves are the branches of second and third division of fifth cranial nerve (trigeminal nerve). These nerves follow the same path as blood vessels. In apical region, the nerves run toward the cervix. The nerves which are present along the root, branch and these run coronally as well as apically. The nerve fibers may be myelinated and of large diameter. The diameter of myelinated fibers is 5 to 15 micron and unmyelinated fibers is 0.5 micron. Alternatively, they may be of small diameter and may be myelinated or not. The small diameter fibers are concerned with pain. These end in the fine branches throughout the ligament.

Large diameter fibers are concerned with pressure and they terminate in variety of endings as knob-like, spindle like, and Meissner's corpuscle-like. Mechanoreceptors of periodontal ligament respond to force applied to the tooth in specific direction. Cell bodies of 75 percent of mechanoreceptors are located in the trigeminal ganglion and remaining 25 percent are located in mesencephalic nucleus.

Cementicles

Cementicles are the small calcified bodies present in periodontal ligament. These are not the cells of cementum. They may form into the large calcified bodies and fuse with cementum, or they remain free (Fig. 8.32). When they join with cementum, cementicles form the excementoses. The origin of cementicles is not known. Cementicles are generally found in old age. It is a possibility that degenerated epithelial cells form a nidus for their calcifications.

FUNCTIONS

The periodontal ligament has many functions, important among them are as follows.

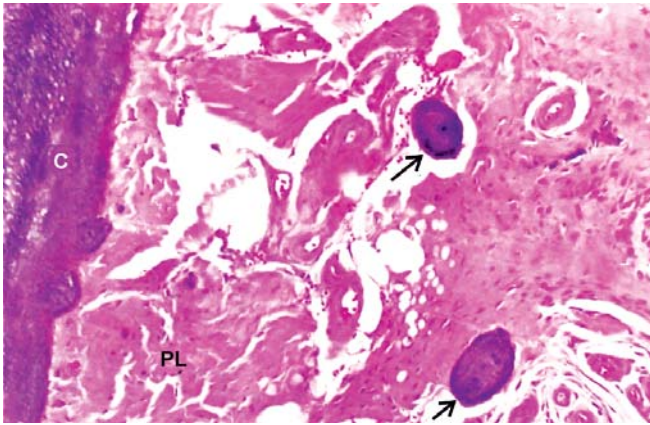


Figure 8.32: Cementicle: Deposits of cementum (C) can be seen in the periodontal ligament (PL). Arrows indicate cementicles, (x 100)

Supportive Function

When a force acts on the tooth and the tooth moves in its socket, the periodontal ligament gets compressed while the other part of the periodontal space get widened, and provides support for the tooth. The collagen fibers present in the periodontal ligament act as a cushion to withstand the masticatory forces. The pressure of the blood in the ligament also provides support for the tooth as it also acts as a cushion (Figs 8.33 A to D).

In the widened part of the periodontal space, the collagen fibers extend to a certain limit after the application of a force to the tooth.



Figures 8.33 A and B: Distribution of faciolingual forces (arrow) around the axis of rotation (black circle on root) in a maxillary central incisor; A. The periodontal ligament fibers are compressed in areas of pressure (a) and stretched in area of tension (b); B. Right, the same tooth in a resting state

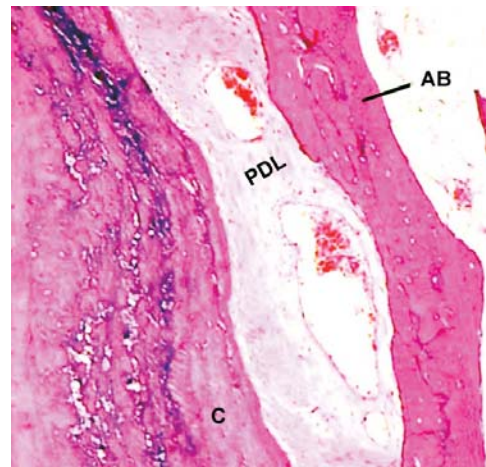


Figure 8.33 C: Periodontal ligament of a functioning tooth is broad and contains alveolar spaces through which blood vessels and nerves traverse. Periodontal ligament (PDL), Cementum (C) Alveolar bone (AB). x 100

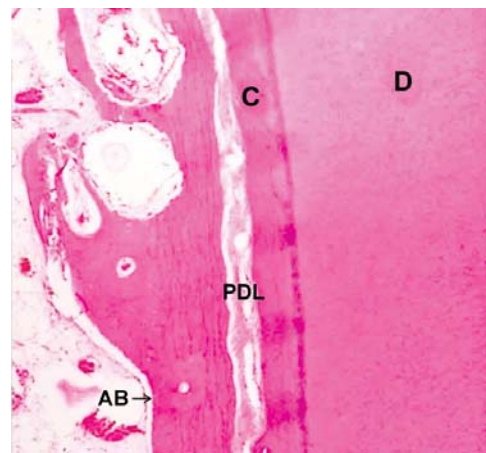


Figure 8.33 D: Periodontal ligament of a nonfunctioning tooth is markedly narrow or shrunk, in comparison with Fig. 8.33 C Periodontal ligament (PDL), Cementum (C) Dentin (D), Alveolar bone (AB)

Because these fibers are non-elastic in nature these help to prevent the tooth from being moved too far. Thus collagen fibers acts as an attaching apparatus between cementum and alveolar bone and it also acts as a cushion.

Sensory Function

The nerves of periodontal ligament provide the proprioceptive mechanism to detect an application of small force to the teeth and also very small displacement of teeth. Thus, it helps in protecting the supporting structures of tooth and substance of crown from effects of the masticatory force.

Nutritive Function

Blood vessels of the periodontal ligament provide nutrition to the cells of periodontium because they contain various anabolites and other substances, which are required by the cells of the ligament. Compression of the blood vessels (due to heavy forces applied on the tooth) leads to the necrosis of cells. Blood vessels also remove catabolites.

Formative Function

The cells of periodontal ligament produce the cementoblast and the osteoblasts. The cementoblast and osteoblast can form new cementum and bone, respectively (Fig. 8.34).

Homeostatic Functions

The cells of periodontal ligament have an ability to resorb and synthesize the extra cellular substances of the ligament, alveolar bone and cementum. The mechanism by which the resorption and synthesis occurs is unknown. It is a continuous process which occurs throughout the life of the tooth. Among all the connective tissues in body, the turn over rate of the collagen of the periodontal ligament is fastest. The cells on bone side half of the ligament may be more active than on cemental side. Deposition of cementum is a slow, continuous process while resorption does not occur regularly.

The resorption and synthesis are the controlled procedures. Under normal conditions, all tissues of the

periodontium maintain their integrity. If the homeostatic mechanism is upset, there is derangement of the periodontium. If there is a long standing damage to periodontal ligament, which is not repaired, the bone is deposited in the periodontal space. This results in the obliteration of the space and ankylosis between bone and tooth.

The quality of the tissue changes, if the balance between synthesis and resorption is disturbed. If there is deprivation of vitamin C or protein, which are essential for collagen synthesis, resorption of collagen will continue. The synthesis and the replacement of the collagen is reduced. So, there is progressive destruction and loss of extracellular substance of the ligament. This occurs more on the bone side of the ligament. Hence, there is a loss of attachment between the bone and tooth and at last there is loss of tooth. This occurs in scurvy where there is a complete lack or severe long standing deficiency of vitamin C from the diet. The connective tissue cells of the periodontal ligament are turned over. The old cells are replaced by the new cells that are provided by cell division of the progenitor cells.

Important molecules secreted by the cells of periodontal ligament which prevent the ankylosis and maintain the unmineralized periodontal ligament are as follows.

1. Msx 2- Repressing Run x 2 (runt related transcription factors 2) and prevent the osteogenic differentiation of periodontal ligament fibroblast.
2. Glycosaminoglycans – Maintain the unmineralized state of the periodontal ligament.

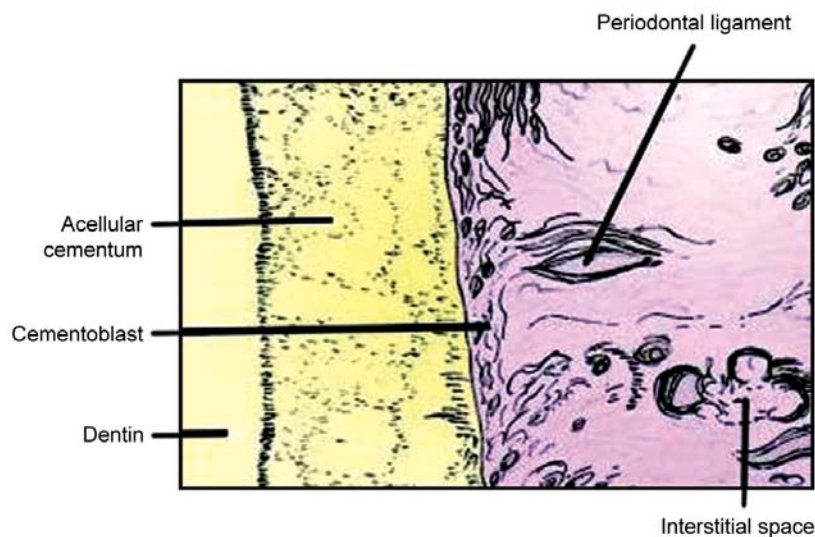


Figure 8.34: Transverse section of periodontal ligament

3. Matrix 'Gla' protein - Inhibit mineralization and preserve the width of the ligament.
4. Balance between the function of osteopontin and bone sialoprotein also maintain the region of unmineralized periodontal ligament.
5. Bone stromal cells inhibit mineralized bone nodule formation. The inhibition is dependant on production of prostaglandin.

A functioning periodontal ligament exhibits all the structures but with loss of function, the extracellular substance of the ligament is lost. This occurs because of the less synthesis of substances required to replace structural molecules. So, there is an increased deposition of cementum and decrease in alveolar bone tissue which is reversible, and is largely controlled by cells. It is called as the homeostatic mechanism.

So, the main function of the periodontal ligament is to attach and support the tooth in the bony socket.

AGING OF LIGAMENT

Aging occurs in all tissues of the body including all ligaments. The number of cells and their activity decrease, scalloping occurs in cementum and alveolar bone. Some fibers are attached at the peaks of those scallops only and not in the depressions. This adversely affect the support to the teeth. If general and dental health is good, periodontium may remain healthy even in advanced age. However the activity of cells and their number decreases with aging. With aging due to restricted and soft diet physiological stimulation to the PDL is reduced. Gingival recession and chronic periodontal diseases cause destruction of PDL. Due to reduced or non functioning of some teeth PDL becomes weaker.

CLINICAL CONSIDERATIONS

The main function of periodontal ligament is to support the tooth in its socket. Its thickness varies in different teeth in the same person (Tables 8.2 and 8.3) and in different locations on the same tooth.

The periodontal ligament is thinnest in the middle region of the root. This indicates that the fulcrum of movement is in this region. The functional movement of the tooth helps in maintaining the thickness of the periodontal ligament. So, it is thick in the teeth that are exposed to excessive occlusal stress and thin in functionless and embedded teeth.

If a tooth is long out of function, the supporting tissue of this tooth are not fully adapted to carry the load suddenly placed on the tooth by a restoration like full cast crown. Hence the patient cannot use the tooth immediately. After the restoration, supporting tissues take some time to adapt to new functional demands. Therefore, after restoration an adjustment period should be allowed and tooth should be used gradually first for soft food before the tooth is put to full use to bite hard food.

It shows that the width of the periodontal ligament decreases as the age advances.

Table 8.2: Average of the thickness of periodontal ligament of 154 teeth from 14 human jaws*

S. No.		At alveolar crest (mm)	At midroot (mm)	At apex (mm)	For Entire tooth (mm)
1.	Ages 11 to 16 (83 teeth from 4 jaws)	0.23	0.17	0.24	0.21
2.	Ages 32 to 50 (36 teeth from 5 jaws)	0.20	0.14	0.19	0.18
3.	Ages 51 to 67 (35 teeth from 5 jaws)	0.17	0.12	0.16	0.15

From Coolidge ED: J Am Dent Assoc 24: 1260, 1937

**The table shows that the width of the periodontal ligament decreases with age and that it is wider at the crest and at the apex than at the midroot.*

Table 8.3: Comparison of periodontal ligament in different locations around the same tooth (subject 11 years of age)

S. No. (mm)		Mesial (mm)	Distal (mm)	Labial (mm)	Lingual (mm)
1.	Upper right central incisor mesial and labial drift	0.12	0.24	0.12	0.22
2.	Upper left central incisor, no drift	0.21	0.19	0.24	0.24
3.	Upper right lateral incisor and labial drift	0.27	0.17	0.11	0.15

From Coolidge ED: J Am Dent Assoc 24: 1260, 1937

The table shows variation in width of the periodontal ligament on the mesial, labial and lingual sides of the same tooth

1. In the periodontal ligament, aging results in more number of elastic fibers and decrease in the vascularity, mitotic activity, fibroplasia and in the number of collagen fibers and mucopolysaccharides. A reduction in width represents decrease in strength

of the masticatory musculature. An increase in width may be due to the support of the entire functional load.

2. If the gingivitis is not cured and supporting structure becomes involved, the disease is termed as periodontitis. The periodontitis has been classified according to the role of progression and according to the age of onset.
3. There are fewer coccal cells and more motile rods and spirochetes in the diseased site than in the healthy site. The bacteria consist of gram-positive facultative rods and cocci in the healthy site while in the diseased site, gram-negative rods and anaerobes are more in number.
4. Trauma to the ligament due to mechanical separation can produce pathologic changes such as fracture or resorption of the cementum, tears of fiber bundles, hemorrhage and necrosis. These result in resorption of the bone and the periodontal ligament is widened, so the teeth become loose. If the trauma is eliminated, repair will take place.
5. Resorption and formation of both bone and periodontal ligament play an important role in the orthodontic tooth movement. If the tooth movement takes place, compression of the periodontal ligament is compensated by bone resorption, whereas on the tension side bone apposition takes place (Figs 8.33A and B).
6. Periapical area of the tooth is the main pathologic site. Inflammation of pulp reaches to the apical periodontal ligament and replaces its fiber bundles with granulation tissue, which is called as a granuloma. Granuloma further progresses into apical cyst, which is the most common pathologic lesion of the jaws.
7. Chronic periodontal disease can lead to infusion of microorganisms into the bloodstream.
8. The pressure receptors in the ligament have a protective role. Apical blood vessels are protected from excessive compression by the suspensory apparatus of the teeth.
9. The rate of mesial drift of tooth is related to health, dietary factor and age. It varies from 0.05 to 0.7 millimeters per year.
10. Teeth are slightly more mobile in the early morning than in the evening and this is explained below.

The components of the periodontal ligament act as a shock absorber. The periodontal ligament along with tissue fluid and the blood in blood vessels acts as a viscoelastic system.

During daytime, as a result of functional and parafunctional activities, forces are applied on the teeth. This results in the displacement of fluid through cribriform plates and to other regions of the ligament. The periodontal fibers gradually straighten out and act as inelastic strings. They transmit tension to the alveoli and hence teeth become less mobile.

During the night, when there is lack of activity in the oral cavity, the tissue fluid comes back into the fibers. The fibers regain their wavy course so teeth become comparatively more mobile.

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Oral Mucous Membrane

- Introduction
- General considerations
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 - Functions of oral mucosa
- Histology of oral mucous membrane
 - Oral epithelium
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 - Importance of the corrugated arrangement of interface
 - Basal lamina
 - Basement membrane
 - Lamina lucida and lamina densa
 - Layers of epithelium
- Differences between keratinized and nonkeratinized epithelium
- Connective tissue
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- Clinical considerations

INTRODUCTION

The intestinal tract (through oral cavity and anus), nasal passage and other body cavities communicate with the exterior environment. All these are lined by a moist lining, which is called as 'mucous membrane'. The moist lining of the oral cavity that is in continuation with the exterior surface of skin on one end and esophagus on the other end is called 'the oral mucosa or oral mucous membrane'.

In oral cavity food is tasted, masticated, mixed with saliva and its enzymes initiate the digestion. Inedible, thick fibrous and hard substances are sensed, felt, rejected and thrown out of oral cavity. Depending upon the function, the oral mucosa is divided into three main types.

1. *Masticatory mucosa*: It does not stretch and is attached to bone. During mastication, it bears chewing forces (covers gingiva and hard palate). It constitutes 25 percent of the total mucosa.
2. *Lining or reflecting mucosa*: It covers the musculature and is distensible. It is exposed to very mild forces. It adapts itself to the movements of cheeks, lips, and tongue and of the mandible. It lines most of the surfaces of the oral cavity except the areas of masticatory mucosa and dorsum of the tongue (covers lip, cheek, vestibular fornix, floor of mouth, soft palate and alveolar mucosa). It constitutes 60 percent of total oral mucosa.
3. *Specialized (sensory) mucosa*: It is so called as it bears the taste buds and specialized papillae (covers dorsum of tongue including taste buds). It constitutes 15 percent of the total oral mucosa.

GENERAL CONSIDERATIONS

Considering Oral Mucosa as an Organ

Oral mucosa is situated anatomically between the skin and intestinal mucosa. Hence, it shows some properties of both. The skin, oral mucosa and intestinal lining, all three consist of two separate tissue components.

- i. A covering epithelium
- ii. Underlying connective tissue.

These two tissues together perform common functions. Therefore, the oral mucosa like the skin and the intestinal lining should be considered as an organ.

Regional Modification of Oral Mucosa

Oral mucosa shows regional structural modifications according to the stress, strain and workload borne by it. The areas which are well-protected from masticatory

stress and wear and tear, are thin and delicate, like the mucosa of the floor of the mouth and the mucosa of cheek; in comparison the areas involved in mastication of food and stress-bearing are tough, thick and keratinized like the mucosa of the gingiva and the hard palate.

Organization of Oral Mucosa

Oral cavity is divided into outer vestibule and oral cavity proper

- a. *Outer vestibule*: It is the area bounded by lips, cheeks, alveolar bone and teeth, that is the areas outside teeth and jaws.
- b. *Oral cavity proper*: Separated from the outer vestibule by the alveolus bearing the teeth that is areas inside the teeth and jaws.

Boundaries of Oral Cavity

- a. *Superior border*: Formed by hard and soft palate.
- b. *Inferior border*: Floor of mouth and base of tongue.
- c. *Posterior border*: Pillars of fauces and tonsils.
- d. *Anterior and anterolateral borders*: By the lips and cheeks.

Functions of Oral Mucosa

Oral mucosa performs several functions. Main functions of oral mucous membrane are as follows.

Protection

As a surface lining, oral mucosa separates and protects deeper tissues and organs in oral region from the following.

1. Environment of oral cavity
2. Mechanical forces of biting and mastication, to surface abrasion from hard particles in the diet.
3. Microorganisms that may cause infection if they get access into the underlying tissue
4. Toxins produced by microorganisms.

Thus epithelium of the oral mucosa acts as a major barrier for all these insults.

Sensation

Rich innervation of oral mucosa makes it a very good receptor of temperature, touch, pain and taste. Certain reflexes such as swallowing, salivation and gagging are also initiated by the receptors in the oral mucosa. Certain

other reflexes in the oral mucosa respond to taste of water and signal the satisfaction of thirst.

Thermal Regulation

Thermal regulation function is quite obvious in some animals such as dogs. In these animals, considerable amount of body heat is dissipated through the oral mucosa by panting. There is no specialized blood vessels for controlling heat transfer in human oral mucosa, so it plays little part in regulation of body temperature.

Secretion

Various major and minor mucous and serous salivary glands open into the oral cavity and make it moist which helps in the mastication of food, its swallowing and digestion. The major secretion associated with oral mucosa is the saliva.

HISTOLOGY OF ORAL MUCOUS MEMBRANE

Oral mucous membrane consists of two layers (a) epithelium and (b) connective tissue.

Connective tissue components are the lamina propria and submucosa (Fig. 9.1).

Oral Epithelium

The epithelium forms the surface of oral mucosa. Oral epithelium forms primary barrier between oral environment and deeper tissues. The oral epithelium is ectodermal in origin (except the tongue, which is endodermal) and is a stratified squamous epithelium that is arranged in a number of distinct layers or strata. Oral epithelium maintains its structural integrity by a system of continuous cell renewal in which cells produced by mitotic division in the deepest layers migrate to the surface

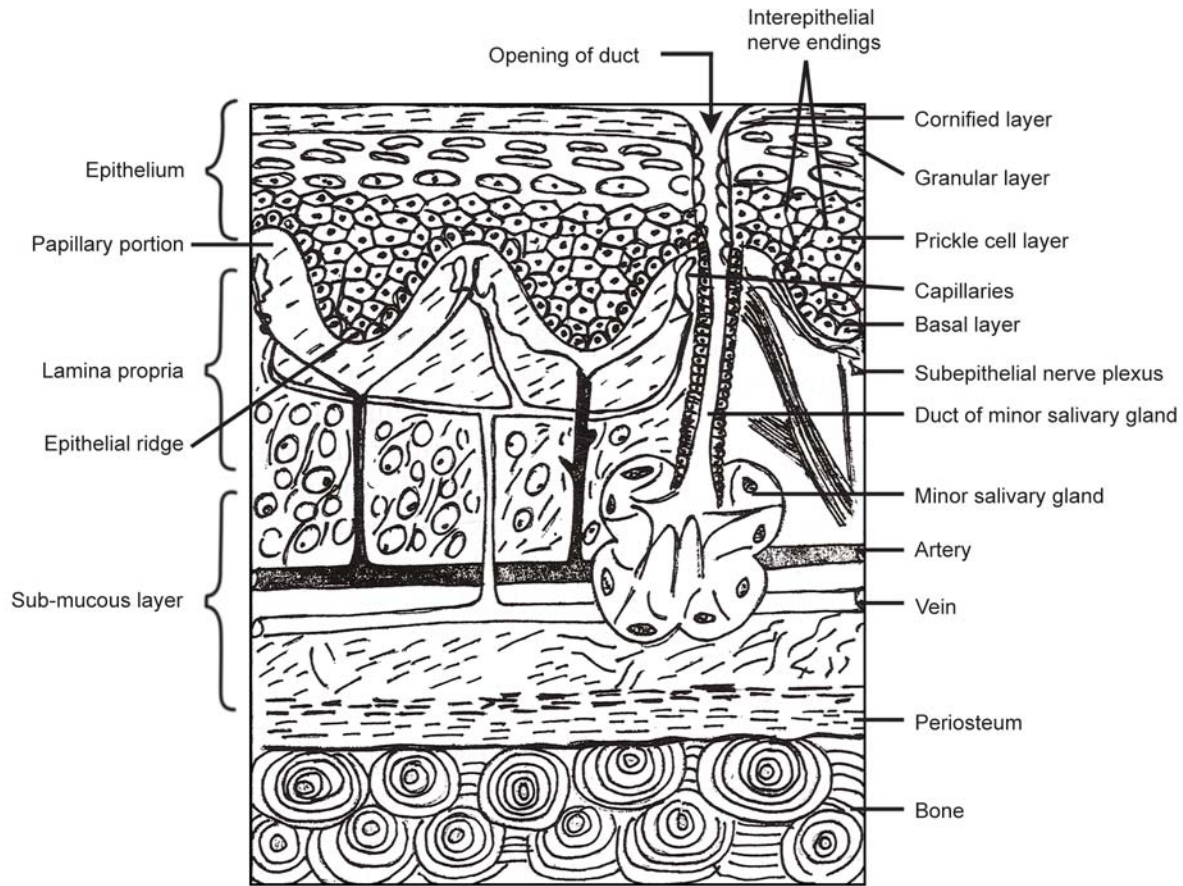


Figure 9.1: Vertical section of oral mucous membrane showing epithelium, lamina propria, submucosa, and other structures

to replace those cells that are shed. Cells of the epithelium are of two types (i) progenitor cells, (ii) maturing cells or keratinocytes.

Progenitor cells divide and produce new cells and maturing cells undergo a process of continuous maturation or differentiation and form a protective surface layer. Thus, there are two important processes that is proliferation and maturation continuously going on.

Regardless of the function, all epithelial cells contain keratin intermediate filaments.

These filaments form the component of their cytoskeleton. The filaments resemble tonofilaments and these are 7 to 11 nanometers in width.

Cell division is a cyclic activity and is commonly divided into four distinct phases.

These four phases are (i) prophase, (ii) metaphase, (iii) anaphase, (iv) telophase. These phases together are called mitosis. After mitotic phase or after a cycle of cell division, each daughter cell either recycles in progenitor population or enters the maturing process.

Diclophase: This is a decision-making period. In this period, the daughter cell decides either to recycle in progenitor population or to enter the maturing compartment. Plant alkaloid colchicine can arrest the dividing cell as they enter metaphase.

Turnover time: It is the time necessary to replace all the cells in epithelium. Turnover time of gingiva is 41 to 57 days. Turnover time of cheek is 25 days. Nonkeratinized buccal epithelium has a faster turnover than the keratinized gingival epithelium.

Factors affecting mitosis: Mitosis is affected by following factors.

1. Time of day
2. Stress
3. Inflammation
4. Intrinsic gene functions
5. Various other intrinsic mechanisms

Presence of slight subepithelial inflammatory cell infiltrate stimulates mitosis while severe inflammation causes a marked reduction in mitotic activity.

Junction of Epithelium and Lamina Propria

The junction where the lamina propria of connective tissue meets the overlying oral epithelium is a corrugated surface or interface at which papilla of connective tissue interdigitate with epithelial ridges. Papillae of connective

tissue protrude towards epithelium and have blood vessels and nerves. Epithelium in turn is formed into ridges that protrude towards lamina propria (connective tissue). These ridges interdigitate with the papillae and are called epithelial ridges.

Lamina propria contains connective tissue of variable thickness (Fig. 9.1).

Importance of the Corrugated Arrangement of Interface

1. This arrangement makes the surface area of interface larger than the simple flat junction does. It provides better attachment as well as the forces that are applied at the surface of epithelium are dispersed over greater area of connective tissue.
2. The junction is also a major route for metabolic exchange between epithelium and connective tissue. Epithelium has no blood vessels.

Basal Lamina

Basal lamina tissue is evident at the junction of epithelium and lamina propria or connective tissue. Basal lamina is evident at only electron microscopic level and is epithelial in origin.

Basement Membrane

The structure of basement membrane is also evident at the junction of the epithelium and the connective tissue, but is evident at the light microscopic level within the connective tissue.

Basement membrane is an acellular structure. It is 1 to 4 microns thick and is made up of neutral mucopolysaccharides (glycosaminoglycans). It also contains fine argyrophilic reticulin fibers as well as special anchoring fibrils. In the skin, it also contains fibronectin and laminin (glycoprotein), heparin sulfate, type IV collagen as well as some special antigens. It promotes differentiation, peripheral nerve regeneration, growth and prevents metastasis. This zone stains positively with Periodic Acid Schiff (PAS) method (Fig. 9.2).

Lamina Lucida and Lamina Densa

The basal lamina is made up of a clear zone called the lamina lucida just below the epithelial cells. A dark zone called the lamina densa is present below the lamina lucida and adjacent to the connective tissue (Fig. 9.3).

Lamina lucida is 20 to 40 nm thick. Lamina lucida contains type IV collagen, glycoproteins laminin, bulbous pemphigoid antigen which is a transmembrane component, probably associated with adhesion of the basal cell- and basement membrane glycoprotein and laminin. The lamina densa is composed of type IV collagen coated with heparin sulfate in chicken wire mesh configuration. Laminin is a large triple chain molecule ($M_r = 10^6$). Along with type IV collagen, it promotes epithelial cell growth and guides epithelial cell movement through chemotaxis.

Layers of Epithelium

The epithelium may be (i) keratinized (ii) nonkeratinized, depending upon the location. Keratinized mucosa can be

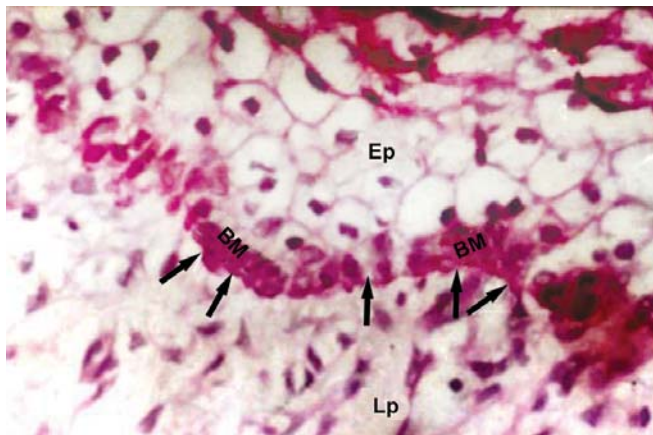


Figure 9.2: PAS stain showing thin magenta colored basement membrane (lamina densa) (BM) indicated by arrows at the base of the epithelium (Ep), separating it from the underlying lamina propria (LP) (Periodic Acid Schiff stain x 400)

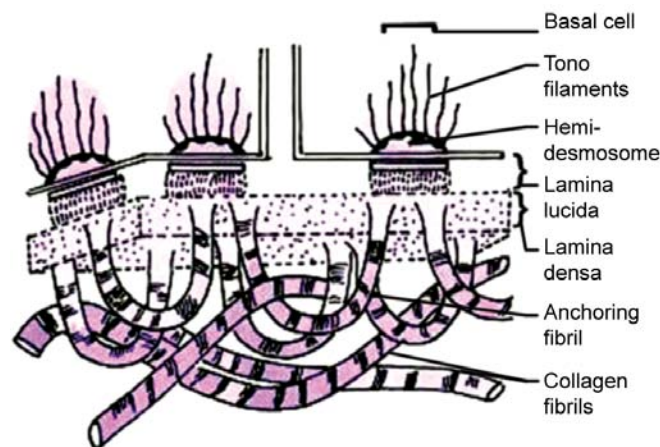


Figure 9.3: Fine structures of junction between epithelium and connective tissue

(a) orthokeratotic or (b) Parakeratotic depending upon the nature of keratinization. In orthokeratinization, the process of keratinization is absolutely complete with the formation of acellular and anuclear keratin layer on surface. In parakeratinization though cells of superficial most layer show keratin formation in the cytoplasm the cells retain the flattened to oval nuclei (Fig. 9.4).

Epithelium of the gingiva and hard palate is keratinized while some gingival tissue may be parakeratinized. Nonkeratinized tissues are (i) inner lining of cheek (ii) faucial pillars (iii) sublingual tissues (iv) floor of mouth, (Figs 9.4 to 9.10).

Cytokeratins

Cytokeratins are intermediate filament of 7 to 11 nm in diameter, they are intermediate between the larger microtubules of 25 nm in diameter and smaller microfilaments of 4 to 6 nm in diameter. The molecular weight of cytokeratins is 40 to 200 Kda. Type I (basic cytokeratins) and Type II (acidic cytokeratins) are the two types of cytokeratins. They always occur in pairs of type I with type II. Functions of cytokeratins are as follows.

1. Form the cytoskeleton of epithelial cells and maintain the shape of the cells.
2. Act as a stress bearing structure within the epithelial cells.
3. Distribute the forces over a wide area and provide mechanical linkages.

The oral epithelium of both types keratinized and nonkeratinized, consists of four layers.

Keratinizing epithelium consists of the following.

1. Basal layer (Stratum basale).
2. Prickle cell layer (Stratum spinosum).

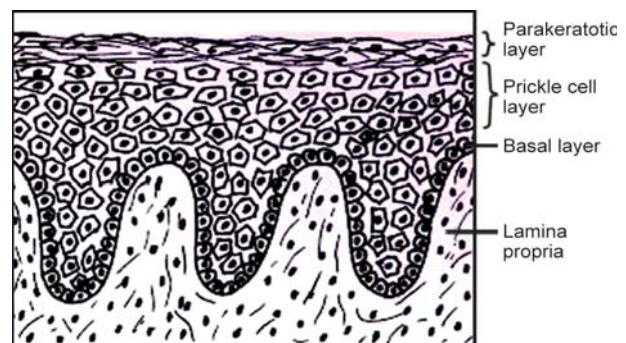


Figure 9.4: Parakeratinized oral mucosa (as seen under light microscope)

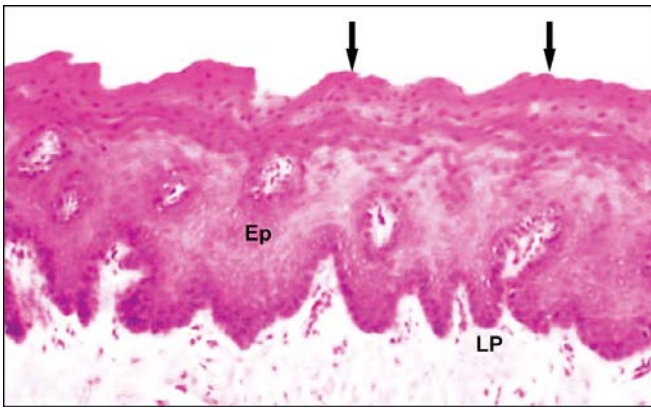


Figure 9.5: In parakeratotic epithelium (Ep) the superficial layer consists of 3 to 4 layers of flattened cells with deep pink cytoplasm and flattened to pyknotic nuclei (indicated by arrows) (LP) Lamina propria. (H & E x 200)

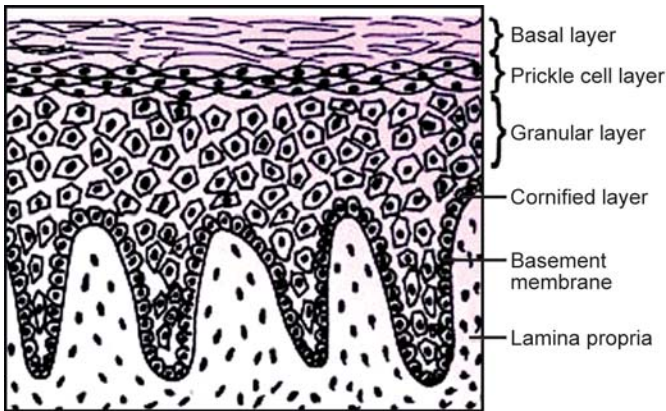


Figure 9.6: Layer of keratinized or orthokeratinized gingival epithelium (as seen under light microscope)

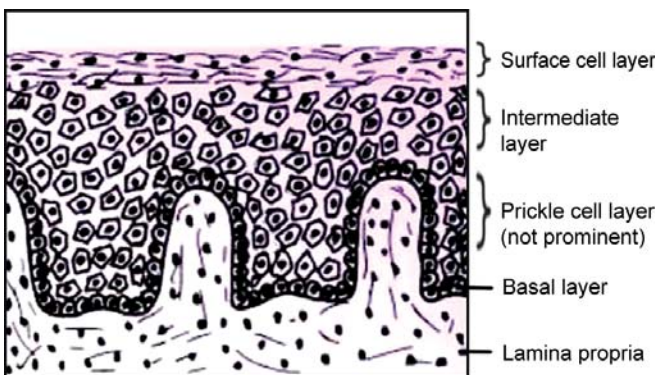


Figure 9.7: Layer of nonkeratinized oral mucosa (cheek mucosa), (as seen under light microscope)

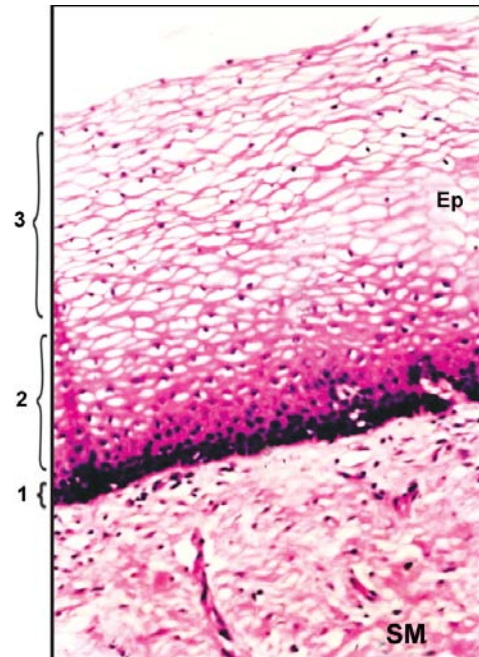


Figure 9.8: Nonkeratinizing squamous epithelium: There is no keratinization of epithelium. It consists of 4 layers x 150. (Ep) Epithelium, (1) Stratum basale or basal layer, (2) Stratum intermedium or intermediate layer, (3) Stratum superficiale or superficial layer, (SM) Submucosal fibrous tissue

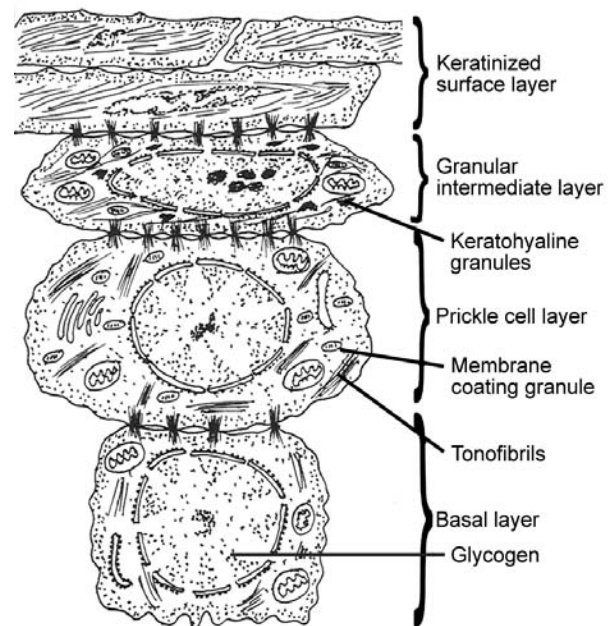


Figure 9.9: Principal structural features of cells in different layers of orthokeratinized epithelium (as seen under Electron Microscope)

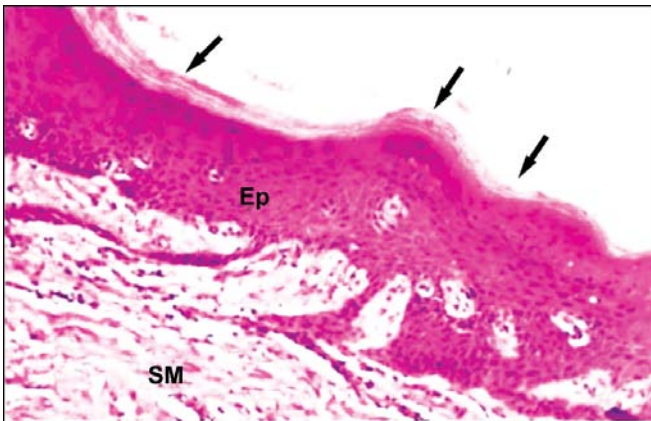


Figure 9.10: In keratinizing epithelium (Ep), the superficial layer of cells is covered with a layer of enucleated thin flattened glassy looking cells (indicated by arrows). This layer is rich in keratin protein. (SM) Submucosa (H & E x 100)

3. Granular layer (Stratum granulosum).
 4. Cornified layer (Stratum corneum).
- Nonkeratinizing epithelium consists of following.
1. Basal layer (Stratum basale)
 2. Prickle cell layer (Stratum spinosum)
 3. Intermediate layer (Stratum intermediae)
 4. Surface cell layer (Stratum superficiale)

These layers are named according to their situation (no. 1, 3 and 4) and the morphology of the cells no. 2. At different time, a single cell is a part of each layer. After mitosis, either it may remain in basal layer and divide again or it migrates and is pushed upward. The cell which migrate is a keratinocytes. Certain biochemical and morphologic changes occurs in this keratinocyte and forms keratinized squama. This is a dead cell filled with densely packed protein contained within a toughened cell membrane. It reaches the surface and desquamates after a while. This whole process is known as keratinization.

Epithelium

Keratinization is a process by which epithelial cells exposed to the external environment lose their moisture and are replaced by horny tissue. This horny tissue contains keratin which is fibrous in nature and contains protein. This protein is insoluble in most solvents including gastric juice. Keratinized epithelium is more resistant to infections and irritations than nonkeratinized epithelium (Fig. 9.11).

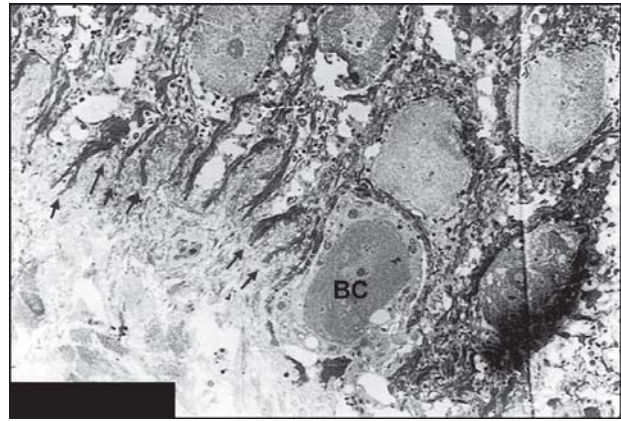


Figure 9.11: Oral epithelium- Basal layer: Hemidesmosomes indicated by arrows anchor the basal cells (BC) to the basement membrane (Electron micrograph x 3800)

Basal Layer (Stratum Basale)

Basal layer is also called as the proliferative or germinative layer. The cells of the basal layer are capable of division. The basal layer consists of a layer of cuboidal or columnar cells adjacent to the basement membrane (Figs 9.6 to 9.8). The cells of the basal layer synthesize DNA and undergo mitosis, providing new cells. So, new cells are generated in the basal layer. The basal cells are of two types. One type is serrated and is packed with tonofilaments. These are adapted for attachment. The other type is non-serrated and is made up of slowly cycling stem cells. These cells give rise to cells which are amplified for cell division.

The serrated basal cells are a single layer of cuboid cells. These have protoplasmic process, which project from their basal surface towards the connective tissue. On the basal surface hemidesmosomes are present which abut on basal lamina, that is connect epithelium to the connective tissue (Figs 9.12 A and B).

Adjacent basal cells are connected by desmosomes. These consists of adjacent cell membrane and or pair of denser region (attachment plaque). Other types of cell junctions may be present, these are, tight, close and gap junctions. The presence of ribosomes and rough endoplasmic reticulum indicates the protein synthesizing activity. Basal cells synthesize some proteins of basal lamina

The basal cells and parabasal spinous cells are referred to as stratum germinativum but only the basal cells can divide.

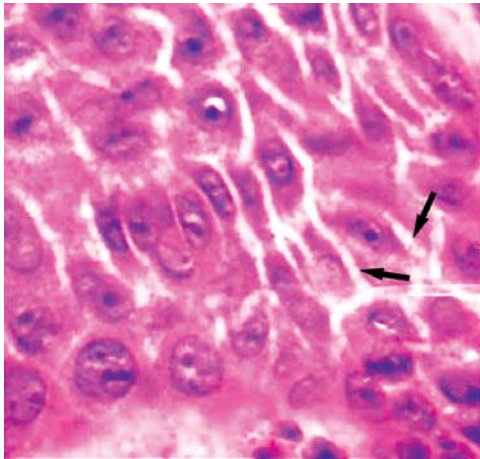


Figure 9.12A: Microscopic section of oral epithelium showing intercellular bridges (shown by arrow) traversing the space between two adjacent epithelial cells (H&E x 400)

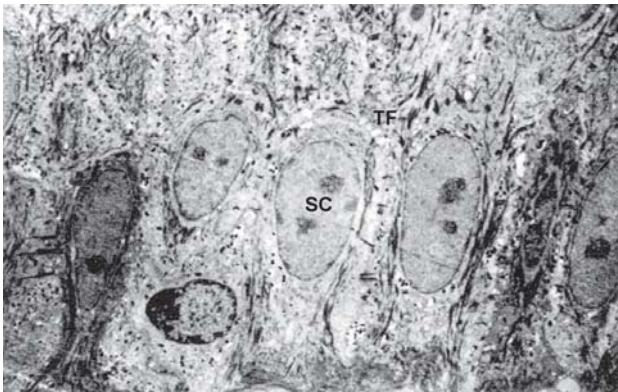


Figure 9.12B: Oral epithelium showing tonofilaments (TF) in the cytoplasm of squamous cells (SC) (in stratum spinosum) (Electron micrograph x 3800)

Main features of basal layer stratum basale: Cuboidal or columnar cells contain bundles of tonofibrils and other cell organelles. It is a site of cell division.

Prickle Cell Layer (Stratum Spinosum)

Stratum Spinosum is also called the prickle cell layer, as the shape of cells on histological examination is like a prickle (Figs 9.6 to 9.8).

The spinous cells are irregularly polyhedral and these are larger than the basal cells. The cells are joined by intercellular bridges. Tonofibrils cross from one cell to the next across intercellular bridges. These tonofibrils are bundles of tonofilaments that run next to the attachment plaque and do not crossover into the adjacent cells. The tonofilaments are joined to attachment plaques by an agglutinating substance (Figs 9.12A and B and 9.13). The

desmosome attachment plaque contain polypeptides desmoplakin I and II. Monoclonal antibodies to these polypeptides can be used to detect carcinomas by immunocytochemistry.

The intercellular spaces contain glycoprotein, glycosaminoglycans and fibronectin. The tonofilament network and desmosomes appear to make up a supporting system for the epithelium.

The basal cells of the gingiva and the palate contain a greater number of hemidesmosomes than the alveolar mucosa, buccal mucosa and tongue.

The desmosomes become more prominent because the intercellular spaces of the spinous cells in the keratinizing epithelia are large and distended. When prepared for histologic examination, cells of stratum spinosum shrink away from one another, remaining in contact only at point known as intercellular bridges or desmosomes. This arrangement gives the cells a spiny or prickle-like profile.

Acanthosis and Acantholysis

Acanthosis: Acanthae is a word used for prickle cell layer. Acanthosis is an increase in thickness of prickle cell layer in pathologic conditions.

Acantholysis: Separation of cells due to loss of intercellular bridges.

The tonofilament network and desmosomes appear to make up the tensile supporting system for epithelium. Amongst the four layers, spinous cells are most active in protein synthesis. Proteins produced by stratum spinosum are of great help in the keratinization process. The basal cells and prickle cells together constitute from half to two thirds of the thickness of epithelium.

Granular Layer (Stratum Granulosum)

This layer contains flatter and wider cells. These cells are larger than spinous cells. This layer contains a number of small granules that stain intensely with basic dyes, such as hematoxylin. These granules are called keratohyalin granules (basophilic in nature). Keratohyalin granules contain two types of proteins, one is rich in the amino acid histidine. Tonofilaments are denser in quantity and are found in association with keratohyalin granules.

The nuclei in the cells of this layer show signs of degeneration and pyknosis. This layer also synthesizes protein but in lesser quantity and at different rate. Cells of stratum granulosum are more regularly arranged and more closely applied to the adjacent cell surface.

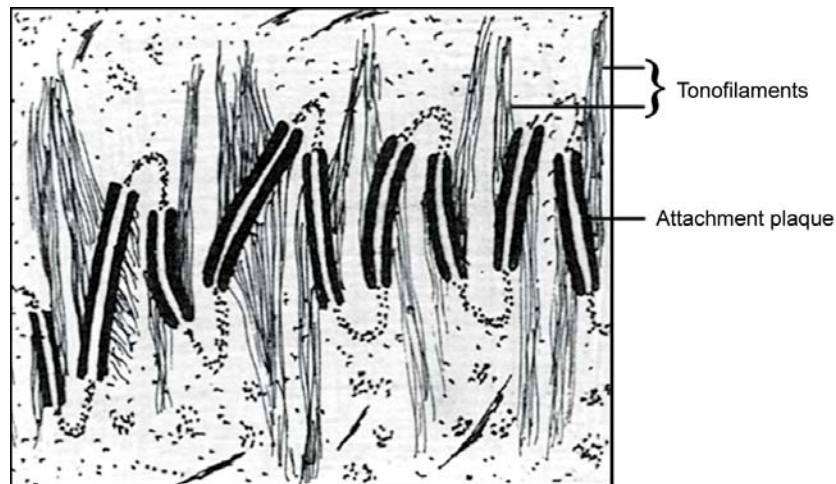


Figure 9.13: Prickle cell layer in keratinized oral epithelium with intercellular bridges between adjacent cells (As seen under electron microscope)

Odland body or Keratinosome: Keratinosomes are also called membrane-coating granules. They are formed in the upper spinous and granular cell layers. These are small membrane bound structures, 250 nanometers in size and originate from the Golgi bodies system. In keratinized epithelium, the granules are elongated and contain a series of parallel lamellae. In non-keratinized epithelium, they appear to be circular.

Lamellar granules or keratinosomes discharge their contents into intercellular space forming an intercellular lamellar material. It contributes to form a permeability barrier between the stratum corneum and the stratum granulosum. The intercellular space of this region has a lamellar structure that resembles the structure of the lamellar granular, and contains glycolipid.

Involucrin (keratolinin) is a sulfur rich protein, present in upper half of the stratum spinosum, which contributes to thickening of the inner unit of cell membrane, forming a 'cornified cell envelope'; The crosslinking of involucrin, periplakin and envoplakin form a scaffold on which loricrin and S P R R (small proline rich protein) are added. Sulfur rich proteins are stabilized by covalent crosslinks. Genes of cornified layer are located in I q 21 region of chromosome and called as epidermal differential complex. This is a highly resistant structure. This change occurs at about the same time of formation of the permeability barrier.

A smaller organelle similar to lamellar granule forms in the non-keratinizing epithelium. The only difference being that its contents are granular rather than lamellar. It may however, perform a similar function.

Cornified Layer (Stratum Corneum)

This layer is keratinized and the cells are larger and flatter than granular cells and are eosinophilic in nature. In this layer, all the nuclei and other organelles like mitochondria and ribosomes disappear (Figs 9.6, 9.7, 9.9 and 9.10). The cells are composed of densely packed filament coated by basic protein of keratohyalin granule, filaggrin. Disulfide bonds closely pack the tonofilaments and provide chemical and mechanical resistance to this layer.

There are two types of maturation of keratinized cells.

1. Orthokeratinization or True keratinization
2. Parakeratinization

Sometimes part of the hard palate and usually all of the gingiva are parakeratinized.

In parakeratinized epithelium, surface layer stains for keratin but importantly retains shrunken or pyknotic nuclei in many or all of the squamous epithelium. Keratohyalin granules are present in parakeratinized areas but fewer than orthokeratinized areas.

The keratinized cells become compact and they cover a greater surface area than the basal cells from which they develop. Importantly the keratinized layer does not synthesize protein.

Keratocytes or keratinocytes: Keratocytes or keratinocytes is the name given to the epithelial cells that ultimately keratinize. Keratinocytes increase in volume in each successive layer from basal to granular.

Major features: Cells of the keratinized epithelium are extremely flattened and dehydrated. All the organelles are lost and cells are packed with keratin.

DIFFERENCES BETWEEN KERATINIZED AND NONKERATINIZED EPITHELIUM (TABLE 9.1)

1. In the non-keratinized epithelium also there are four layers, but in place of the granular layer (stratum granulosum) and cornified layer (stratum corneum) there are the intermediate layer (stratum intermedium) and the surface cell layer (stratum superficiale).
2. Intercellular spaces are less conspicuous in the non-keratinized epithelia as compared to the keratinized, hence the cells do not have a prickly appearance.
3. Above the prickle cell layer (stratum spinosum), non-keratinized epithelium has no conspicuous differentiating feature between stratum intermedium and stratum superficiale, thus generally only three layers are prominent that is basal layer, intermediate layer and surface cell layer.
4. Non-keratinizing epithelia do not produce cornified surface layer and do not stain intensely with eosinophilic stain.
5. Superficial layer cells contain nuclei, which are quite big and plump and not flattened.
6. Turnover and mitotic ratio of the nonkeratinizing epithelium is higher than keratinized epithelium.
7. The surface layer of non-keratinized epithelium consists of cells filled with loosely arranged filaments that are not dehydrated. The surface of non-keratinized epithelium is flexible and can tolerate compression and distension as the non-keratinizing epithelia are present in cheeks and floor of mouth (Figs 9.5 and 9.14 to 9.16, Table 9.1).

Common Factors Affecting Keratinization

1. *Linea alba*: Sometimes a non-keratinized area in the cheek opposite the occlusal plane, gets keratinized due to continuous stress of friction. This line is called the linea alba. Continuous stress of friction causes linea alba.
2. *Smoking*: The palate of smokers-becomes hyperkeratotic due to irritation produced by tobacco smoke.

Table 9.1: Differences between keratinized and nonkeratinized epithelium

Keratinized Epithelium		Nonkeratinized Epithelium	
Present as Masticatory mucosa, Gingiva, Hard Palate, Vermilion buccolingual border.		Present as lining mucosa and specialized mucosa.	
Cell Layer	Features	Cell Layer	Features
1. Basal	(A) Cells cuboidal or columnar, (B) Cells contain bundles of tonofibrils and other cell organelles, (C) Site of most cell divisions.	1. Basal	(A) Cells cuboidal or columnar, (B) Cells contain separate tonofilaments and other cell organelles, (C) Site of most divisions.
2. Prickle cell	(A) Cells larger ovoid, (B) Cells contain conspicuous tonofibril bundles, (C) Membrane-coating granules appear in upper part of this layer.	2. Prickle cell	(A) Cells larger ovoid, (B) Cells contain dispersed tonofilaments, (C) Membrane-coating granules appear in upper part of this layer, (D) Filaments increase in number.
3. Granular	(A) Cells flattened, (B) Cells contain conspicuous keratohyaline granules in association with tonofibrils, (C) Membrane-coating the granules fuse with cell membrane in upper part, (D) Thickening of internal membrane also occurs.	3. Intermediate	(A) Cells slightly flattened, (B) Cells contain many dispersed tonofilaments and glycogen.
4. Keratinized	(A) Cells extremely flattened and dehydrated, (B) In cells all organelles and nuclei have been lost, (C) Cells filled only with packed fibrillar material, (D) When pyknotic nuclei are retained, it is called parakeratinization.	4. Superficial	(A) Cells slightly flattened, (B) Cells with dispersed filaments and glycogen are present, (C) Fewer organelles are present, (D) Nuclei persist.

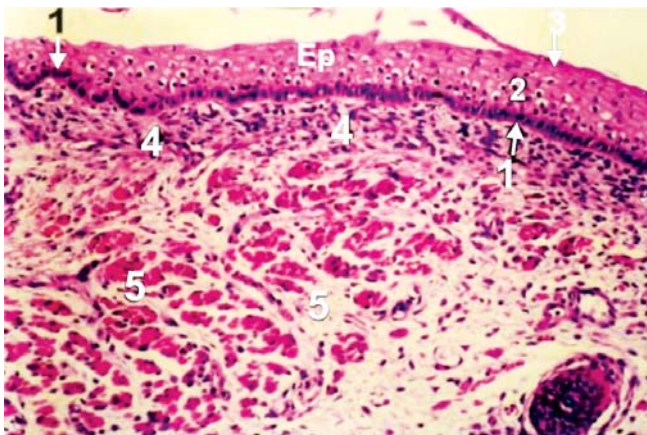


Figure 9.14: Non-keratinizing epithelium (Ep) of fetal oral mucosa consisting of 5 to 6 layers of cells. Basal layer (1) is single layer of columnar cells. The cells above this layer are polyhedral with central nuclei and perinuclear clear space due to presence of glycogen. The topmost layer of epithelium also shows presence of nuclei. Keratinization is not present. (2) Stratum intermedium, (3) Stratum superficiale, (4) Submucosa, (5) Muscle (H & E x 100)

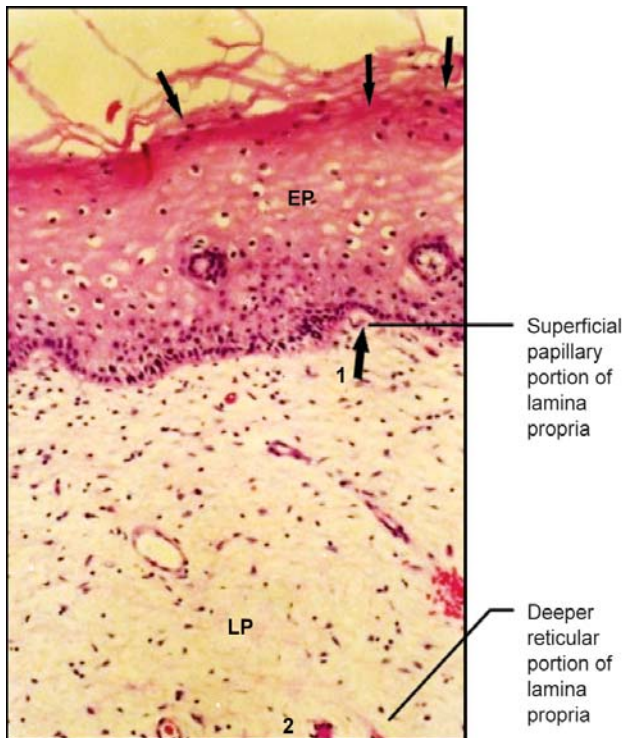


Figure 9.15: In nonkeratotic epithelium (Ep), the superficial layer consists of three to four layers of cells (indicated by arrows) Lamina propria (1) Superficial papillary portion of lamina propria. (2) Deeper reticular portion of lamina propria (H & E x 200)

3. Presence of chronic mild inflammation increases keratinization but severe inflammation reduces the degree of keratinization.

Keratosi and Parakeratosis

Keratosi: When keratinization occurs in a normally non-keratinized tissue it is called as keratosi.

Parakeratosis: When normally keratinized tissue such as the epidermi becomes parakeratinized it is called parakeratosis.

Table 9.2: Arterial blood supply to various part of the oral mucosa

Oral Region	Subterminal Branches
Upper lip	Superior labial artery (anastomoses with buccal artery)
Upper gingiva Anterior	Anterior superior alveolar artery
Lingual	Major palatine artery
Buccal	Buccal artery
Posterior Hard palate	Posterior superior alveolar artery Major palatine artery Nasopalatine artery Sphenopalatine artery
Soft palate	Minor palatine artery
Cheek	Buccal artery Some terminal branches of facial artery Posterior alveolar artery Infraorbital artery
Lower lip	Inferior labial artery (anastomoses with buccal artery) Mental artery Branch of inferior alveolar artery
Lower gingiva	
Anterior buccal	Mental artery
Anterior lingual	Incisive artery and sublingual artery
Posterior lingual	Inferior alveolar artery and sublingual artery
Posterior buccal	Inferior alveolar artery and buccal artery
Floor of mouth	Sublingual artery Branch of lingual artery
Tongue (dorsal and ventral surfaces)	
Anterior two thirds	Deep lingual artery
Posterior third	Dorsal lingual artery, to base of tongue, about posterior third

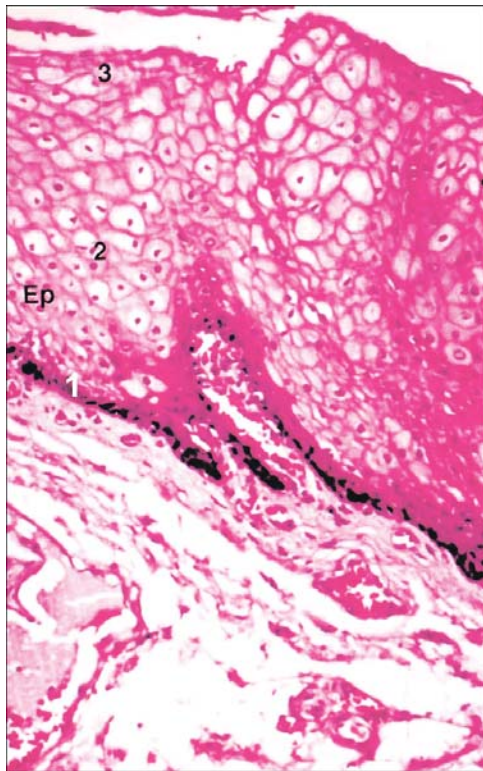


Figure 9.16: Nonkeratinizing epithelium: Epithelial lining (Ep) of adult cheek showing three layers of nonkeratinizing epithelium. (1) Basal layer, (2) Intermediate layer, (3) Superficial layer. The latter shows presence of polyhedral cells with no attempt at formation of parakeratin or keratin (H & E x 200)

CONNECTIVE TISSUE

Lamina Propria

1. The lamina propria is the connective tissue supporting the oral epithelium. The lamina propria at different regions is of different thickness (Fig. 9.15).
2. Lamina propria is divided into two parts: (i) superficial papillary portion (ii) deeper reticular portion.

Papillary portion is associated with epithelial ridges. The papillary layer consists of finger-like projections of connective tissue that interlock with similar epithelium projections (Fig. 9.1).

The reticular portion is named because of the reticular fibers. Because of the difference in the length and width of the papillae in different areas, the papillary portion is of variable depth. Basement membrane can be distinguished from the connective tissue of lamina propria as it takes silver stain.

The ridge and papillary arrangement helps to increase the attachment area. It also increases the strength of bond

between the epithelium and the lamina propria. It facilitates exchange between the epithelium and blood vessels. Reticular portion is associated with a net-like arrangement of collagen fibres. Though collagen fibers are also present in the papillary layer but these are thin as compared to the reticular layer where the collagen fibers are thick and dense and are arranged parallel to the surface plane.

In the alveolar mucosa, the papillary arrangement is missing, but reticular portion is present in every area. The lamina propria may attach to periosteum of the alveolar bone. It may also overlay the submucosa.

Submucosa

Submucosa is a connective tissue layer that attaches the lamina propria of the oral mucosa to the underlying bone or muscle. Submucosa is of variable thickness and density. The character of submucosa ascertains whether the attachment is loose or firm.

Glands, blood vessels, nerves and adipose tissue are present in the submucosa. Large blood vessels enter the submucosa and divide here into smaller branches, which then enter the lamina propria, where these branches again divide into further smaller branches. In this way, a subepithelial capillary network in the papillae is formed. Lymph vessels also accompany blood vessels.

Nerve Supply: The nerve supply to the oral mucosa is sensory. The nerve fibers are myelinated at first but as they traverse through the submucosa, they lose their myelin sheath before splitting into their ends. Various types of nerve endings are found in the papillae. Some fibers enter the epithelium and they may terminate between epithelial cells as free nerve endings. The smooth muscles of the blood vessels are supplied by non-myelinated visceral nerve fibers. They form a network in the reticular layer of the lamina propria that finally terminates in a subepithelial plexus.

The sensory nerve terminates into nerve endings, both free and organized. Free nerve endings are found in the lamina propria and within the epithelium. The primary sensations perceived in the oral cavity are those of warmth, cold, touch, pain and taste. The anterior parts of the tongue, lips and the hard palate are most sensitive to touch, as the nerve endings are mostly concentrated in these regions. Temperature perception is more acute in the vermilion border of the lip, at the tip of tongue and the anterior region of the palate (Tables 9.2 and 9.3).

Table 9.3: Principal sensory nerve fibers supplying the oral mucosa

Oral Region	Innervation
Upper lip and vestibule	Twigs from infraorbital branch of maxillary nerve.
Upper gingivae	Anterior, posterior, and (when present) middle superior alveolar branches of maxillary nerve.
Hard palate	Greater, lesser, and sphenopalatine branches of maxillary nerve.
Soft palate	Lesser palatine branch of maxillary nerve; tonsillar branch of glossopharyngeal nerve; and nerve of pterygoid canal (taste; originating from facial nerve).
Cheek	Twigs from infraorbital branch of maxillary nerve; superior alveolar branch of maxillary nerve; buccal branch of mandibular nerve; and possibly some terminal branches of facial nerve.
Lower lip and vestibule	Mental branch of inferior alveolar nerve and buccal branch of mandibular nerve.
Lower gingivae: buccal, lingual	Inferior alveolar branch of mandibular nerve; buccal branch of mandibular nerve; and sublingual branch of lingual nerve.
Anterior two thirds of tongue	Lingual branch of mandibular nerve (taste) provided by fibers carried in lingual nerve but originating in facial nerve and passing by way of chorda tympani to lingual nerve.
Posterior third of tongue, facial, and tonsillar	Glossopharyngeal nerve (taste and general sensation).

SUBDIVISIONS OF ORAL MUCOSA ON THE BASIS OF KERATINIZATION

On the basis of keratinization oral mucosa is divided into the following areas (Fig. 9.17, Table 9.4).

- A. Keratinized areas
 - 1. Masticatory mucosa
 - a. Hard palate
 - b. Gingiva
- B. Non-Keratinized areas
 - 1. Lining mucosa
 - a. Soft palate
 - b. Ventral surface of tongue
 - c. Floor of the oral cavity
 - d. Lips and cheek
 - e. Vestibular fornix and alveolar mucosa
 - 2. Specialized mucosa
 - a. Dorsal lingual mucosa
 - b. Taste buds

Keratinized Areas

Masticatory Mucosa

The areas that bear masticatory force and compression form the masticatory mucosa. These areas are the (a) Hard palate (b) Gingiva.

The epithelium of the masticatory mucosa is thicker than other regions and is generally orthokeratinized or

parakeratinized. Masticatory mucosa has the greatest number of papillae per unit area of mucosa. In the lining mucosa, papillae are shorter and fewer. Parakeratinized areas are generally found in areas of gingiva.

Hard palate: Histologically, the masticatory mucosa of the hard palate can be divided into three parts, each part having characteristic histologic features (Figs 9. 17 A to D and 9.18).

Covering epithelium: The epithelium of the hard palate is thick orthokeratinized [some parts may show parakeratinization], stratified squamous epithelium and anterolaterally it is thrown into transverse palatine ridges or rugae.

Lamina Propria: It characteristically shows long papillae and dense network of thick collagen tissues especially under rugae. The blood supply is moderated by short capillary loops.

Submucosa: It tightly attaches the mucosa to the periosteum by dense collagen fibers, so, it is immovable. Fat and minor salivary glands are present in the submucosal layer. The mucous membrane of the hard palate is inextensible and tightly fixed to the underlying periosteum and well adapted to withstand abrasion (Fig. 9.19). It is pale pink in color like the gingiva.

The junction between the epithelium and the underlying lamina propria is convoluted or corrugated, and the papillae are long and elongated to prevent the epithelium from being tipped under masticatory force.

Table 9.4: Important feature of main layers oral mucosa in different regions of the oral cavity

Region	Covering Epithelium	Lamina Propria	Submucosa
I. Masticatory Mucosa			
A. Hard Palate	a. Orthokeratinized (often parakeratinized in parts), stratified squamous epithelium b. Thick, palatine, ridges (rugae).	a. Thick, dense collagenous tissue, especially under rugae, b. Long papillae, c. Moderate vascular supply.	a. Dense collagenous connective tissue attach mucosa to periosteum.
B. Gingiva	a. Orthokeratinized or parakeratinized, stratified squamous epithelium with stippled surface b. Thickness (250 micron).	a. Dense collagenous connective tissue, b. Moderately vascular, c. Long, narrow papillae.	a. Mucosa firmly attached by collagen fibers to cementum and periosteum, b. No distinct layer.
II. Lining Mucosa			
A. Floor of Oral Cavity	a. Nonkeratinized, stratified squamous epithelium, b. Very thin (100 micron).	a. Short papillae, b. Few elastic fibers, c. Rich vascular supply.	a. Loose fibrous connective tissue with minor salivary glands and fat.
B. Labial and Buccal Mucosa	a. Nonkeratinized, stratified squamous epithelium, b. Very thick (500 micron).	a. Dense fibrous connective tissue, b. Long, slender papillae, c. Rich vascular supply.	a. Mucosa firmly attached to underlying muscle, b. Dense collagenous connective tissue, c. Plenty of minor salivary glands.
C. Alveolar Mucosa	a. Nonkeratinized, stratified squamous epithelium, b. Thin.	a. Connective tissue containing many elastic fibers, b. Capillary loops close to the surface, c. Short papillae.	a. Loose connective tissue, containing thick elastic fibers, b. Minor salivary glands.
D. Soft Palate	a. Nonkeratinized stratified squamous epithelium, b. Thin (150 micron), c. Taste buds present.	a. Thick with many short papillae, b. Elastic fibers forming an elastic lamina, c. Highly vascular.	Diffuse tissue with plenty of minor salivary glands.
E. Ventral Surface of Tongue	a. Thin, b. Nonkeratinized, c. Stratified squamous epithelium.	a. Thin with plenty of short papillae and some elastic fibers, b. Capillary network in subpapillary layer, c. A few minor salivary glands.	Thin and irregular.
F. Lips: Vermilion Zone	a. Orthokeratinized, stratified squamous epithelium, b. Thin.	a. Capillary loops close to surface, b. Plenty of narrow papillae.	a. Mucosa firmly attached to underlying muscle.
G. Lips: Intermediate Zone	a. Parakeratinized, stratified squamous epithelium, b. Thin.	a. Elastic and collagen fibers in connective tissue, b. Irregular papillae.	
III. Specialized Mucosa			
Dorsal Surface of Tongue	a. Keratinized and nonkeratinized, stratified squamous epithelium, b. Thick, c. Form three types of lingual papillae some bear taste buds.	a. Minor salivary glands in posterior portion, b. Long papillae, c. Rich innervation especially near taste buds.	a. Mucosa is bound to connective tissue, b. No distinct layer.

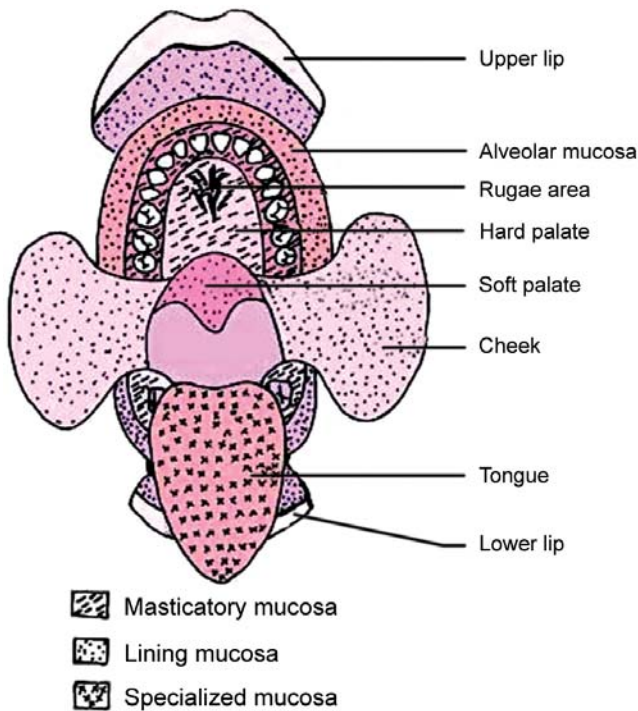


Figure 9.17A: Anatomic locations of three main types of oral mucosa, masticatory mucosa, lining mucosa and specialized mucosa

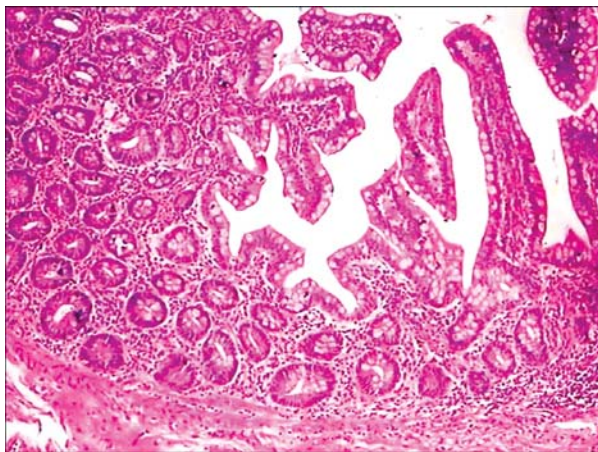


Figure 9.17B: Intestinal mucosa. Intestinal mucosa contains villi and glands and lamina propria (loose fibrous tissue between glands), the submucosa is separated from mucosa by a thin layer of muscle called muscularis mucosae unlike in oral mucosa where lamina propria merges into submucosa (H & E x 100)

Because of the varying structure of the submucosa, there is difference in various regions of the hard palate.

Zones of Hard Palate

1. Gingival region - adjacent to teeth
2. Palatine raphe - extending from the incisive or palatine papilla posteriorly to soft palate

3. Anterolateral fatty zone between palatine raphe and gingiva
4. Posterolateral glandular area between raphe and gingiva

i. *Gingival area and the median palatine raphe:* These do not have the submucous layer beneath the lamina propria and they are directly attached to the bone. The thick layers of the submucosa are present in a wide region extending between the palatine gingiva and the palatine raphe. In this region, a dense band of fibrous connective tissue joins the lamina propria to the mucoperiosteum. Thus, despite the heavy band

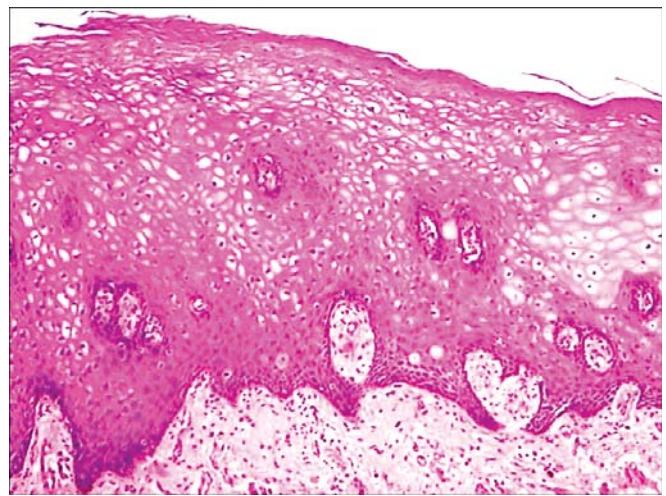


Figure 9.17C: Nonkeratinizing mucosa: Epithelium is made up of polyhedral cells. Intercellular bridges are not prominent. Surface layer does not show pink hyaline structure (H & E x 100)



Figure 9.17D: Masticatory keratinized mucosa: Mucosa has prominent intercellular bridges and is covered by a layer of pink cellular keratin epithelial ridges are long. (H & E x 100)

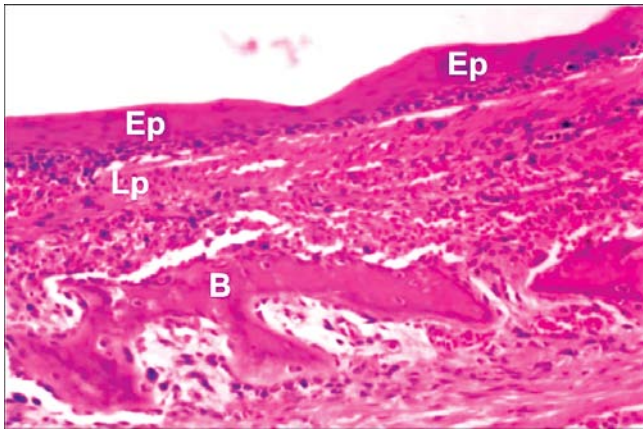


Figure 9.18: Hard Palate: in 14-week-old fetus: Epithelium (Ep) is resting on the fibrous tissue of lamina propria (LP) which is attached to the underlying bony plate (B) (H & E x 150)

of submucosa, the mucous membrane is immovably attached to the periosteum of the maxillary and palatine bones.

The thick submucosa in the palatal region is divided into irregular compartments filled with adipose tissue in the anterior part and with glands in the posterior part of the hard palate. The glandular zone extends onto the soft palate (Figs 9.17 and 9.19).

The oral mucosa of the hard palate contains the anterior palatine nerves and blood vessels that pass and course through the junction of the alveolar process and the horizontal plate of the hard palate.

- ii. *Incisive papilla*: In the midline of the hard palate, immediately posterior to the maxillary central incisors is a dense elevation called the incisive papilla. This small projection of tissue is composed of dense connective tissue. It overlies the oral opening of the incisive canal. The incisive canal contains remnants of the oral part of the nasopalatine duct, nasopalatine nerve and vessels. This duct is lined by simple or pseudostratified columnar epithelium which are rich in goblet cell (Fig. 9.20).

The nasopalatine ducts are vestigial in human beings and functional in the lower mammals as a part of Jacobson's organ of olfaction. The Jacobson's organ is a small ellipsoid (cigar-shaped) structure lined by olfactory epithelium that extends from the nose to the oral cavity. In humans, Jacobson's organ appears from the 12th to 15th weeks of fetal life after which it degenerates.

In humans, the anterior part of the papilla contains cartilage. But this has no bearing on the nasopalatine

ducts. Sometime in humans, the nasopalatine ducts get pathologically involved to form the nasopalatine cyst.

- iii. *Palatine rugae (Transverse palatine ridges)*: Mucosa of the anterior one-third of the hard palate is formed into elevated and irregular transverse fold of the palatine rugae. These extends laterally from the incisive papilla and anterior part of the raphae.

The rugae are supported laterally by a submucosal cushion of adipose tissue, thus forming a fatty anterolateral zone. The core of the rugae is made up of dense connective tissue layer that makes them immovable.

- iv. *Epithelial pearls*: These are circular or concentrically arranged, keratinized epithelia cells and are remnants of epithelium formed in the line of fusion of palatine processes. These may be found in the lamina propria, mostly in the region of incisive papilla (Fig. 9.21).

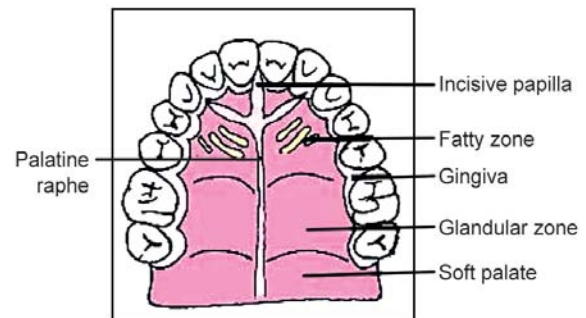


Figure 9.19: Surface view of hard and soft palates indicating various zones of palatine mucosa

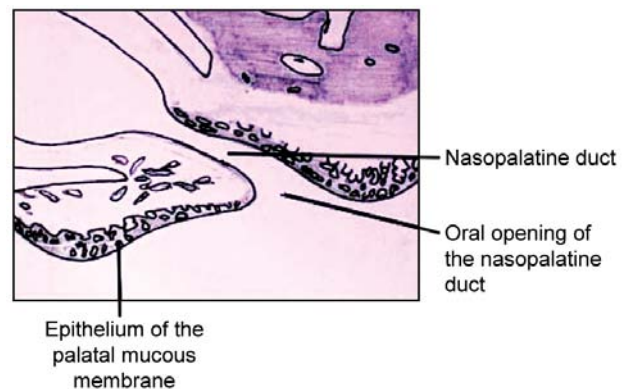


Figure 9.20: Nasopalatine duct as seen in histological section

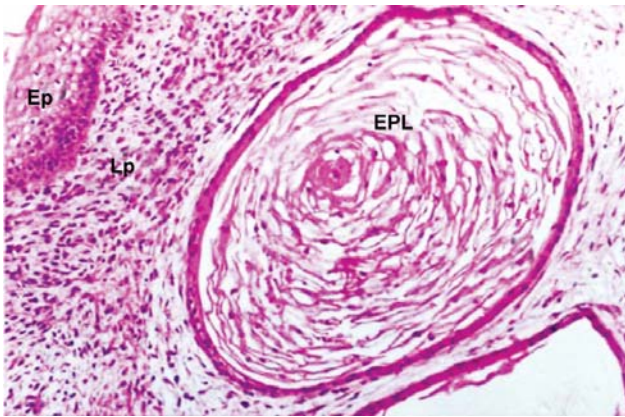


Figure 9.21: Epithelial pearl (EPL) with concentric layers of keratinized epithelial cells is seen in the lamina propria (LP). It represents remnant of epithelium (Ep) formed in the line of fusion of facial prominences during development (H & E x 200)

Gingiva

It is a part of the oral mucosa that surrounds the neck of erupted tooth and covers the alveolar process of the jaws.

Parts of Gingiva

Gingiva consists of the following two parts.

- The part facing the oral cavity is the masticatory mucosa. This part bears the masticatory stresses and is keratinized or parakeratinized.
- The part facing the tooth that forms the gingival sulcus and joins gingiva to the tooth. This is a part of the periodontium and is nonkeratinized.

The mucogingival junction separates the alveolar mucosa from the gingiva which is limited on the outer surface of both jaws. On the inner surface of the lower jaw a line of demarcation is present between the gingiva and mucosa on the floor of the mouth. On the palate, there is no sharp distinction between the gingiva and the peripheral palatal mucosa.

Gingiva is made up of stratified squamous epithelium that is most often parakeratinized and may be keratinized or non-keratinized (Fig. 9.22). The gingiva is normally pink but may sometimes have a grayish tint. Color of the gingiva depends on the following:

- The surface (whether keratinized or not)
- Thickness
- Pigmentation

Reddish or pinkish tint is due to the color given to underlying tissue by the blood vessels and the circulating blood. The gingiva is:

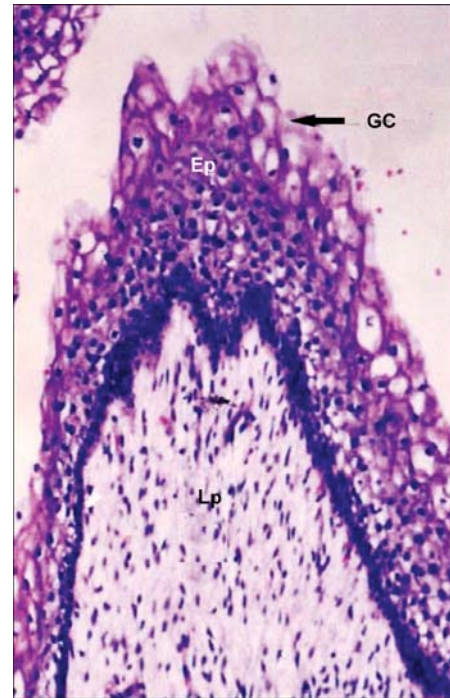


Figure 9.22: Gingiva: Gingival crest (GC) of 14-week-old fetus lined by squamous epithelium (Ep). The stromal tissue of lamina propria (LP) is made up of fibroblasts (H & E x 150)

- Parakeratinized in 75 percent cases, where superficial cells retain pyknotic nuclei.
- Keratinized in 15 percent cases, there superficial cells form scales of keratin and lose their nuclei.
- Nonkeratinized in 10 percent cases, where surface cells are nucleated and shows no signs of keratinization. Inflammation interferes with the keratinization. If the tissue is highly keratinized, it is whiter and less translucent. Apart from keratinocytes, the gingiva also contains three other types of cells.

1. Melanocytes (Derived from Neural Crest Cells)

Presence of melanin pigment gives it a brown or black pigmentation. This pigmentation is maximum at the base of the interdental papilla. It may increase in disease state. Melanin is stored by the basal cells in the form of melanosomes. But it is released by the melanocytes that reside in the basal layer (Fig. 9.23) and then it is transferred to the basal cells. Each melanocyte is connected with 30 to 40 keratinocytes by their dendritic processes. These processes transfer melanin from melanocytes to keratinocytes. Melanocytes are also known as clear cells or dendritic cells.

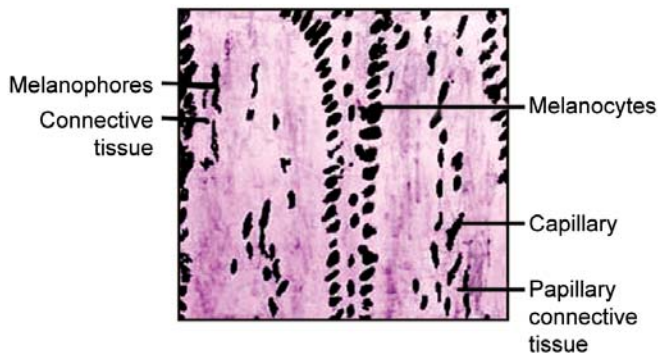


Figure 9.23: Pigmented gingiva showing melanocytes, melanophores and capillaries. Melanocytes are present in basal layer, melanophores in connective tissue and capillaries in papillary connective tissue

By dopa reactions or by silver staining techniques, oral pigmentation can be studied. In dopa reaction, the cells which contain tyrosinase enzyme appear dark. So, the melanin-producing cells, which contain tyrosinase can be seen. Silver stains also dye the melanin pigment.

2. Langerhans' Cells (Clear or Dendritic Cells)

These are found in the upper layer of the skin and the mucosal epithelium confined to areas of orthokeratinization. A Langerhans' cell is free of melanin and does not give a DOPA reaction. Both the melanocytes and Langerhans' cells do not form desmosomal attachments to the epithelial cells. Langerhans' cell is stained with gold chloride, ATPase and immunofluorescent markers. These cells are of hematopoietic origin and are involved in the immune response (Fig. 9.24). The cell contains convoluted nucleus and rodlike Birbeck granules. Langerhans cells are important in graft rejection, contact hypersensitivity and in anti-tumor immunity

3. Merkel Cells

These are the specialized neural pressure-sensitive receptor cells with nerve tissue lying just below them. Merkel cells are commonly observed among basal cells in masticatory mucosa and respond to touch sensation. Nucleus of Merkel cells show deep invagination and characteristic rodlet. Cytoplasm contains dense granules and intermediate type junctions are present between the axon terminals and Merkel cells. These are slow acting, have neurosecretory activity, and are migrant from the neural crest cells. The Merkel cells lack neural filaments but are neurally related.

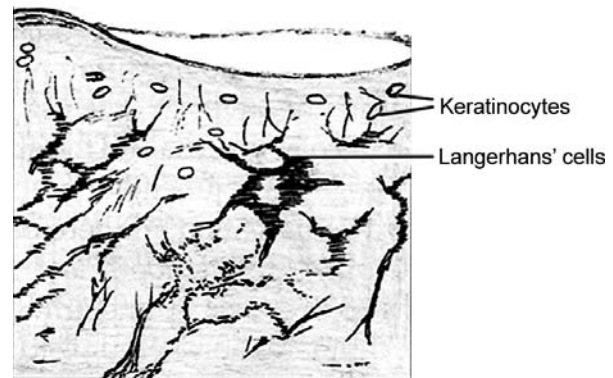


Figure 9.24: Oral aspect of gingival epithelium showing Langerhans' cells by immunoperoxidase technique

Lamina propria beneath the epithelium is dense and there is no submucosa beneath it (Fig. 9.25). The dense lamina propria of the gingiva does not contain large vessels. Numerous long and slender papillae are present in the gingiva that help in histologic differentiation between the gingiva and the alveolar mucosa (Fig. 9.26). This distinction between the alveolar mucosa and the gingiva can be made by the mucogingival junction/mucogingival line, which forms a demarcation line between the two. The alveolar mucosa is red, smooth and shiny rather than pink and stippled. The epithelium of the alveolar mucosa is thinner, non keratinized and contains no rete pegs. The connective tissue of the alveolar mucosa is loosely arranged and the blood vessels are numerous. This demarcation line is quite obvious on the facial surfaces of the gingiva and on the lingual surfaces of mandibular gingiva, where the demarcation line is between the gingiva and the floor of the mouth. On the palatal side, there is very faint demarcation between the gingiva and the palatal mucosa. Only a few elastic fibers are present in the connective tissue of the lamina propria, which are localized to the walls of the blood vessels. Oxytalan fibers are also present.

Divisions of Gingiva

Free or unattached gingiva: This is also called as marginal gingiva and is a terminal edge or border of the gingiva surrounding the teeth in a collar-like fashion. Generally it is demarcated from the attached gingiva by a shallow linear depression called the free gingival groove which runs parallel to the gingival margin at a distance of 0.5 to 1.5 millimeter. Free gingiva is usually 1 mm wide and it forms the soft tissue wall of the gingival sulcus.

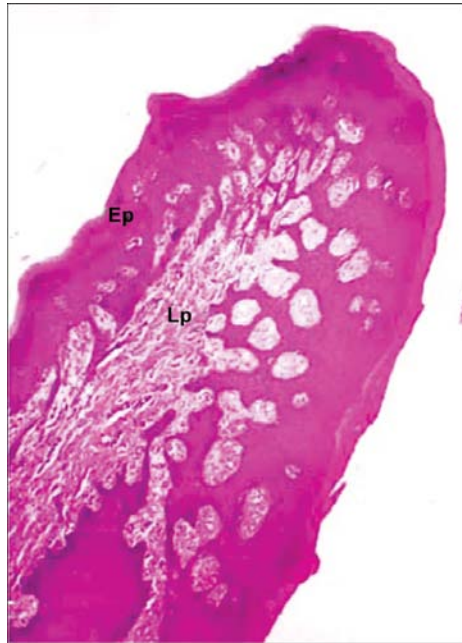


Figure 9.25: Section from interdental papilla of gingiva shows squamous epithelium (Ep) covered with thin layer of keratin. Rete ridges are elongated lamina propria (LP) is dense fibrous in nature (H & E x 50)

Gingival sulcus: It is a shallow space around the tooth bounded by the tooth surface on one side and the epithelium lining the free gingiva or the sulcular epithelium on another side. Gingival sulcus is V-shaped and under absolute normal conditions or in Gnotobiotic animals (Germ-free animals), its depth is about 0 mm (in this condition there is no free gingiva). In normal healthy individuals, the depth of gingival sulcus is 1.8 to 2 mm. The probing depth of the sulcus is different from its histologic depth. The histologic depth generally is not equal to the probing depth or the depth of penetration of probe. The probing depth of the clinically normal gingival sulcus is 2 to 3 mm (Fig. 9.27).

Attached gingiva: The attached gingiva is a region between the free gingival groove and the alveolar mucosa or the mucogingival junction. It is a continuation of the marginal gingiva but is firm, resilient and tightly bound to the underlying periosteum of the alveolar bone (Fig. 9.28).

Width of attached gingiva: It is the distance between the mucogingival junction and the projection on the external surface of the bottom of the gingival sulcus or periodontal pocket or the free gingival groove.

Width of the attached gingiva in the incisor region is the maximum and in the premolar region it is the minimum (Fig. 9.29).

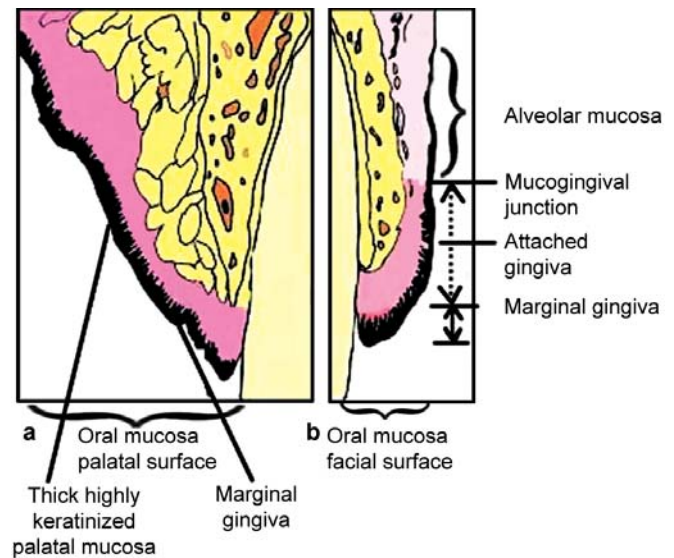


Figure 9.26A: Faciapalatal section of anterior tooth—Oral mucosa showing differences in (a) palatal surface and (b) facial surface

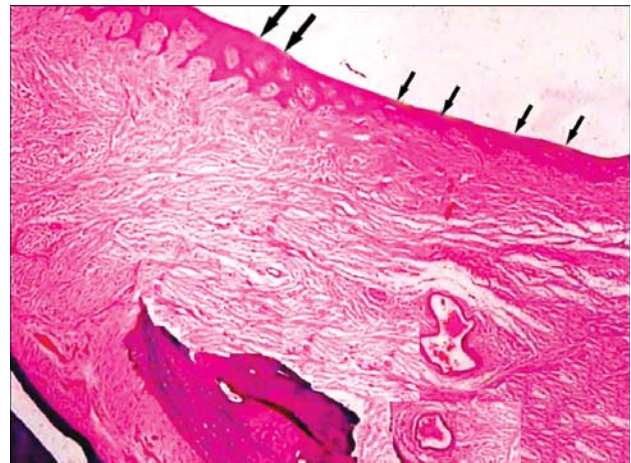


Figure 9.26B: Gingival mucosa (shown by long arrows) is thick and contains prominent rete ridges while alveolar mucosa (shown by short arrows) is thin and rete ridges are not conspicuous. x 40

Width in the incisors is 3.5 to 4.5 mm in maxilla 3.3 to 3.9 mm in mandible

Width in the premolars is 1.9 mm in maxilla and 1.8 mm in mandible

The width of the attached gingiva increases (a) with age and (b) in supra-erupted teeth. The reduction or increase in the width of the attached gingiva is due to the modifications in the position of its coronal end or free gingiva as the position of the mucogingival junction always remains constant.

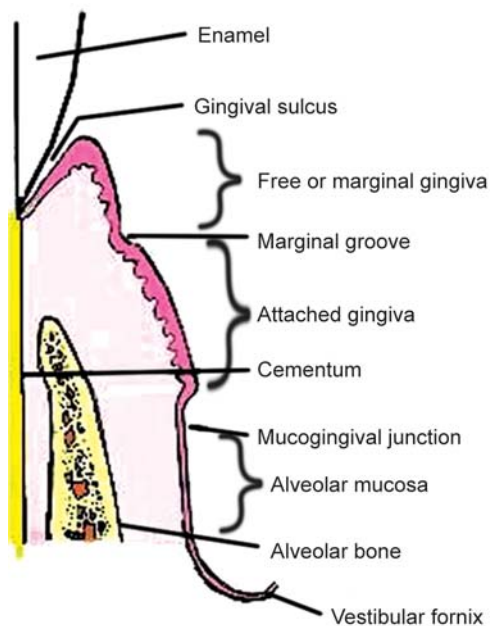


Figure 9.27: Section of gingiva showing divisions of gingiva and gingival sulcus

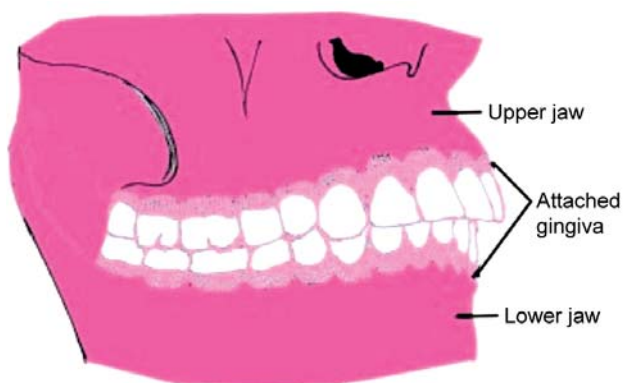


Figure 9.28: Mean width of attached gingiva in permanent dentition. Width of attached gingiva has been shown by light pink color

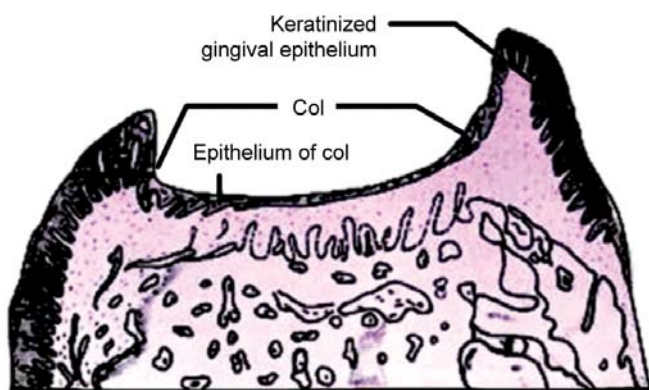


Figure 9.29: Histological section of gingiva through col

Interdental gingiva or interdental papilla: Interdental gingiva is that part of the gingiva that occupies the gingival embrasure or fills the space between the two adjacent teeth.

Interdental gingiva three dimensionally has various shapes (1) pyramidal shaped between anterior teeth (2) tent shaped between posterior teeth and (3) triangular shaped in vestibular view. Shape of interdental gingiva is directly dependent on the location and width of contact point between the two adjacent teeth.

- i. *Interdental grooves:* The gingiva appears slightly depressed between two adjacent teeth as it conforms to the depression on the alveolar process in this region. The depression in gingiva forms vertical folds called interdental grooves.
- ii. *Col:* In between the buccal and lingual peaks of interdental gingiva of posterior tooth generally the central concave area fits below the contact point. This depressed part of the interdental papilla is covered by thin non-keratinizing epithelium and is called 'col'.

It was at one time suggested that the col epithelium is highly susceptible and vulnerable to the initiation of periodontal disease, but now it is evident that the col epithelium is identical to the junctional epithelium and has the same origin as the dental epithelium. Thus, there is no evidence that the morphology of the col is vulnerable to disease (Fig. 9.29).

- iii. *Blood supply of gingiva:* The blood supply of the gingiva is derived from the periosteal vessels or branches of the alveolar arteries (Figs 9.30 and 9.31)

Blood vessels run through the interdental septa, perforate the septa through its crest and enter the gingiva and form loops within the connective tissue of gingiva supplying the dentogingival junction and the buccal and lingual gingivae.

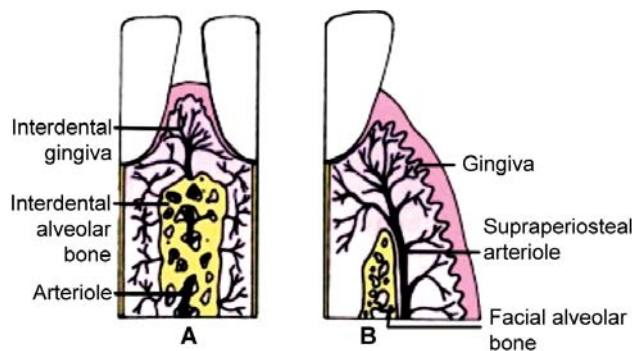


Figure 9.30: Arteriole penetrating the interdental alveolar bone to supply the interdental gingiva. (B) Suprapariosteal arteriole overlying the facial alveolar bone, spreading branches to the surrounding gingiva

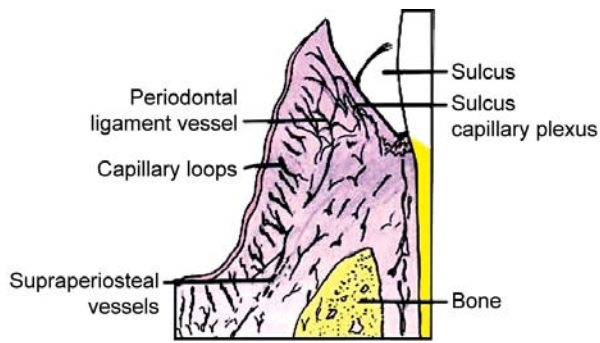
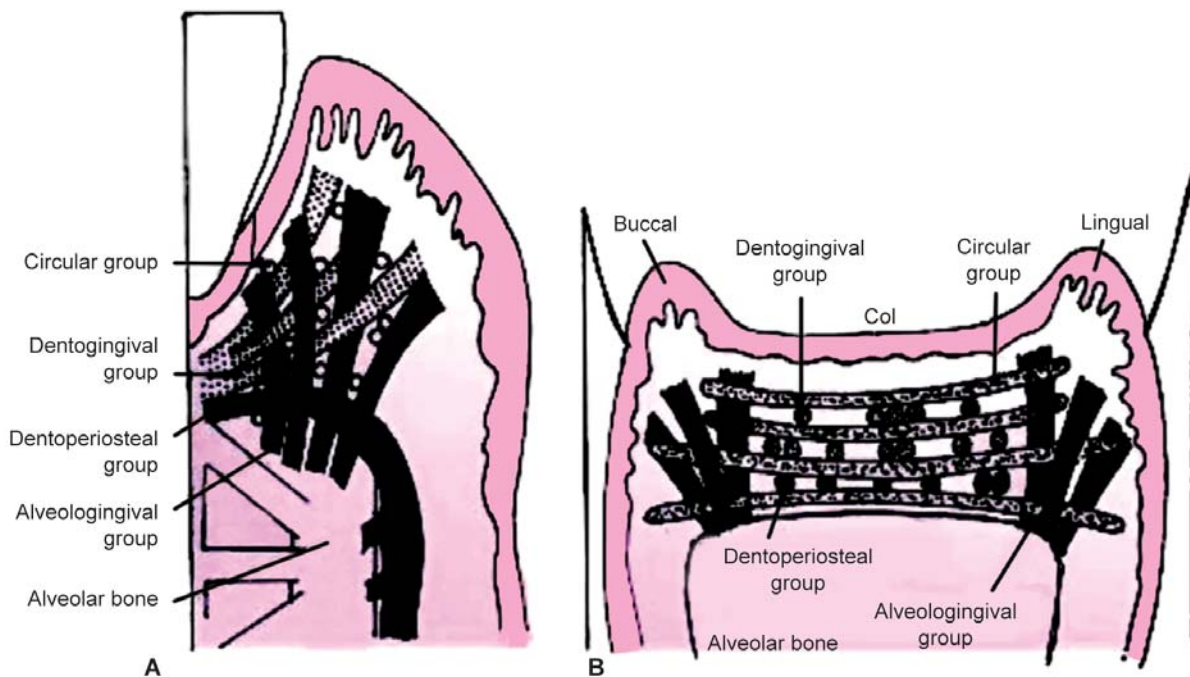


Figure 9.31: Peripheral vascular circulation of the gingiva. Sulcus capillary plexus is parallel to the sulcus. Capillary loops are present in the outer papillary layer. Supraperiosteal vessels are present external to the bone which supplies the gingiva. Periodontal ligament vessel anastomosing with the sulcus capillary plexus

- iv. *Lymphatic drainage of gingiva:* Lymph from the gingiva is drained in the submental and submandibular lymph nodes
- v. *Nerve supply of gingiva:* The gingiva is innervated by the terminal branches of the periodontal nerve fibers, originating from infraorbital, palatine, lingual, inferior alveolar, and buccal nerves. Nerve endings are Meissner or Krause corpuscles, end bulbs and loops.
- vi. *Age changes in gingiva:* With aging, apical migration of the dentogingival junction occurs.

Histologically, the epithelium becomes thinner and friable and flattening of the epithelium connective tissue junction occurs.

- vii. *Gingival fibers: (Gingival Ligament)* There are five groups of gingival fibers, which collectively form the gingival ligament (Fig. 9.32). These are the dense fibers of collagen and are divided into following major groups
 - a. Dentogingival
 - b. Alveologingival
 - c. Circular
 - d. Dentoperiosteal
 - e. Transseptal group.
- a. *Dentogingival:* It is also called as gingivodental. These are the most numerous of all the fibers. They extend from cervical cementum into the lamina propria of free and attached gingiva on all surfaces of teeth.
- b. *Alveologingival:* As the name suggests, they extend from the alveolar crest to the gingival margin.
- c. *Circular:* These form a band around the tooth and encircle it in a ring like fashion, interlace with other fibers and bind the free gingiva to the tooth.
- d. *Dentoperiosteal group:* These run from the tooth to the bone, from the cementum to the periosteum of the alveolar crest and insert either into the alveolar process or the muscles in the floor of mouth.



Figures 9.32A and B: A. Principal fiber groups of the gingival ligament; B. Fiber groups of the gingival ligament as seen interproximally related to the col

e. *Transseptal group*: These are sometimes described with the principal fibers of periodontal ligament. These arise from the area between base of gingival sulcus and alveolar crest from the cementum of one tooth and run interdentially over the alveolar crest and attach into the cementum of adjacent tooth.

Collectively these fibers form the transseptal fiber system. On the whole forming an interdental ligament which connect all the teeth of the arch. The supracrestal fibers especially the transseptal fiber system are the main cause of postretention relapse of orthodontically treated teeth.

viii. *Stippling of gingiva*: Gingiva is characterized by the stippled surface. It refers to the surface texture of the gingiva that is like an orange peel. It is a normal physiologic appearance. Its absence or reduction shows diseased gingiva. It is produced by an elongated papillary layer of the connective tissue that gives round protuberances to the gingiva. Stippling is produced by the rounded protuberances and depressions of the gingival surface. These depressions are the center of heavier epithelial ridges (Figs 9.33 and 9.34).

It is important to note that the central portion of the gingiva or the attached gingiva is stippled and the marginal gingiva is not stippled. Stippling is not very prominent on the lingual side as compared to the labial, It is less or absent in infancy and old age as compared to adulthood, when it is prominent. In younger females, the connective tissue is more finely textured than in males. Males tend to have a more heavily stippled gingiva.

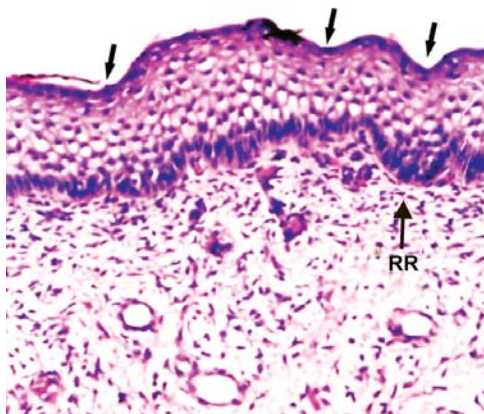


Figure 9.33: Gingiva is lined by stratified squamous epithelium showing undulations at the surface called stippling. The rete ridges (RR) show elongation under the epithelial depression. (shown by arrows). (H & E x 100)

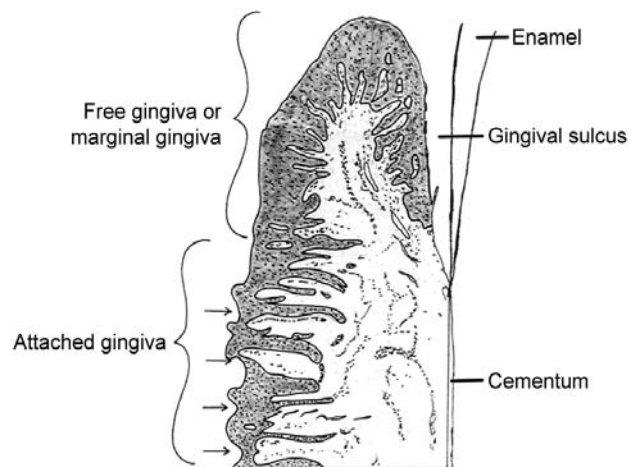


Figure 9.34: Section of gingiva showing alternate elevations and depressions (arrows) in the attached gingiva is responsible for stippled appearance

Vermilion Border of Lip

Vermilion border of lip is the transitional region or area between the skin of lip and the mucous membrane of the oral cavity (Fig. 9.35). This area is pink or red in color due to the thin epithelium and high vascularity. Because the papillae of lamina propria are long, the blood vessels are very near to the epithelial surface, giving it a red color.

This is a zone where keratinization ends, as skin of the lip is keratinized and the mucous membrane of lip is non-keratinized. It is found only in humans. The skin contains appendages such as the hair follicles, sweat glands and sebaceous glands. In the oral mucosa, there is a lack of appendages. The mucocutaneous junction of the lip is synonymous with the vermilion border.



Figure 9.35: Section through upper lip: 18 week old fetus: (S) Skin of lip, (TZ) Transition zone or vermilion border, (MM) Mucous membrane of lip, (M) Muscle, (BV) Blood vessel (x 30)

As this transitional zone contains only occasional sebaceous glands and lacks salivary glands, it tends to become dry, often becoming cracked and sore in cold weather. Between the vermilion border and the thicker non-keratinized labial mucosa, is an intermediate zone covered by parakeratinized oral epithelium (Fig. 9.36). In infants, this region is thickened as an adaptation to suckling and is called the suckling pad.

Nonkeratinized Areas

Lining Mucosa

The epithelia of the lips, soft palate, cheeks, floor of mouth are covered by a non-keratinized or lining epithelium. As the lining mucosa is loosely textured it allows easy movement of the lip, cheek and tongue.

Mucosa of soft palate: The mucous membrane of the soft palate has thin non-keratinized stratified squamous epithelium with taste buds present. The lamina propria is thick and highly vascularized, being red in color. It shows few, short papillae (Fig. 9.37). The elastic fibers are rich in this layer. The submucosa is loose and contains a continuous layer of mucous glands, taste buds and

numerous minor salivary glands. The oral mucosa continues around the free border of the soft palate for some distance and is then replaced with pseudostratified, ciliated columnar epithelium of the nasal mucosa. The submucosa of soft palate shows numerous minor salivary glands (Figs 9.38, 9.39 and 9.41).

Ventral surface of tongue: The epithelium of the ventral surface of tongue is thin, non-keratinized, and loosely textured. The lamina propria is thick having short papillae with a rich capillary network. Due to the loose texture of the mucosa, it is loosely attached to the underlying structures so the tongue is freely mobile. The submucosa cannot be identified as a separate layer (Fig. 9.40). It is thin and irregular and is hardly distinguishable from the

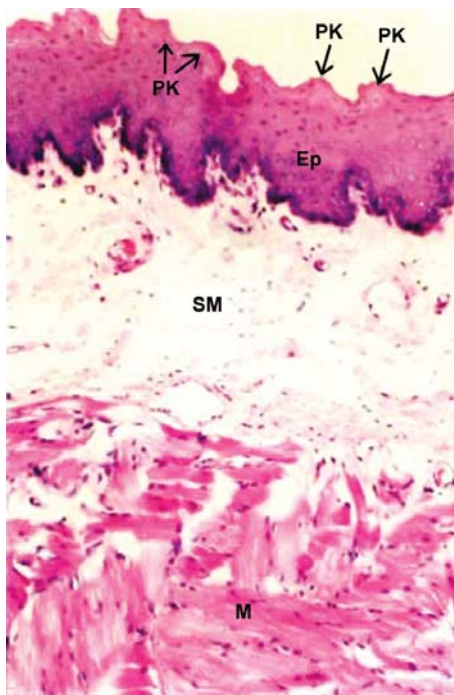


Figure 9.36: Parakeratotic Epithelium: Epithelium (Ep) lining the lip shows thin layer of parakeratin (PK) covering the surface (x 150) (SM) Submucosa, (M) Muscle

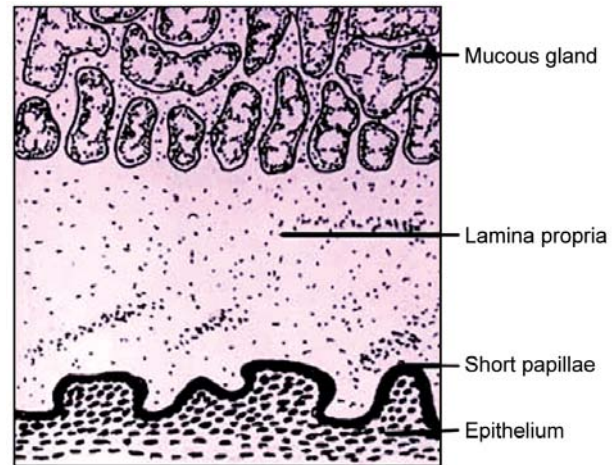


Figure 9.37: Mucous membrane of soft palate (as seen under light microscope)

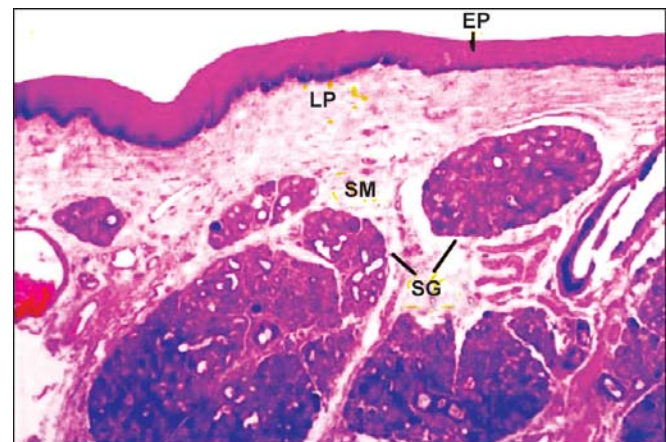


Figure 9.38: Soft palate is lined by nonkeratinizing stratified squamous epithelium (Ep). Submucosa (SM) contains continuous band of minor salivary glands (SG). (LP) Lamina propria (H & E x 50)

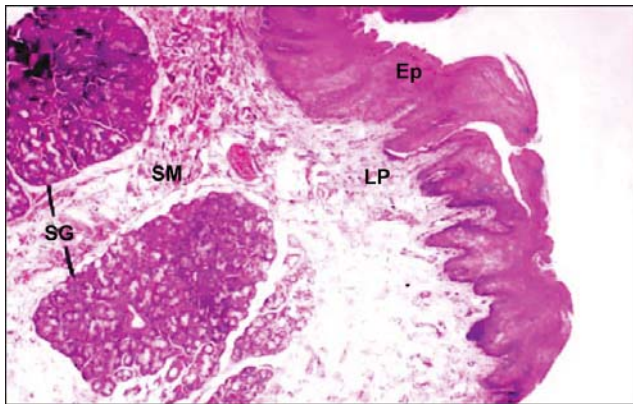


Figure 9.39: Non keratinized oral mucosa present in soft palate and floor of the mouth. (Ep) Epithelium, (LP) Lamina propria, (SM) Submucosa, (SG) Minor salivary glands

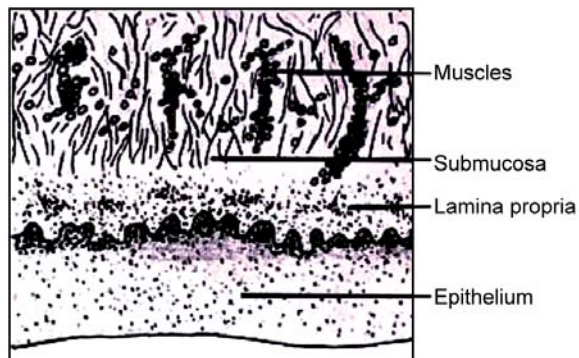


Figure 9.40: Section of mucous membrane on inferior surface of tongue

connective tissue layers of the lamina propria. The submucosa binds the mucous membrane tightly to the connective tissue of the muscle.

Floor of the oral cavity: The histological features are similar to those found on the ventral surface of the tongue. The epithelium of the mucosa of the floor of the oral cavity is non-keratinized, thin and loosely adherent to the underlying structures, which allows free mobility to the tongue. Papillae of the lamina propria are numerous but short. The submucosa contains adipose tissue but cannot be identified. The junction between the sublingual mucosa and the lingual gingiva corresponds to the mucogingival junction on the vestibular surface. The sublingual mucosa is reflected onto the lower surface of the tongue and continues as the ventrolingual mucosa. Floor of the mouth shows mixed minor salivary glands (Fig. 9.41).

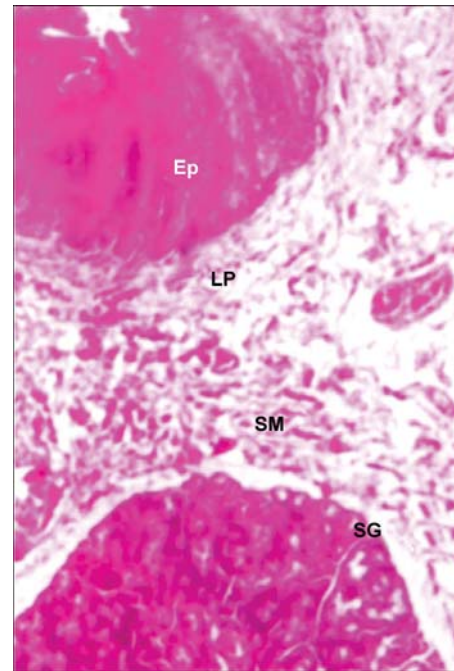


Figure 9.41: Floor of oral cavity shows presence of mixed minor salivary glands (SG) in the submucosa (SM) which is loose and consists of short collagen fibers. Ep=Epithelium, LP= Lamina propria. (H & E x 200)

Lips and Cheeks

- i. **Lips:** The epithelium of the lip is characterized by a broad transitional zone between the oral mucosa and hairy skin on the external surface of the lips. This transitional zone (as earlier described) is called the vermilion border or the mucocutaneous junction. The epithelium over the vermilion border is thin orthokeratinized and is semitransparent. The lamina propria of the labial and buccal mucosa consists of dense connective tissue and has short, and irregular papillae. The lamina propria is connected to the thin fascia of muscles by a submucosal layer. The submucosal layer contains strands of densely grouped collagen fibers. Loose connective tissue consisting of fat and small mixed glands is interspersed between these strands. The mobility of the mucous membrane is limited by strands of connective tissue, which hold it to the musculature. This prevents trauma to the mucous membrane of lips and cheeks by not letting it come in between the biting surfaces of the teeth during mastication. The submucosa of the lips also contains mixed minor salivary glands.

- ii. *Cheeks*: Buccinator muscle forms the major portion of cheek. The submucosa tightly adheres non-keratinized epithelium to the muscle (Figs 9.42 and 9.43). The serous or mucous salivary glands are situated between bundles of buccinator muscle, and sometimes on its outer surface.
- iii. *Fordyce spots*: These are ectopic (isolated) sebaceous glands found within the oral mucosa of cheek opposite the maxillary molars and lateral to the corner of mouth. These glands are pale yellowish in appearance, found in 75 percent of adults and are a normal anatomic feature (Fig. 9.44).

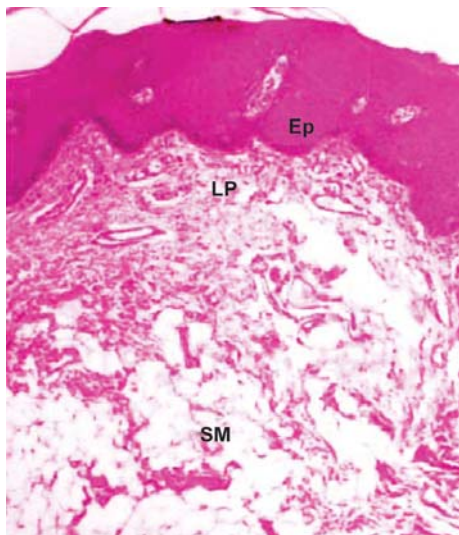


Figure 9.42: The buccal or cheek mucosa shows very thick layer of submucosa (SM) which contains loose strands of fibrous tissue, fat cells and muscle fibers. Epithelium (Ep) is stratified, squamous nonkeratinizing or parakeratotic in nature. LP= Lamina propria, (H & E x 100)

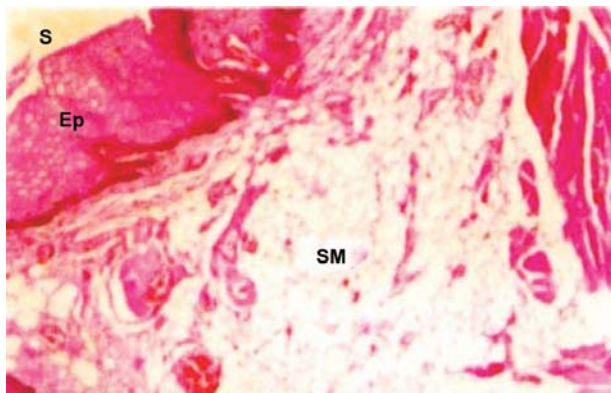


Figure 9.43: Non-keratinized cheek oral mucosa. S = Surface Ep = Epithelium SM = Submucosa

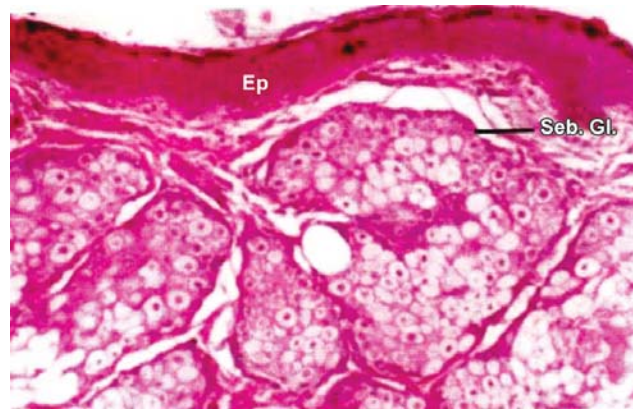


Figure 9.44: Fordyce's Granules: Sometimes buccal and upper lip mucosa shows presence of sebaceous gland (Seb GI) in the submucosa, These are called Fordyce's spots. Ep= Epithelium. (H & E x 100)

Vestibular fornix and alveolar mucosa: Mucogingival junction separates the gingiva and alveolar mucosa.

- *Vestibular fornix:* The mucosa of the vestibule is thin, non-keratinized and loosely textured allowing free movement of the cheek and lips.
- *Alveolar mucosa:* It is thin and non-keratinized stratified squamous epithelium. It is attached loosely to the underlying periosteum by submucosa that is made up of loose connective tissue containing thick elastic fibers and numerous mixed salivary gland. The epithelial ridges and papillae are low and often missing. The alveolar mucosa is dark red in color when compared to gingiva that is pale pink in color due to superficially running capillaries close to the surface.

Specialized Mucosa

Dorsal lingual mucosa: The dorsal surface of the tongue is covered by specialized mucosa.

This mucosa is specialized in the sense that it is non-keratinized or parakeratinized containing numerous papillae having taste buds showing specialized sensory function of taste. The anterior two-thirds is supplied by the trigeminal nerve through its lingual branch and the posterior one-third is supplied by the glossopharyngeal. Mucous membrane on the upper surface of the tongue is moist, pink, rough and irregular. Tongue is divided into two parts.

1. Anterior part or papillary part which faces superiorly and contains many fine, pointed, cone-shaped papillae which gives it a velvety appearance.
2. Posterior part that faces posteriorly, is called the lymphatic part.

Papillary part forms the anterior two-third of the tongue and the lymphatic part forms the posterior one third. Both the parts are separated by a V-shaped furrow termed the *sulcus terminalis*, the limbs of which run laterally and forward from a median pit named the *foramen caecum* [Foramen caecum is a remnant of the thyroglossal duct]. (Figs 9.45 and 9.46).

The two parts of the tongue differ embryologically and in the nature of their covering epithelium. The anterior papillary part and the posterior lymphatic parts are derived from different visceral arches. Both these parts are supplied from the different source of nerves. The anterior two-thirds are supplied by trigeminal nerve through its lingual branch and the posterior one-third by glossopharyngeal nerve.

The covering epithelium of the tongue is non-keratinized or para-keratinized. Stratified squamous epithelium is covered by four types of papillae, and some papillae contain taste buds.

Papillae of Tongue

- i. *Filiform papillae*: These are also called the conical papillae because they bear conical epithelial caps. These cover the entire anterior surface of the tongue. These are pointed, minute conical or cylindrical in shape having a core of connective tissue containing numerous connective tissue papillae (Figs 9.47 and 9.48). The conical or covering epithelium is keratinized. The filiform epithelium is not associated with taste perception and does not contain taste buds.
- ii. *Fungiform papillae*: These are found chiefly at the side and tip of tongue, scattered between numerous filiform papillae. These are round, elevated, red and mushroom shaped (fungus-like) papillae (Figs 9.49). The word fungiform is taken from fungus like. Their color is derived from a rich capillary network visible through thin epithelium. The surface is covered by thin nonkeratinized epithelium bearing only one to three taste buds, present on the dorsal surface.
- iii. *Foliate papillae*: Foliate papillae are found on the posterior side of the tongue, in front of the palatoglossal arches as four to ten vertical folds. These are raised, round to oval follicles with lymphoid aggregate in the center. These are lined by stratified squamous epithelium Taste buds are present in the epithelium of the lateral wall of folds. They are more frequently seen in mammals other than human (Fig. 9.50).
- iv. *Circumvallate papillae (vallate papillae)*: These are 8 to 12 in number, larger in size and situated in the dorsum of the tongue. In front of the sulcus terminalis, these are bound by deep circular furrows. The ducts of the minor salivary glands called the von Ebner's glands, open into these furrows. The lateral wall of the

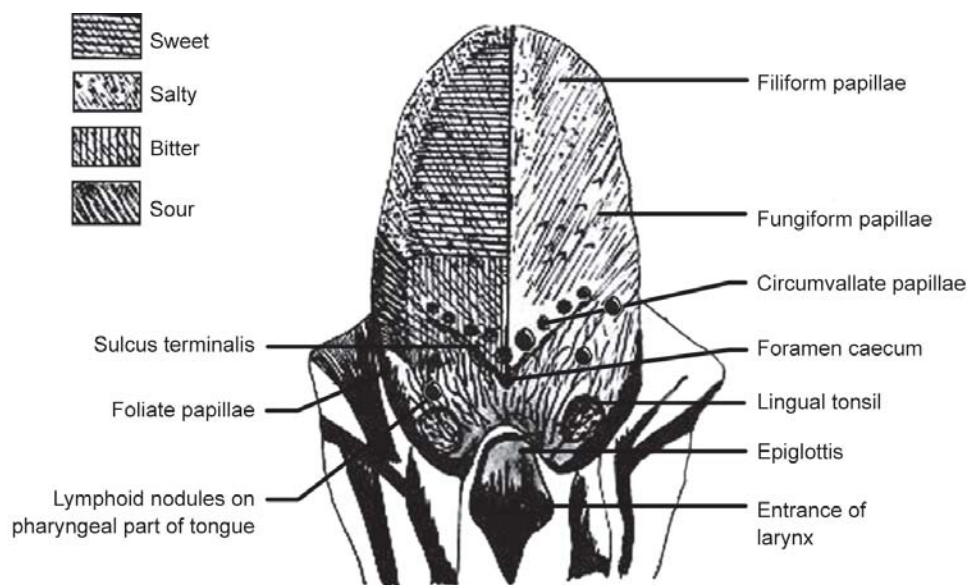


Figure 9.45: Various types of papillae, structures and regions of taste perception on dorsal surface of tongue

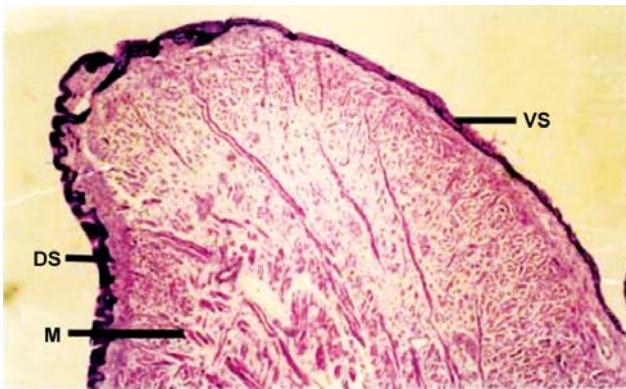


Figure 9.46: Embryonal development of tongue: Fetus at 14 weeks: Dorsal surface (DS) of tongue is lined by thick mucosal layer showing developing papillae. Ventral surface (VS) is lined by thin and smooth mucosa. The main body of tongue is made of immature mesenchyme and developing muscle bundles (x 30) (M) Muscle fibres

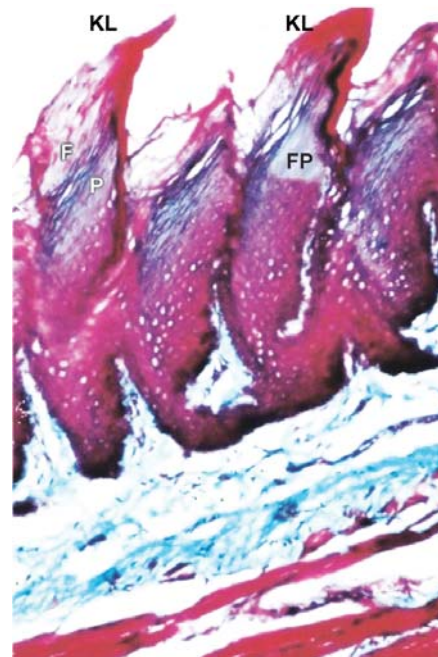


Figure 9.48: PAS stain showing deep magenta color taken up by the keratin lining (KL) on the tip of filiform papillae (FP)

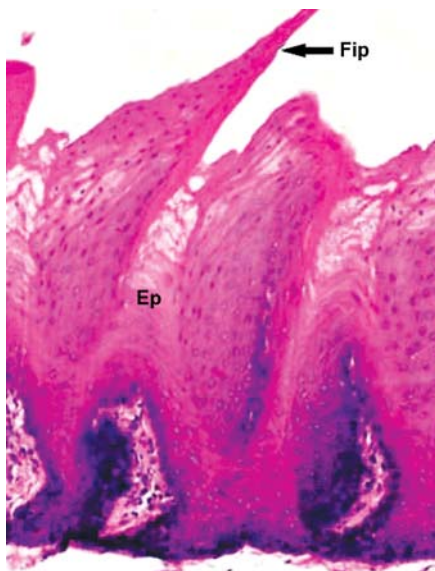


Figure 9.47: Filiform papillae of tongue: Adult tongue: Filiform papillae (Fip) are thin pointed highly keratinized structures arising from the surface epithelium (Ep) of dorsal surface of tongue (x 150)

circumvallate papillae is non-keratinized and it contains the most numerous taste buds of all the papillae (Figs 9.51 and 9.52). The serous secretion of the von Ebner's gland serve to wash out the soluble elements of food and these are the main source of the salivary lipase.

Taste Buds

These are also called as gustatory calculi (a small cup-shaped structure) or organ. These are made up of modified epithelial cells arranged in groups. These are found in

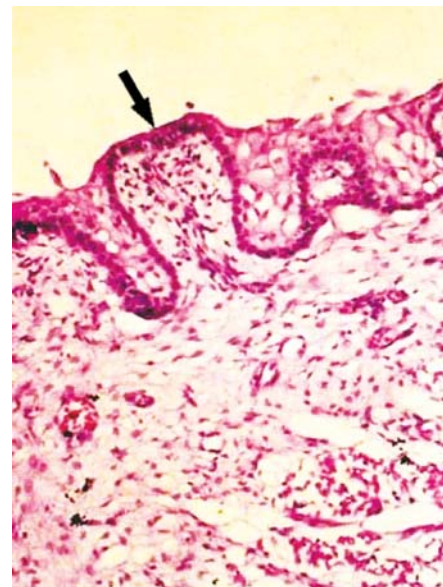


Figure 9.49: Fetal tongue showing developing fungiform papillae indicated by arrows (x 150)

the tongue, soft palate, epiglottis and the posterior wall of the oral part of the pharynx. But they are most numerous on the vallate papilla of the tongue. Comparatively taste buds are more in number in infants than in adults and in adults they are more in number than in old and more aged persons because with the increase in age they get

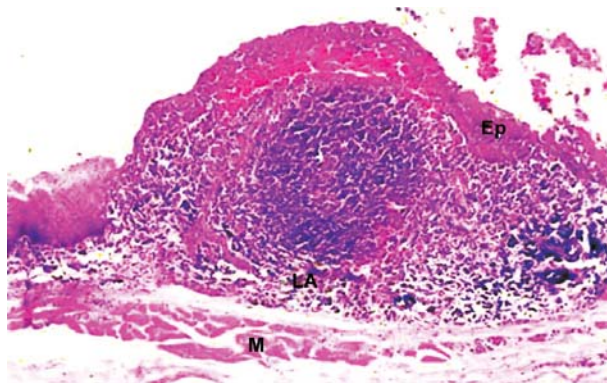


Figure 9.50: Lingual Follicle/ Foliate papilla. At the posterior part of the tongue and on its lateral border, raised round to oval follicles with lymphoid aggregates (LA) in the center are present. The epithelial lining (Ep) is stratified squamous in nature. M = muscle (H & E x 100)

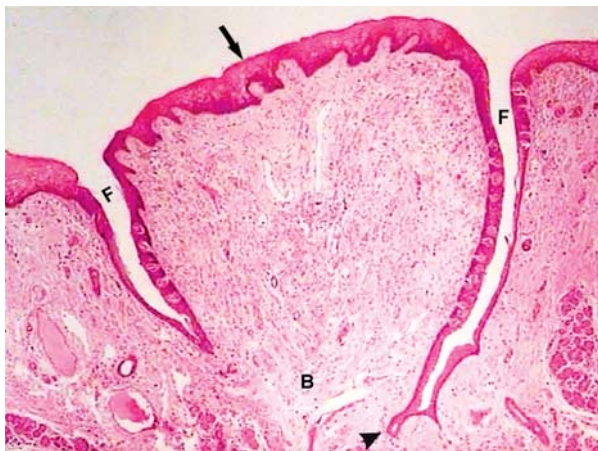


Figure 9.51: Circumvallate papilla of adult tongue. Papilla (indicated by arrow) is surrounded by a furrow (F) on both sides and does not project above the surface of tongue. Base (B) of papilla is narrow. Right side furrow shows opening of the duct of von Ebner's gland (shown by arrow head) x 20

atrophied. The mid dorsal region of oral part of the tongue contains no taste buds. The younger the age more are the taste buds.

Taste buds are ovoid structures or barrel-shaped structures (80 micron high and 40 micron thick) found in the epithelium, and are separated from the connective tissue by a basement membrane. The taste buds open on the surface of the epithelium by a taste pore or gustatory aperture. The outer surface of a taste bud is covered by flat epithelial cells, which cover the gustatory pore of a taste bud. The gustatory pore leads into a small canal that is lined by supporting cells of taste buds. The outer supporting cells are arranged like the staves of a barrel. The inner and shorter supporting cells are spindle-shaped (Figs 9.53 to 9.56).

The taste receptors or the neuroepithelial cells, the receptors of taste stimuli, are found in between the inner supporting cells. These receptor cells have finger-like projections. The base of each taste bud is penetrated by a group of gustatory nerve fibers. Substance to be tasted dissolves in saliva and enters the taste bud, tasted by neuroepithelial receptor cells and stimuli is taken by a rich plexus of nerves found below the taste bud. Some of the fibers enter the epithelium and end when it comes in contact with the sensory cells of taste buds.

Numerous taste buds are found on the inner wall of trough surrounding the vallate papillae, in folds of foliate papillae on the posterior surface of epiglottis. These are also found on some of the fungiform papillae at tip and the lateral border of tongue.

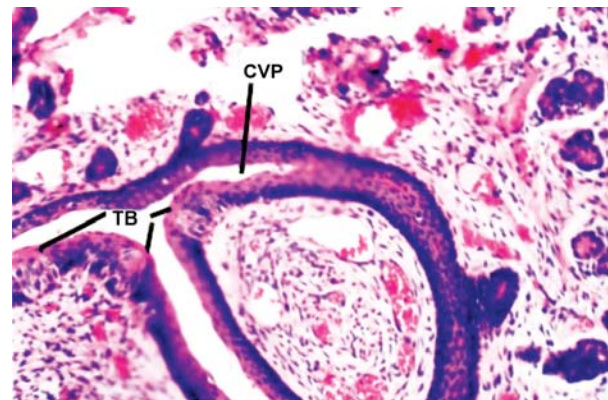


Figure 9.52: Circumvallate papillae: Fetus at 16 weeks: Circumvallate papilla (CVP) is seen lying deep in a cleft arising from surface of the tongue. Many taste buds (TB) are present in the lining of a circumvallate papilla. Tongue mesenchyme is immature. Many immature minor salivary gland units are also present (x 150)

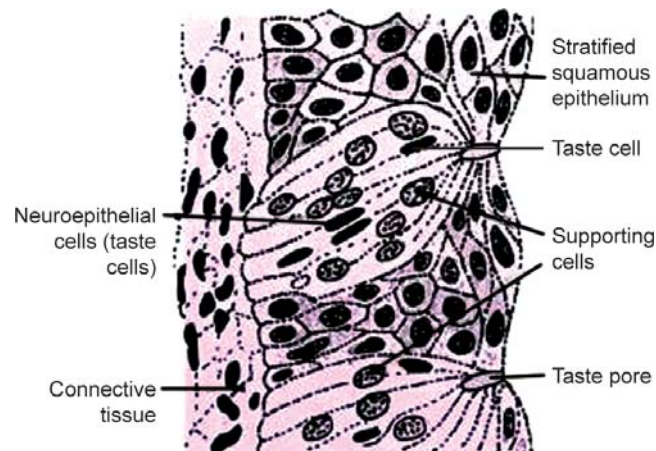


Figure 9.53: Structure of taste buds

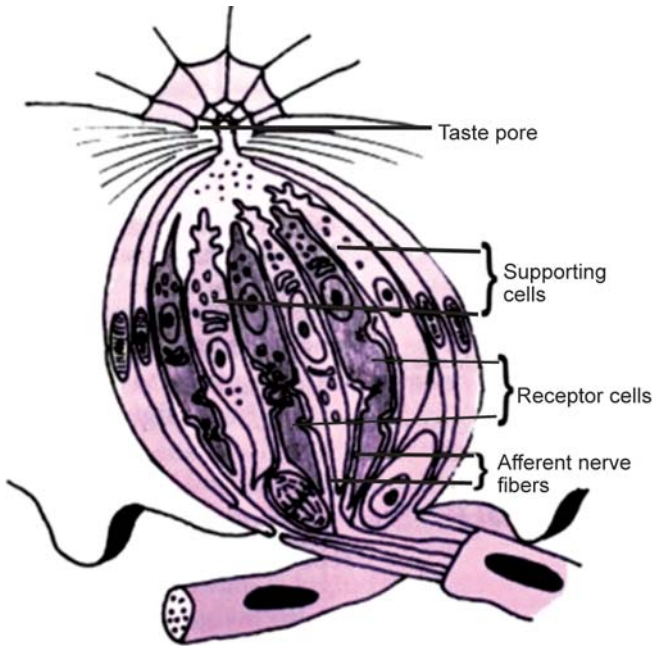


Figure 9.54: Taste bud from slope of dental papilla

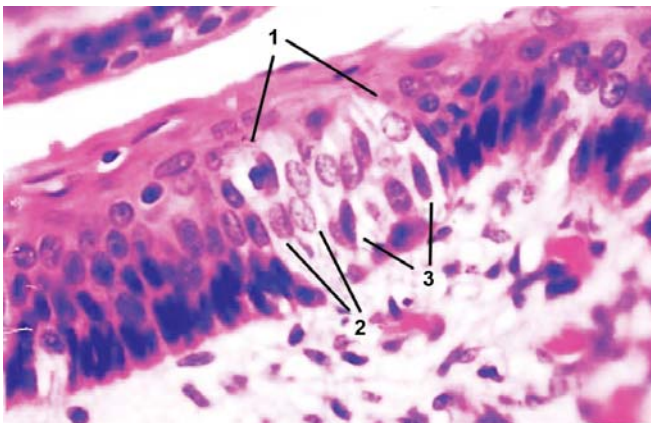


Figure 9.55: Taste Bud: High power view showing its structure. The cells with plump oval nuclei are the supporting cells (2). Spindle cells with thinner nuclei and finger like process of cytoplasm at superficial aspect are sensory cells (3). Two to three layers of epithelial cells cover the superficial aspect of taste bud (1). Taste pore is not visible in the photograph (x 500)

Regions of Distribution of Taste

Primary taste sensation are perceived in different regions of the tongue and the palate (Fig. 9.45).

Sweet: Tip of the Tongue

Salty: Lateral borders of the tongue. Both the sweet and salty tastes are mediated by the intermediofacial nerve by the chorda tympani.

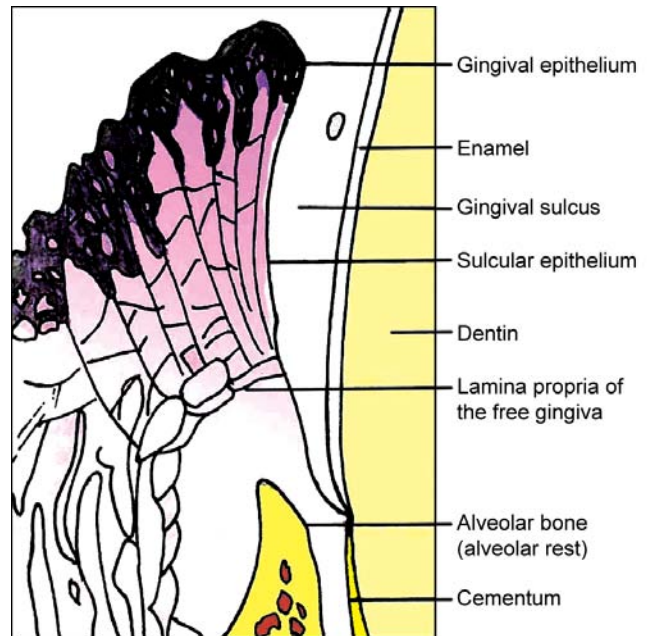


Figure 9.56: Periodontium near the cervical margin of the tooth showing gingival sulcus and epithelium

Bitter and Sour: Palate and posterior part of the tongue. Bitter in the middle and sour in the posteriolateral part of the tongue. Bitter and sour tastes are mediated through the glossopharyngeal nerve.

Lingual Tonsil

The lingual follicles are round or oval prominences containing a small pit at the center called the lingual crypt. The lingual crypt is lined with stratified squamous epithelium. The lingual follicles are present posterior to the terminal sulcus. Many lymphocytes migrate into the lingual crypts through epithelium. The posterior lingual mucous glands open via ducts into these crypts. The lingual follicles together form the lingual tonsil.

DENTOGINGIVAL JUNCTION, GINGIVAL SULCUS AND OTHER RELATED STRUCTURES

Dentogingival Junction

This is the junction of the gingiva and tooth and can be easily attacked by microorganisms.

Junctional epithelium: Epithelium of the gingiva which gets attached to the tooth.

Epithelial attachment: The union between the junctional epithelium and tooth. This dentogingival junction is formed

by junctional epithelium and gingival fibers and is together considered as a functional unit called the dentogingival unit. Clinically, the junction is very important. It is a point of reduced resistance to mechanical forces and bacterial attack.

The junctional epithelium has highest turnover rate of 5 to 6 days, highly permeable and extends up to 2 mm on the tooth surface. This epithelium is highly permeable, permits easy flow of gingival fluid and large intercellular spaces permit movement of neutrophils in and out of the epithelium.

The gingiva consists of two important tissues that help to maintain the junction intact. The lamina propria, which is dense and resilient, withstands forces during mastication. The gingival epithelium, either keratinized or parakeratinized, functions similarly.

When the epithelium is injured, the mitotic capacity and the migratory capabilities of the cells help repair this injury. In cases of injury to the connective tissue, ribosomes present within the fibroblasts form procollagen (the precursor protein of collagen) and ground substance, thereby helping in repair. Any condition that causes breakdown of collagen weakens the dentogingival junction. Hence, the firmness of the junction is maintained by the gingival portion of the periodontal ligament. The integrity of the dentogingival junction is therefore maintained both by the epithelium and the connective tissue, which are attached to the tooth.

Defense mechanism of the body helps in defense against bacterial injury. The lysosomes of the junctional epithelium may have a phagocytic function. The epithelium and connective tissue are attached to the tooth and these contribute to the integrity of the dentogingival junction. The gingival portion of the periodontal ligament helps to maintain the firmness of the junction. This junction is weakened by any condition in which collagen is broken down.

Gingival Sulcus

Gingival sulcus is a small pocket or crevice that extends from the free gingival margin to the dentogingival junction (Fig. 9.56) In health, its depth is at the approximate level of the free gingival groove on the outer surface of the gingiva.

The groove may be formed by the gingival sulcus, as the sulcus leaves the gingival margin without firm support.

It is believed that the groove is formed by the functional folding of the free gingival margin during mastication.

Periodontal Pocket

It is a pathologically deepened gingival sulcus.

Sulcular Epithelium

It is the epithelium lining the gingival sulcus, extending from the coronal limit of the junctional epithelium to the crest of the gingival margin. Sulcular epithelium is non-keratinized, thin, stratified squamous epithelium. It lacks epithelial ridges or rete pegs. The junction between the epithelium and the lamina propria is smooth and straight.

This epithelium though non-keratinized, if exposed to the oral cavity, may get keratinized. Sulcular epithelium is semipermeable and allows passage of toxins into the gingiva and tissue fluid from the gingiva into the sulcus. The sulcular epithelium is continuous with the gingival epithelium and the attachment epithelium.

DEVELOPMENT OF JUNCTIONAL AND PRIMARY ATTACHMENT EPITHELIUM

Junctional Epithelium

After completion of enamel matrix formation, the ameloblasts leave a thin membrane on the surface of the enamel, the primary enamel cuticle. Once the primary enamel cuticle has been formed, the epithelial enamel organ is reduced to a few layers of flat, cuboidal cells called the reduced enamel epithelium. It covers the entire enamel surface (Figs 9.57A and B). The origin of the junctional epithelium is from the reduced enamel epithelium of the enamel organ. It continues from the base of the gingival sulcus apically and surrounds the tooth as a collar-like band. It is stratified squamous and non-keratinized in nature. The length of the junctional epithelium ranges from 0.20 to 1.30 mm and is 4 to 5 cell layers thick in early life and increases to ten to twenty cell layers later in life.

As the tooth erupts, the reduced enamel epithelium becomes continuous or merges with the oral epithelium and together forms the attachment epithelium covering the enamel surface and extending up to the cemento-enamel junction and remains attached to the primary enamel cuticle.

Junctional epithelium is attached to the enamel surface or to the acellular cementum by basal lamina. Basal lamina

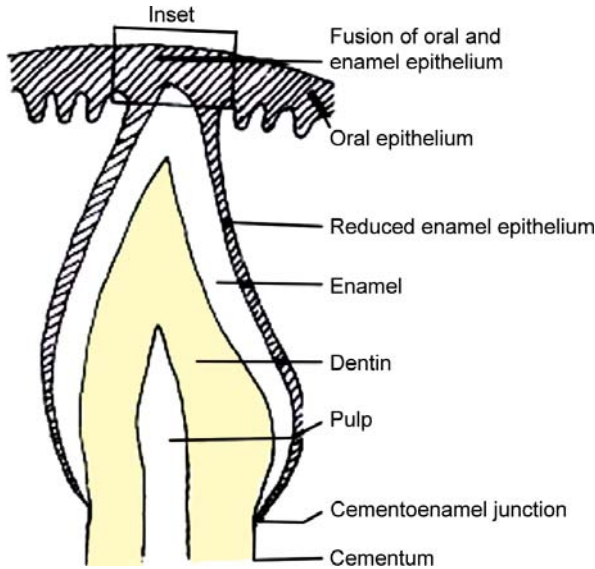


Figure 9.57A: Fusion of reduced enamel epithelium with oral epithelium in unerupted tooth

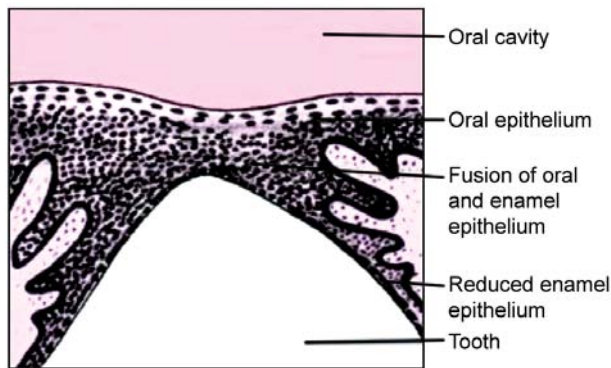


Figure 9.57B: Magnified view of the incisal one twentieth part of the clinical crown of inset in figure 9.57A

consists of the lamina densa adjacent to the enamel and lamina lucida which is attached to the junctional epithelium by the hemidesmosomes (Figs 9.11 and 9.58A and B). The turnover rate of the junctional epithelium is high. The cells continuously shed into the gingival sulcus and are replaced by the daughter cells from the division of the most apically located cells.

Junctional epithelium is divided into three zones (i) Coronal; (ii) Middle (iii) Apical (Fig. 9.59).

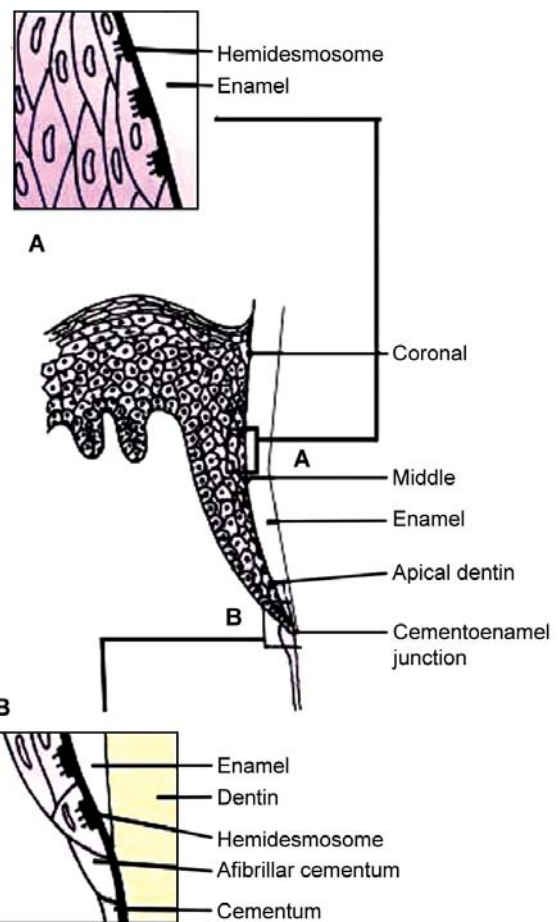
1. Coronal zone is highly semipermeable and allows entry of toxins and passage of gingival fluid into the sulcus.
2. Middle zone is an adhesive zone (Attachment epithelium).

3. Apical zone has a proliferative capacity to replace shedded cells of the junctional epithelium.

Primary Attachment Epithelium

Attachment of the junctional epithelium onto the tooth surface is called the **attachment epithelium**. During eruption, the tip of the tooth approaches the oral mucosa. As a result of this, the reduced enamel epithelium and the oral epithelium meet and fuse. The remnant of primary enamel cuticle is called the Nasmyth's membrane.

The epithelium covering the tip of the crown degenerates in its center, and the crown emerges through this perforation into the oral cavity. The reduced enamel epithelium remains organically attached to the part of the enamel, which has not yet erupted. Once the tip of the



Figures 9.58A and B: Dentogingival junction showing the junctional epithelium adhering to enamel of tooth with hemidesmosomes Inset, (A) junctional of epithelium with enamel, Inset (B), junction of enamel, cementum and epithelium

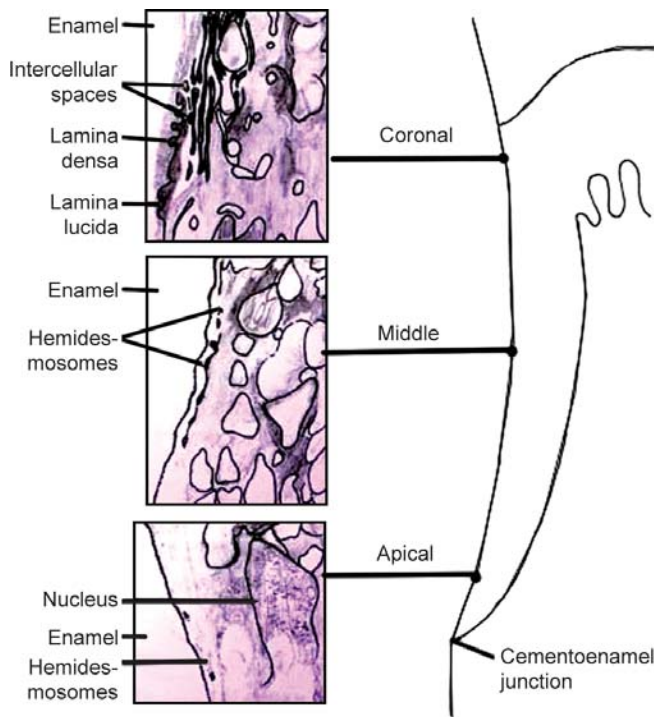


Figure 9.59: Junctional epithelium attached to enamel showing three zones - coronal, middle and apical. Coronal zone shows greater permeability characterized by numerous intercellular spaces, some of which open directly to the internal basement lamina. Middle zone shows greater number of hemidesmosomes, hence area of greater attachment. Apical zone shows cells with germinative characteristics and fewer hemidesmosomes (as seen under electron microscope)

When the crown has emerged, the reduced enamel epithelium is called the primary attachment epithelium (Fig. 9.58 A and B).

With the eruption of the tooth, the reduced enamel epithelium moves apically, reducing in length. Eventually a shallow groove develops in between the gingiva and the surface of tooth called the gingival sulcus. This gingival sulcus extends around the circumference of the tooth which is bordered by the attachment epithelium at its base, and laterally by the free gingival margin (Figs 9.60 A and B).

SHIFTING OF DENTOGINGIVAL JUNCTION AND APICAL SHIFT OF GINGIVAL SULCUS

Shifting of Dentogingival Junction

Dentogingival junction is the region where the tooth is attached to the gingiva and is formed as soon as the tooth erupts into the oral cavity. With time, the position of the gingiva on the tooth surface changes. Almost the entire

enamel is covered by the epithelium when the tip of the enamel first emerges through the mucous membrane of the oral cavity. The eruption of the tooth continues until it reaches the plane of occlusion. The firmness and mechanical strength of the dentogingival junction is because of the connective tissue attachments. This attachment of epithelium to the enamel is not weak. As the crown continues onto the oral cavity, the attachment epithelium separates from the enamel surface gradually. One-third to one-fourth of the enamel is still covered by the gingiva when the tooth first reaches the plane of occlusion (Fig. 9.61). A very slow exposure of the crown follows.

Preclinical or active eruption: The actual movement of teeth towards the occlusal plane is called active eruption.

Passive eruption: The separation of the primary attachment epithelium from the enamel surface is called passive eruption.

Secondary attachment epithelium: Further recession exposing the cementum occurs. When the reduced enamel epithelium has disappeared, the primary enamel

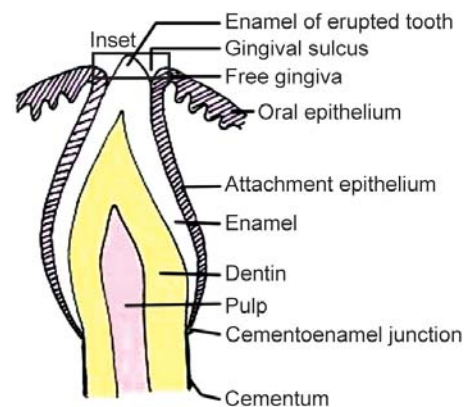


Figure 9.60A: Attached epithelial cuff and gingival sulcus at an early stage of tooth eruption

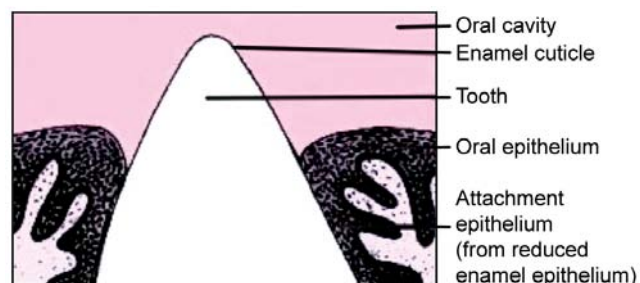


Figure 9.60B: Magnified view of the incisal one-fifteenth portion of the clinical crown of inset figure 9.60A

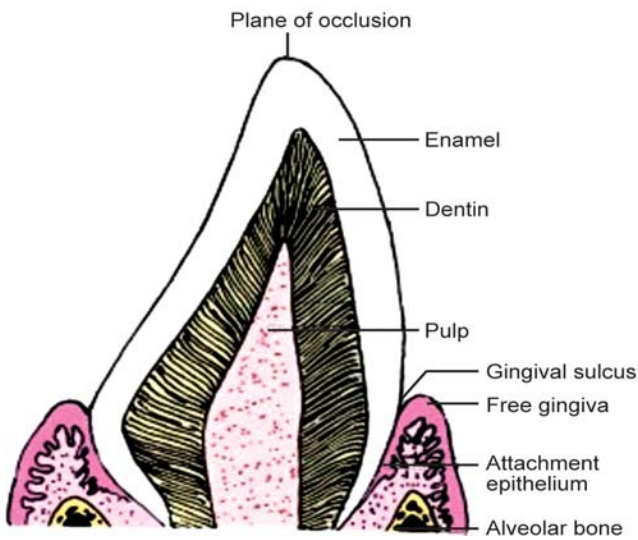


Figure 9.61: During eruption attachment epithelium covers one-fourth of enamel when tooth comes in occlusion

epithelium is replaced by a secondary attachment epithelium derived from the gingival epithelium.

Apical Shift of Gingival Sulcus (Passive Eruption)

Crown exposure which involve passive eruption and further recession has been described in four stages that may be physiologic or pathologic.

- i. *Stage I*: This is a physiologic stage and is usually present upto one year before shedding in primary and for up to twenty to thirty years of age in permanent dentition. In this stage, the bottom of the gingival sulcus remains on anatomical crown that is on the enamel portion and the apical end the attachment of epithelium (which is formed by the reduced enamel epithelium) lies at the cemento enamel junction (CEJ) (Fig. 9.62). But this relation is subject to a wide range of variation (Figs 9.63 A to C).
- ii. *Stage II*: This stage is also physiologic and is present usually up to the age of forty years. In this, the bottom of the gingival sulcus lies on the enamel but the apical end of attachment of epithelium has shifted from the cemento enamel junction to the cementum. The down growth of the attachment epithelium along the cementum is only one aspect of the dentogingival junction. This involves dissolution of fiber bundles that were anchored to the cervical parts of the cementum (which are now covered by the epithelium), and an apical shift in the gingival and

transseptal fibers. The destruction of the fibers is caused by enzymes, formed by the epithelial cells, by plaque, or by immunologic reactions which occur as manifestations of periodontal disease (Fig. 9.64).

- iii. *Stage III*: In this stage, anatomical (enamel covered) crown is fully exposed in oral cavity. The bottom of the gingival sulcus shifts to the cemento enamel junction. The epithelial attachment also entirely shifts on the cementum (Fig. 9.65). This is not a passive manifestation. The epithelium keeps shifting along tooth surface, and no longer remains at the cemento enamel junction. This is a slow and not a continuous process, which is an effort by the body to maintain the integrity of the dentogingival junction.
- iv. *Stage IV or gingival recession*: This stage represents gingival recession. Recession can be defined as an exposure of root surface by an apical shift in the position of gingiva, or recession refers to the position of gingiva and not its condition. It is a result of the pathology that may be inflammatory or non-inflammatory. In this stage, both the sulcus and epithelial attachment are on the cementum. The gingiva may appear normal but it is believed to have receded (Figs 9.66 and 9.67). This stage does not show any definite time of its arrival and may occur early in life depending upon the health of gingiva, but generally gingival recession increases with age. The rate varies also in different teeth of the same jaw and on different surfaces of the same tooth. One side may depict the first stage and the other side may be in the second or even third stage.

Anatomical crown: It is the portion of the tooth which is covered with enamel.

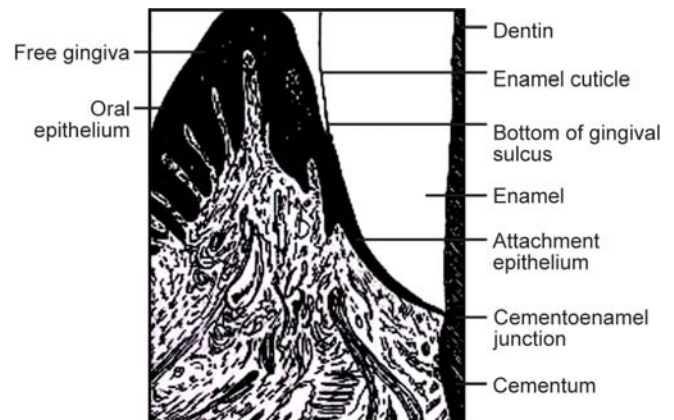
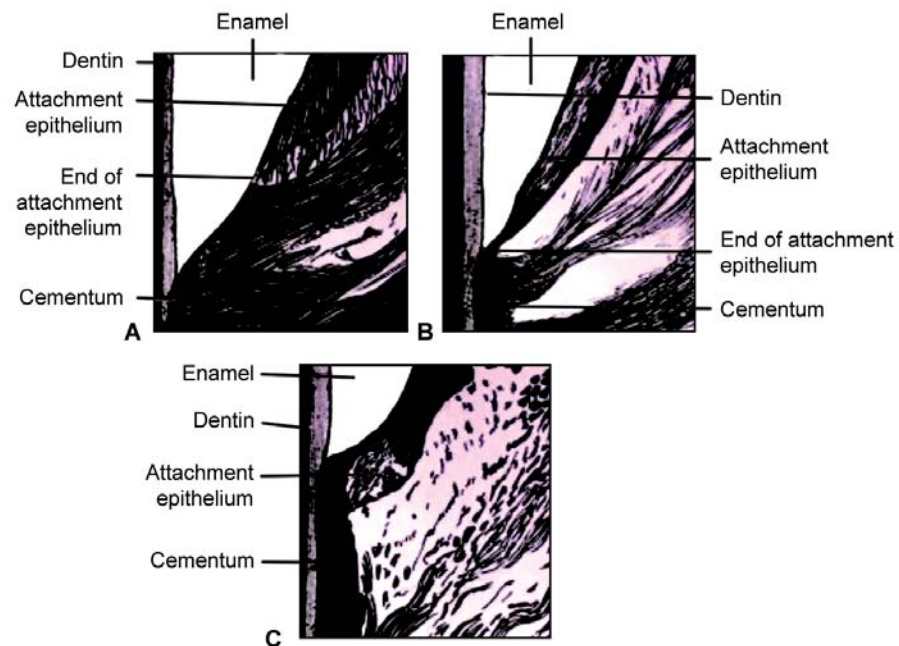


Figure 9.62: First stage of crown exposure. Attachment epithelium on the enamel



Figures 9.63 A to C: Three relations of epithelium attachment with cemento-enamel junction; A. Attachment of epithelium on enamel; B. Attachment of epithelium reaching to cemento-enamel junction; C. Attachment of epithelium is partly on enamel and partly on cementum. Cementum overlaps edge of enamel

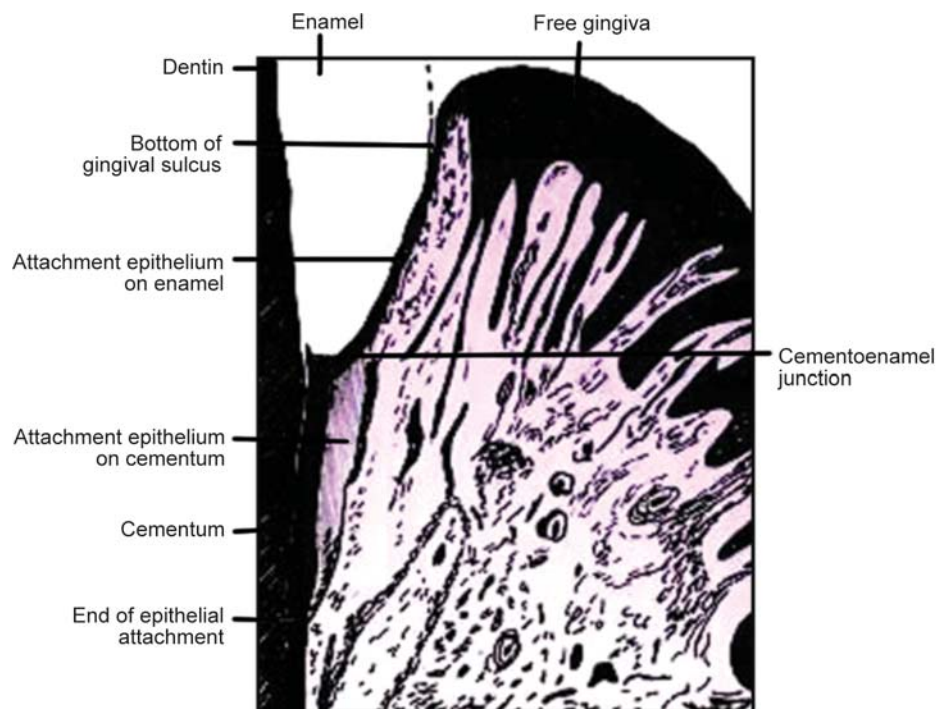


Figure 9.64: Second stage of passive tooth eruption. Bottom of gingival sulcus lies on enamel. The apical end of epithelial attachment has shifted to cementum. Attachment epithelium is partly on enamel and partly on cementum

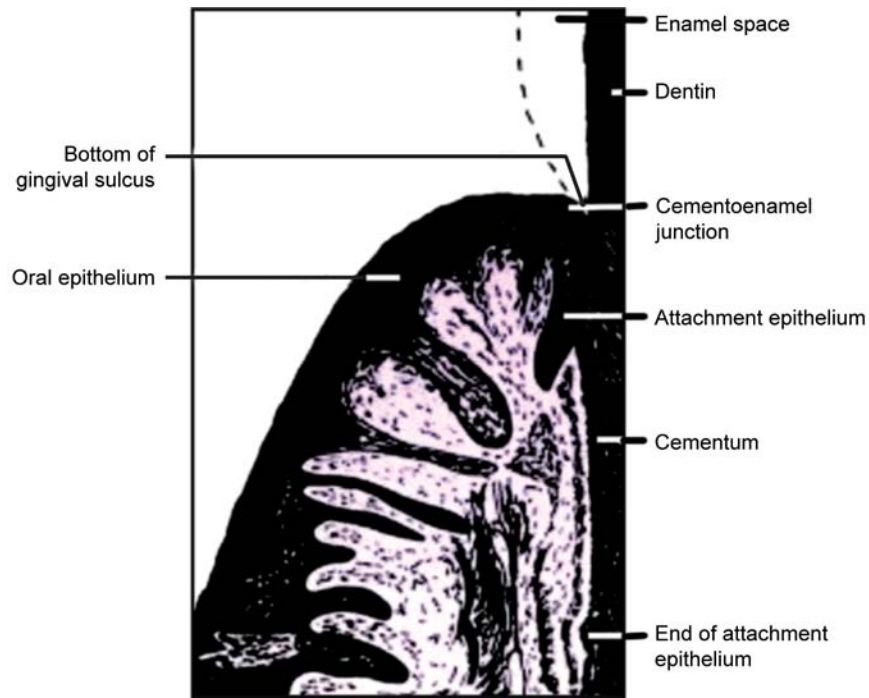


Figure 9.65: Third stage of tooth eruption. Recession is at bottom of gingival sulcus at cementoenamel junction. Attachment epithelium is on cementum

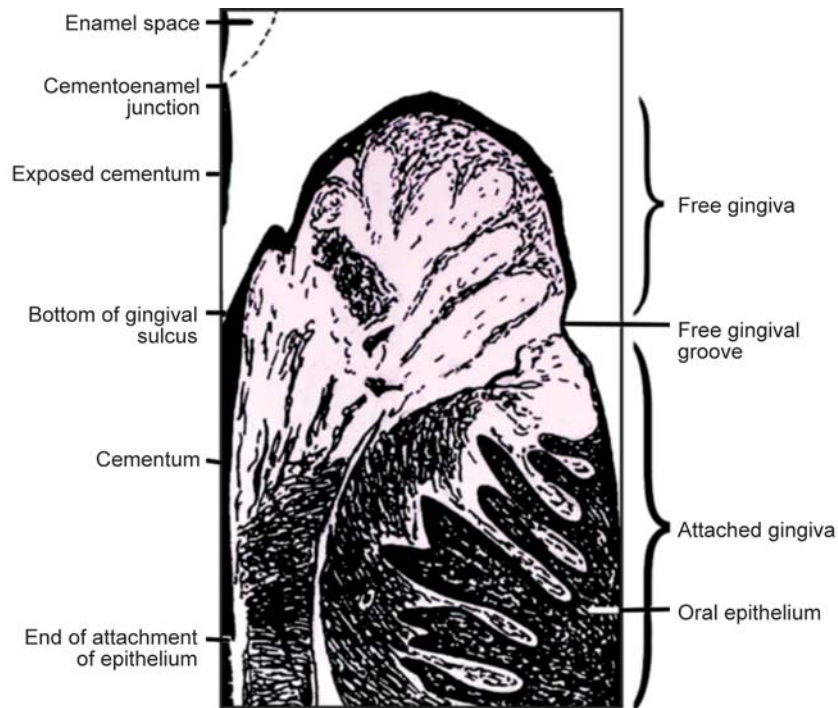
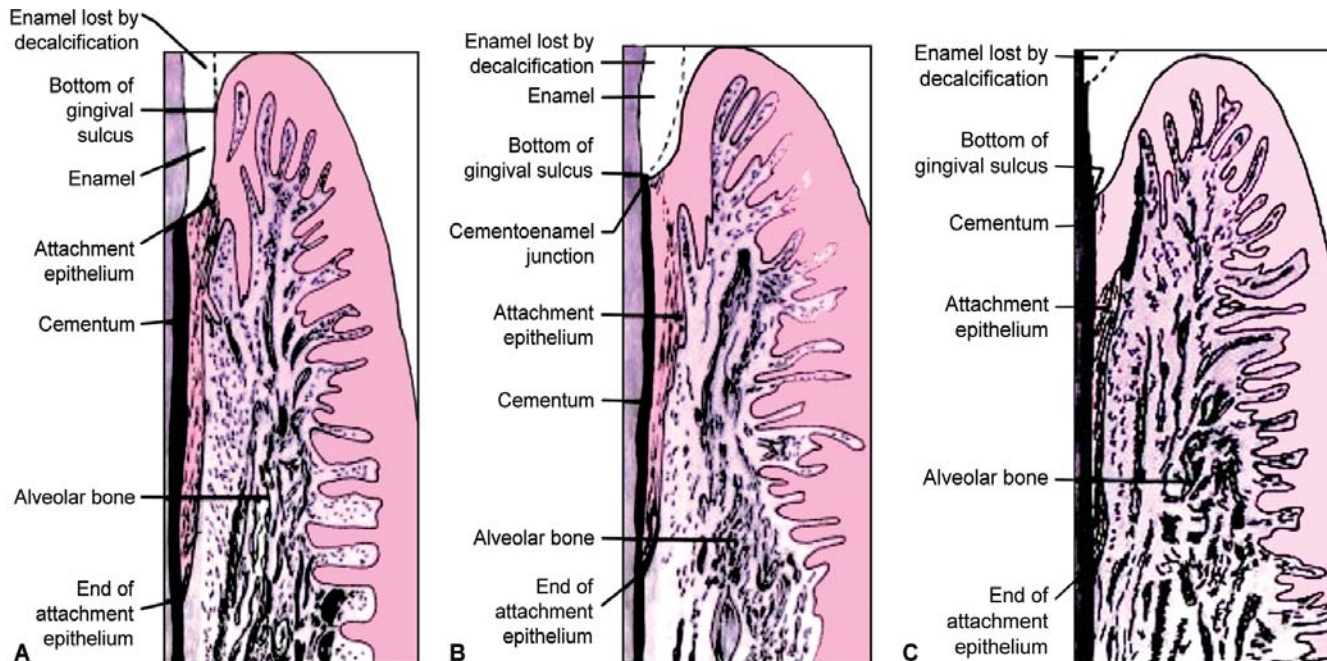


Figure 9.66: Fourth stage of tooth eruption. Recession due to aging or pathology at bottom of gingival sulcus and attachment epithelium both on cementum. Continued recession may reduce the width of attached gingiva



Figures 9.67A to C: Relationship of soft to hard tissues in the three sections of a tooth; A) Bottom of sulcus is present on enamel; B. Bottom of sulcus is present at cemento-enamel junction; C. Bottom of sulcus is present at cementum

Clinical crown: It is the portion of the tooth exposed in the oral cavity. Clinical crown may be smaller or equal or larger than the anatomic crown depending upon the position of the gingiva on the tooth surface (Fig. 9.68).

During the first two stages, the clinical crown is smaller than the anatomic crown. In the third stage (the beginning of recession), the entire enamel covered part of the tooth is exposed, and the clinical crown is equal to the anatomic crown.

During the fourth stage, the clinical crown is larger than the anatomic crown, as parts of root have been exposed.

Actual and Apparent Position of Gingiva

Actual position of the gingiva is level of epithelial attachment on the tooth. Apparent position is the level of crest of gingival margin. It is the actual position of gingiva, not its apparent position that determines the severity of recession.

Sulcus and Cuticles

An organic attachment called the epithelial attachment is present between the epithelium and the tooth. The

mechanism of the epithelial attachment was given by Orban and Gottlieb. It involves the primary cuticle forming an organic union between the ameloblasts and the enamel. When the oral epithelium replaces the ameloblasts, a secondary cuticle is formed.

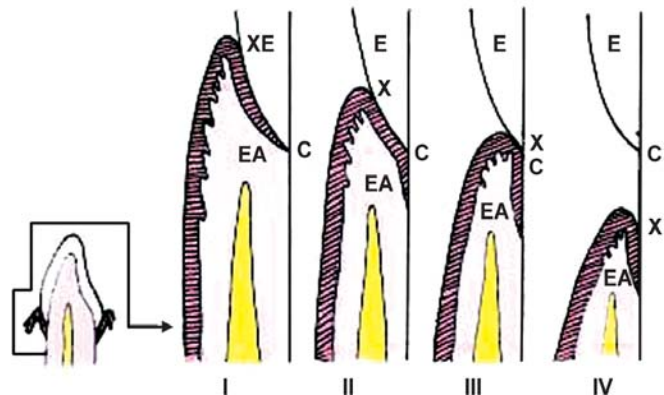


Figure 9.68: Four stages in eruption. (E) Enamel, (C) Cemento-enamel junction, (EA) Epithelium attachment, (X) Bottom of gingival sulcus. In stages [I] and [II]- (Passive eruption) anatomic crown is larger than clinical crown. In stage [III]-MILD Recession, Anatomic and clinical crowns are equal. In stage [IV]- Recession, Anatomic crown is smaller than the clinical crown. Box and arrow on the left diagram indicates the area which has been enlarged

When the epithelium proliferates beyond the cemento enamel junction, the cuticle extends along the cementum (Figs 9.69 and 9.70). The cemental cuticle and the secondary enamel cuticle are called as dental cuticle. These cuticles exist as amorphous materials between the attachment epithelium and tooth.

Deepening of Sulcus (Pocket Formation)

As the tip of crown emerges through the oral mucosa, the gingival sulcus forms. Separation of the reduced enamel epithelium from actively erupting tooth causes deepening of the sulcus. Initially, after the tip of the crown has appeared in the oral cavity, the epithelium separates rapidly from the tooth surface. Later, when it comes to occlude with its antagonist, the separation of the attachment from tooth surface slows down. In the beginning it was believed that from the time when tip of the crown tears the oral mucosa, the gingival sulcus extends to the cemento enamel junction (Fig. 9.71A)

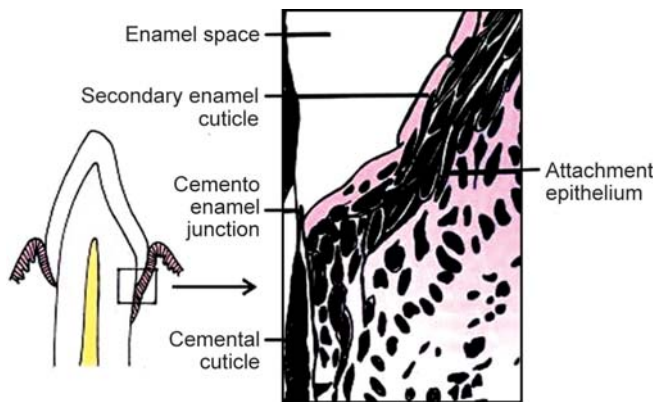


Figure 9.69: The epithelium proliferates beyond the cemento enamel junction "Secondary enamel cuticle" follows attachment epithelium to cementum forming the dental cuticle. Arrow and in left diagram indicates the area from which photomicrograph was taken

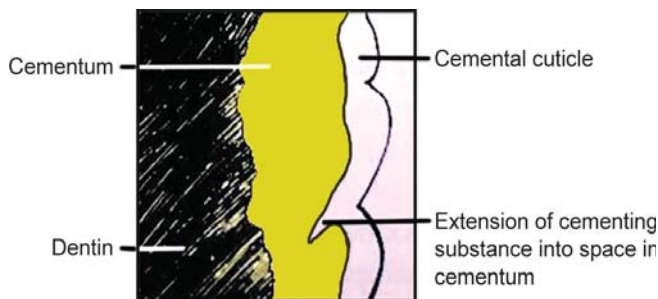


Figure 9.70: Extensions of cemental cuticle are seen extending into cementum

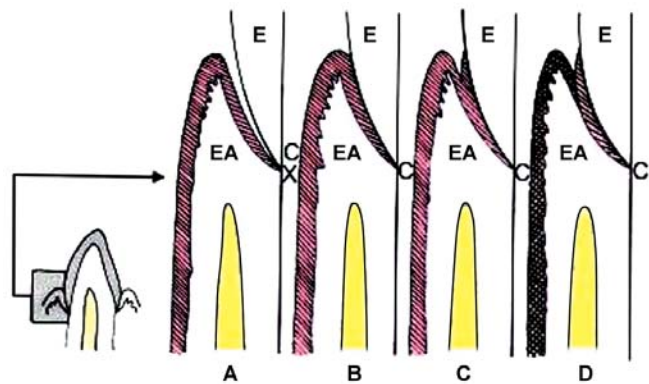
The concept of epithelial attachment was introduced by Orban and Gottlieb, who showed that no cleft was present between the epithelium and enamel, and that these tissues were organically connected. The gingival sulcus was shown to be a shallow groove, the bottom of which is at point of separation of the attached epithelium from the tooth (Fig. 9.71B)

Some believed the deepening of sulcus to be caused by a tear in the attached epithelium (Fig. 9.71, C). Others thought that it occurred because of the downgrowth of oral epithelium alongside the reduced enamel epithelium (primary attachment epithelium) (Fig. 9.71, D). Under normal conditions, the sulcus depth differs; 45 percent of all measured sulci are below 0.5 mm. The average sulcus is 1.8 mm. The possibility of gingival margin not being inflamed increases with the decrease in the depth of sulcus.

Lymphocytes, plasma cells and Langerhans' cells are usually seen in the sulcular and oral epithelium whenever infection or inflammation is present. These cells produce defense reactions to the bacteria in sulcus, which acts as barrier against the invasion of bacteria.

Attachment Epithelium

The primary attachment epithelium, which is, the attachment of the ameloblasts to the tooth, is a basal lamina to which hemidesmosomes are attached.



Figures 9.71A to D: Different views on formation of gingival sulcus. Arrow and box in left small diagram indicates area which has been enlarged. A. The gingival sulcus extending to the cemento enamel junction; B. The bottom of the gingival sulcus at the point of separation of attached epithelium from the tooth; C. Deepening of the gingival sulcus is seen caused by a tear in the attached epithelium; D. Deepening of the sulcus occurs as a result of the downgrowth of the oral epithelium alongside the reduced enamel epithelium; E. Enamel, (C) Cemento enamel junction, (EA) Epithelium attachment, (X) Bottom of gingival sulcus

The secondary attachment epithelium, composed of cells derived from the oral epithelium, forms an epithelial attachment similar to that of the primary attachment epithelium, that is a basal lamina and hemidesmosomes.

The epithelial attachment is submicroscopic, about 40 nanometers or 400 Å wide. It resembles an electron microscopic basal lamina. Cells of the attachment epithelium are held to this structure by hemidesmosomes.

Migration of the Attachment Epithelium

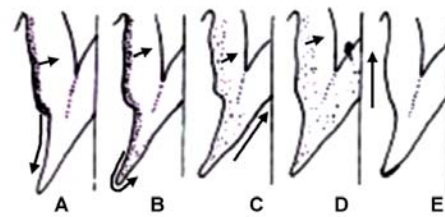
As the cells leave the stratum germinativum, they become specialized. As for example, cells in the oral epithelium specialize and undergo keratinization. In attachment epithelium, the cells specialize and synthesize a basal lamina (the epithelial attachment). They then migrate over it, with their attachment being maintained by the hemidesmosomes. A cell once specialized does not synthesize DNA and does not divide.

When experimental animals are administered titrated thymidine, the cells about to undergo DNA synthesis pick up radioactive thymidine. This radioactivity is detected in histologic sections by using photographic emulsion. After the administration of tritiated thymidine, labeled cells are found in the attachment epithelium.

The time taken for the labeled attachment epithelial cells to migrate and desquamate is called the transit time. It is about 72 to 120 hours for humans (Fig. 9.72).

The epithelial cells are affixed to the connective tissue through the basal lamina, yet they can detach from it and migrate towards the surface. At no time is the epithelium loose from the connective tissue. Both of them are in intimate contact. Imagine the epithelial attachment as the basal lamina of the attachment (junctional) epithelium. It turns about the most apical cell and extends up along the tooth surface. The hemidesmosomes hold the cells to this structure so that the strength of the attachment does not decrease with migration. This causes biologic mechanism, maintains the integrity of the attachment during the four stages of tooth exposure. The reduced ameloblasts do not divide but the basal cells migrate up and along the tooth, desquamating in 4 to 6 days.

While the reduced ameloblasts are still present, the cells of the oral epithelium join them by forming desmosomes. Slowly, the reduced enamel epithelium is lost, and the cells of the oral epithelium contact the tooth surface. These cells form hemidesmosomes and a lamina lucida, by means of which they attach themselves to the tooth. The apical migration of the sulcus is because of a



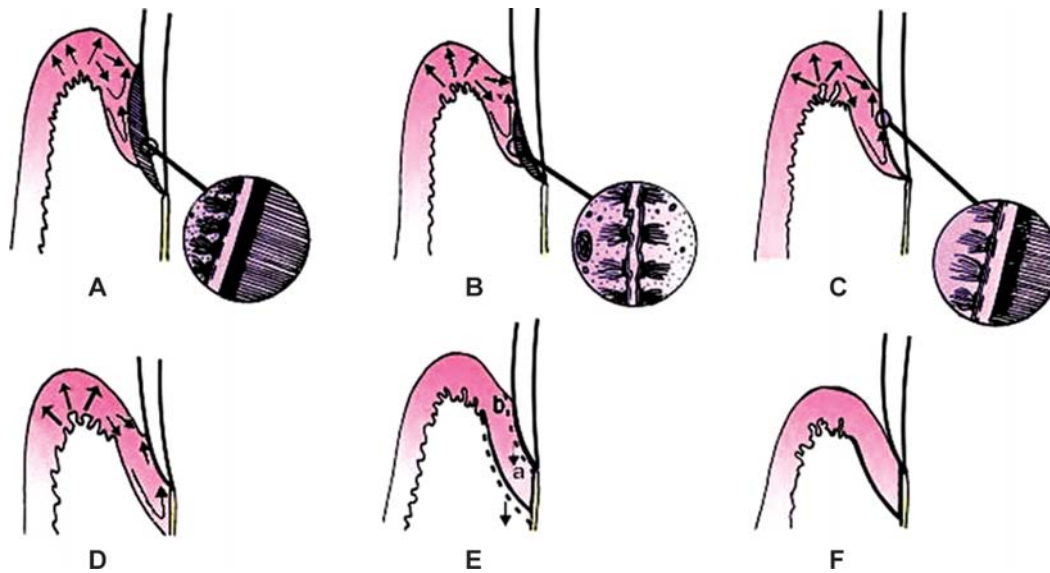
Figures 9.72A to E: Position of labeled cells; A. 1/2 hours, B. 6 hour, C. 24 hours, D. 72 hours, E. 144 hours after administration of titrated thymidine. Diagram of morphology of attachment epithelium and adjacent tissues is representative of gingiva on cemental (oral) surface of incisor (large arrows) show migration of attachment (junctional) epithelium toward and along tooth surface. Small arrows show migration of cells towards sulcus. Above is the enlargement of epithelial attachment (from Anderson GC and Stern IB: Periodontics 4:115, 1966)

detachment of basal cells and a re-establishment of their epithelial attachment at a more apical level (Fig. 9.73).

The intercellular spaces of the junctional epithelium are large. There are very few tight junctions. These spaces are easily penetrated by cells and proteins in transit.

There are certain possibilities which causes the sulcus to move apically. These are as follows.

- i. Toxic or inflammatory influences affect the basal cells. This diminishes the ability of the basal cells to synthesize DNA or may interfere with the physiology of these cells.
- ii. Collagenolysis occurs which destroys the collagen fibers, permitting the epithelium to migrate apically.
- iii. Immunologically competent cells or antibody complexes produce tissue damage, this results in apical migration of the epithelium.



Figures 9.73A to F: Pattern of growth and movements (dynamics of movements) of tissues of dentoepithelial junction; A. Dentoepithelial junction initially consists of reduced ameloblasts attached by hemidesmosomes to lamina lucida. Oral epithelial cells move to gingival surface and keratinize (arrows). Few cells join reduced enamel epithelium, to which they attach; B. Reduced ameloblasts are slowly displaced by junctional epithelium, cells of junctional epithelium are joined by desmosomes by tight and gap junctions. When reduced enamel epithelium produces junctional epithelium (X),- Mitotic activity is increased A locus of proliferation is formed by the cells of outer enamel epithelium, and possibly stratum intermedium; C. Reduced enamel epithelium is completely replaced by junctional epithelium. The attachment occurs by same mechanism as shown in [A, D] in time junctional epithelium may be found attaching to both enamel and cementum what causes this apical migration is not clear; E. In few days junctional epithelium renews itself as does gingival epithelium. Cells migrate in pathways denoted by arrows in [D] from basal lamina. Cells of junctional epithelium travel to epithelial attachment. During inflammation, basal cells at 'a' migrate apically and laterally into areas of collagenolysis. Basal lamina is formed by them. Arrow at 'B' represents deepening of sulcus; F. Even when junctional epithelium has completely migrated onto cementum, attachment is still mediated by basal lamina and by hemidesmosomes

Whatever the conditions, the junctional epithelium moves apically, replicates a new basal lamina, and reestablishes the epithelial attachment. If this results in a deepening of the sulcus a pocket will form.

COMPARISON OF SKIN AND MUCOSA

There are various differences between skin and oral mucosa (Table 9.5).

Table 9.5: Comparison of skin and oral mucosa		
	Skin	Oral mucosa
1. Color	Thicker structure, melanin pigmentation and less vascularity gives it a darker color.	Thinner structure and more vascularity gives it a brighter color.
2. Appendages	In the form of sweat gland, sebaceous gland, hair follicles.	These are absent in the oral mucosa except an occasional finding, sometimes minor salivary gland openings are present.

DEVELOPMENT OF ORAL MUCOSA

Development of the oral mucosa is initiated by a rupture of the buccopharyngeal membrane at about 26 days *in utero*. With this, the fusion of the embryonic stomatodeum and the foregut takes place and the entire stomatodeum or oral cavity is lined by epithelium, which is derived from both the ectoderm and endoderm.

The single layer lining the oral cavity changes to a double layer by 5 to 6 weeks of intrauterine life. The epithelium of the dental lamina vestibule region divides rapidly and thickens by 8 to 9 weeks. At about 8 to 11 weeks, the palatal shelves rise and close. This establishes the future morphology of the adult oral cavity. By about 10 to 14 weeks, the thickened epithelium of dental lamina shows central cellular degeneration causing separation of cells of cheek and alveolar mucosa (Fig. 9.74). This forms a space called the oral vestibule.

Lingual mucosa forms at around seven weeks, with the development of circumvallate and foliate papillae, but filiform papillae develop at around the tenth week of intrauterine life.

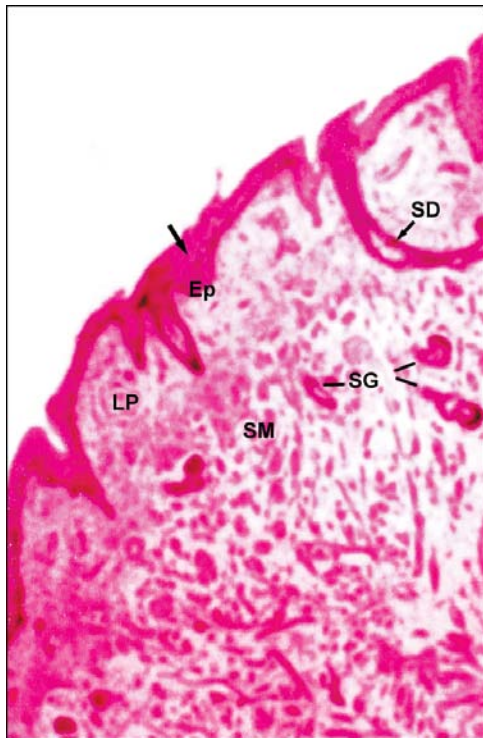


Figure 9.74: Embryonal development of cheek or buccal mucosa: 14-week-old fetus: The submucosa shows presence of immature salivary glands and muscle cells. Salivary ducts (SD) can be seen running toward epithelial lining. (Ep) Epithelium, (LP) Lamina propria, (SM) Submucosa, (SG) Salivary glands (H & E x 100)

Between 10 to 12 weeks, cells of the masticatory and lining mucosa show division and stratification and thicken. The areas which ultimately have to undergo keratinization (the hard palate and the alveolar ridge of the gingiva) contain darkly staining columnar cells. A basement membrane separates these cells from the underlying connective tissue.

The epithelium that eventually forms the areas of lining mucosa retains the cuboidal basal cells. The interface between the epithelium and connective tissue remains flat.

Between 13 to 20 weeks *in utero*, the epithelium is fully developed and shows distinct differentiation between each layer. This change occurs with the development of keratohyaline granules along with the appearance of Merkel cells and Langerhans cells. Till the eruption of teeth, the oral epithelium shows parakeratinization, and it only becomes orthokeratinized after birth with the eruption of teeth.

Along with changes occurring in the oral epithelium, the underlying ectomesenchyme shows gradual changes. In the beginning, widely spaced stellate cells in an amorphous matrix are present in the ectomesenchyme, but reticular fibers start accumulating by 6 to 8 weeks *in utero*. Fewer cells are present in the connective tissue of lining mucosa than in the future masticatory mucosa. Capillary buds and collagen fibers appear by 8 to 12 weeks *in utero*. In the beginning, the collagen fibers show no orientation, but gradually as their number increases, collagen bundles are formed. Subjacent to the epithelium, these bundles are arranged perpendicularly to the basement membrane, where they spread out. Elastic fibers become obvious only in the connective tissue between 17 to 20 weeks.

AGE CHANGES IN ORAL MUCOSA

The following age changes appear in oral mucosa in old age

1. Oral mucosa becomes smooth and dry.
2. Epithelium becomes thin due to decrease in (a) thickness of epithelial ridges and (b) salivary secretion.
3. Reduction in size and number of filiform papillae.
4. Nutritional deficiencies enhance the above changes.
5. On ventral surface of the tongue varicose veins appear which are called as lingual varices
6. In elderly persons ectopic sebaceous glands (Fordyce's spots) also appear.

CLINICAL CONSIDERATIONS

The basic considerations of the oral mucosa are variations in tissue color, dryness, smoothness or firmness and bleeding tendency of the gingiva. Following clinical conditions are important.

1. Periodontal Pocket

Pocket is a pathologically deepened gingival sulcus that forms due to the apical migration of attachment of epithelium generally as a reaction against bacterial attack, toxins, plaque toxins and immunologic response to them.

Scaling and root planing are treatments maneuvers done to remove the layer of calculus and bacterial plaque from the tooth surface so as to reduce pocket depth. Treatment methods should be judged by their ability to reduce the depth of the pockets and prevent their recurrence.

2. Cemental or Root Caries

Cementum or root caries occurs as a result of gingival recession when cementum gets exposed to the oral environment. Cementum is a less hard structure and can easily be attacked by acids and becomes carious.

3. Cemental Abrasion

Hard and strong brushing or the use of very abrasive dentifrice may lead to abrasion of cementum, exposing the dentin. This makes tooth highly sensitive to thermal or chemical stimuli. If pulp is exposed tooth will be very painful.

4. Restoration

It is necessary that whenever a restoration is being planned, it should be well adapted to the cavity by mechanical means. If recession has exposed the root and a restoration needs to be placed, the cavity preparation need not extend up to the gingiva at the cost of healthy dentin. If the gingiva is still on the enamel and the entire interdental space is filled with gingival papilla, then the gingival margin of a cavity should be placed at the level of sulcus. Where periodontal disease is present, treatment should always precede the placement of a restoration.

5. Edema

The submucosa of oral mucosa is made up of very loose connective tissue. Thus, it can show a great amount of swelling very easily. Due to its loose nature, there is no restriction to the spread of infection that can spread very fast along various planes.

6. Keratinization of Gingiva

Regular massaging the gingiva or brushing it with a soft brush helps in keratinization of the gingiva, which gives natural protection to the gingival surface against invasion by microorganisms and harmful substances.

7. Over Hanging Fillings or Carious Broken Teeth

Food accumulation under overhanging fillings or sharp edges of a carious broken tooth may cause chronic irritation of the gingiva, causing its inflammation and recession.

8. Discoloration of Gingiva

Metal poisoning such as silver, copper, lead and bismuth causes discoloration of the gingiva.

- A. *Bismuth pigmentation*: Bluish-black discoloration of the gingival margin.
- B. *Lead intoxication*: Burtonian or Burton's line is a steel gray pigmentation of the gingival margin or blue line along the margin of the gingiva visible in chronic lead poisoning.
- C. *Mercury intoxication*: Dark grayish or black gingival pigmentation is due to deposition of mercury sulphide. It is linear in form.

9. Oral Manifestations of Systemic Diseases

- A. *Measles*: The characteristic oral manifestation of measles is Koplik's spots. These are seen in the mucosa of the cheek particularly in the region opposite the molars as small red spots with bluish-white discoloration at center.
- B. *Scarlet fever*: It shows a characteristic glistening, red tongue showing atrophy of mucosa, called as Strawberry tongue.
- C. *Pernicious anemia and vitamin deficiencies (especially vitamin B complex deficiency)*: Lead to characteristic changes such as magenta tongue and beefy red tongue. Magenta tongue or beefy red tongue is due to deficiency of vitamin 'B' complex.
- D. *Leukaemia and other blood dyscrasias*: These can be indicated by characteristic infiltration of the oral mucosa.
- E. *Aphthous ulcers*: These are characterized by a well-circumscribed margin surrounded by an erythematous halo. These are painful and in fact may interfere with eating for several days.
- F. *Candidiasis*: The oral lesions are characterized by the appearance of painless soft, white slightly raised plaques, mostly seen on the buccal mucosa and the tongue. But these can also be found on the palate, gingiva, and the floor of the mouth. The plaques are described grossly as resembling 'milk curds'.
- G. *Syphilis*: In the primary stage, the oral chancre is seen as an elevated, ulcerated nodule showing local induration. It is mostly found in the oral cavity on the lips, tongue, palate and gingiva. It may become painful because of secondary infection.

In the secondary stage, the oral lesions are called 'mucous patches'. They are usually multiple, grayish white plaques overlying an ulcerated surface.

The tertiary stage is characterized by the 'intraoral gumma' which involves the tongue and the palate. It is a firm nodular mass which may ultimately ulcerates and causes perforation of the palate.

- H. *Chickenpox (Varicella)*: In this there are small, blister-like lesions which may sometimes involve the oral mucosa mainly, the buccal mucosa, tongue and mucosa of the pharynx with elevation of body temperature.
- I. *Diphtheria*: Characteristically, there is formation of a patchy 'diphtheritic' membrane that often begins on the tonsillar mucosa, and enlarges, becoming confluent over the surface.
- J. *Vesiculo-bullous conditions*: Pemphigus vulgaris and bullous pemphigoid can involve oral mucosa sometimes exclusively causing blisters and painful ulcerations.

10. Blanching of Oral Mucosa

In xerostomic individuals, the mucosa becomes atrophic, white, shiny and friable showing high rate of infection.

In the oral cavity, nerve endings are maximum in the lips and the anterior part of the oral mucosa. They are minimum in the posterior part of the oral cavity. Hence, the anterior part tastes food more than the posterior part. Only nerve endings for the perception of pain and cold are present in the posterior palate.

Injections should be made into the loose submucous connective tissue (that is the fornix and alveolar mucosa). The only place where large amounts of fluid can be injected in the palate without damaging the tissues is the loose connective tissue in the furrow between the palatal and the alveolar processes.

In denture construction, it is necessary that in the denture bearing areas, the mucosa should be firm.

In old age, the mucosa may atrophy. It becomes thin and parchment like. The atrophy of the lingual papillae leaves the upper surface of the tongue smooth, shiny and varnished in appearance.

Sometimes, the sebaceous glands are present as small yellow spots at various sites of oral cavity specially in cheeks. They are visible in the form of large, yellowish patches called Fordyce's spots or granules. (Fig. 9.75). These do not represent pathologic change but is a developmental anomaly.

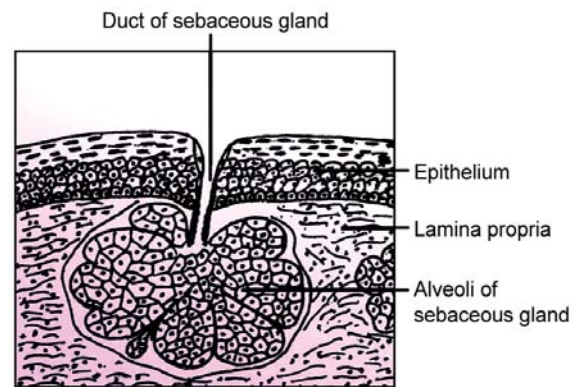


Figure 9.75: Sebaceous gland (Fordyce's granules) in mucous membrane of cheek (as seen under light microscope)

Anticancer drugs affect the oral mucosa and result in ulceration.

The desquamated cells present in saliva are settled on the dorsum of the tongue and form a white coating. This coating is more thick in fever or xerostomia.

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Bone and Alveolus

- Introduction
- Histology of bone
- Development of alveolus
 - Structure of alveolar process
- Alveolar bone proper
 - Bundle bone
 - Supporting alveolar bone
 - Cortical plates
- Types of cells present in formation of bone
- Alveolar process of maxilla and mandible
- Physical properties of alveolar bone
- Physiological changes in alveolar bone
- Internal reconstruction of bone
- Clinical and therapeutic considerations

INTRODUCTION

The periodontium consists of the four connective tissues, two mineralized and two fibrous. The two fibrous tissues are the periodontal ligament and the lamina propria of the gingiva. The mineralized tissues of periodontium consist of cementum and alveolar bone (cementum has been described in Chapter 7). The alveolar process is that part of maxilla and mandible, which primarily supports the teeth. It means that the alveolar process is the bone of jaws that contains sockets for the alveoli of the teeth (Fig.10.1)

HISTOLOGY OF BONE

Compact bone consists of outer sheet of dense bone and central medullary cavity. Cavity is filled with yellow or red bone marrow. Periosteum is an outer condensed fibrocollagen layer of bone. Periosteum consists of two layers, outer, dense, irregular connective tissue layer is known as fibrous layer and inner layer contains bone cells, their precursors and rich blood supply.

Endosteum is a thin cellular layer on the inner surface of the bone which lines the medullary cavities. Haversian system or osteon is a metabolic unit of bone and contains haversian (vascular) canal surrounded by small concentric layers of lamellae known as concentric lamellae. Circumferential lamellae surround the bony surface and contain parallel layers of lamellae.

Reversal line marks the extension of bone erosion prior to the formation of osteon.

This line is formed by the scalloped outline of the Howship's lacunae and rich in glycoproteins and proteoglycans. Resting line marks the period of rest during the formation of bone.

Volkman's canals interconnect the haversian canals. Osteocytes are present at the junction of lamellae in lacunae. Small canaliculi connect all the osteocytes. Interstitial lamellae are the remnants of osteons which are left behind during the bone remodeling.

Type of Bone

Histologically bone can be mature bone or immature bone.

Mature bone is made up of (a) Compact bone or cortical bone and (b) Cancellous bone. Immature Bone also called Woven bone is the bone usually seen in embryonic life or during fracture repair in adults. Alveolar bone has component of Woven bone. This bone is rich in osteocytes and does not have lamellar structure (Figs 10.2A and B).

DEVELOPMENT OF ALVEOLUS

The maxilla as well as the mandible forms a groove at the end of the second month of fetal life which is open toward the oral cavity. These grooves contain tooth germs, alveolar nerves and vessels. Tooth germs gradually develop bony

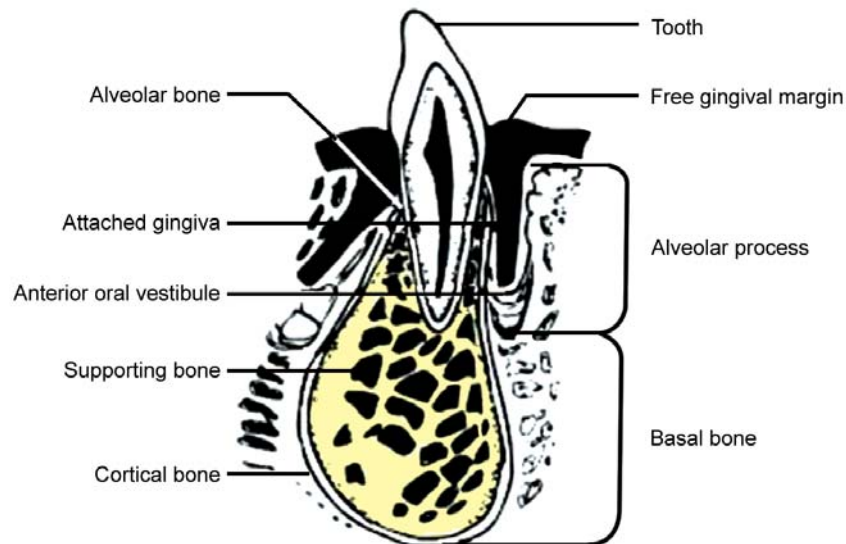


Figure 10.1: Vertical section of mandible showing relationship of tooth to alveolar process and basal bone



Figure 10.2A: Development of alveolar bone: Section from the upper alveolar ridge of 14- week- old fetus showing two tooth germs (1,2) lying close to each other. Alveolar bone formation (3) is seen at the periphery of the dental follicles (H & E x 50)

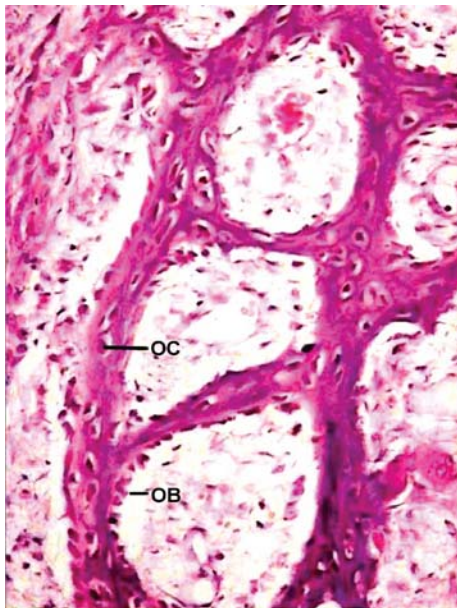


Figure 10.2B: Immature Bone: Immature bone is rich in osteocytes (oc) and prominent osteoblasts (OB) rimming the bone surface are present. Resting lines are not present x 100

septa, in between them. Later on, a horizontal plate of bone separates the mandibular canal from the dental crypts (Fig. 10.2).

The alveolar process develops only during the eruption of teeth and it gradually diminishes in height after the loss of teeth. A part of the alveolar process is included in the maxillary and the mandibular body. During growth its free border grows rapidly. A chondroid bone develops at the alveolar crest that combines the characteristics of cartilage and bone (Fig. 10.3).

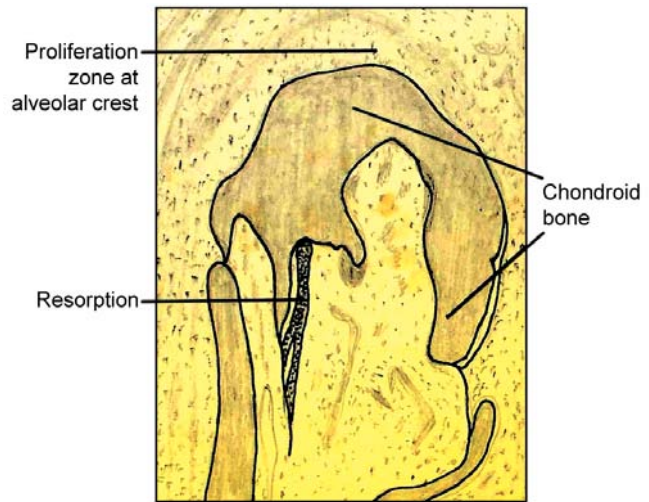


Figure 10.3: Vertical growth of mandible at the alveolar crest to form alveolar process. Formation of chondroid bone is seen which is later replaced by typical bone

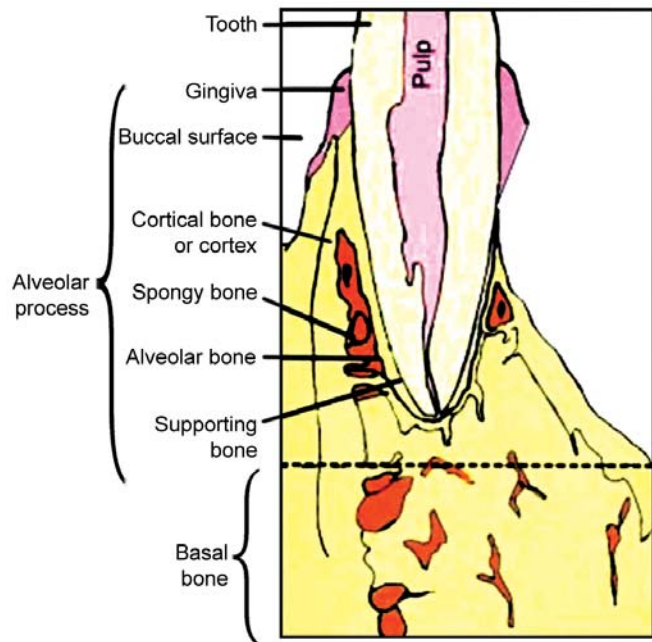


Figure 10.4: Section through mandible showing basal bone and parts of alveolar process and their relationship to the tooth

Structure of Alveolar Process

The part of the maxilla or mandible, which supports and protects the teeth is known as the alveolar bone. The alveolar bone consists of outer cortical plates, a central spongiosa and the bone lining the socket

(Fig. 10.4). The alveolus meets at the alveolar crest, below the level of the cemento-enamel junction of the tooth. There is no differentiation between the body of the maxilla or the mandible and their alveolar process, anatomically. In some places, the alveolar process fuses with bone that is not functionally related to it. For the sake of convenience of description and understanding, the alveolar process is divisible into following portions. It must be emphasized however, that these portions are not separate anatomical entities unto themselves, rather they constitute a unit, anatomically as well as functionally.

According to function, to which the alveolar process adapts itself, it is divided into two parts.

ALVEOLAR BONE PROPER (FIG. 10.5)

It is that part of alveolar process which constitutes the inner wall of the bony socket (or alveolus) surrounding the roots of teeth. This is also known as lamina dura or cribriform plate. It is called cribriform plate as it has many minute perforations or holes through which neurovascular structures pass through. Alveolar bone proper or lamina dura is constituted by the (A) Bundle bone and (B) Cortical plates.

Bundle Bone (Fig. 10.6)

It is the term applied to that portion of inner alveolar wall which gives attachment to the periodontal ligament fibres and into which Sharpey's fibres are inserted. This

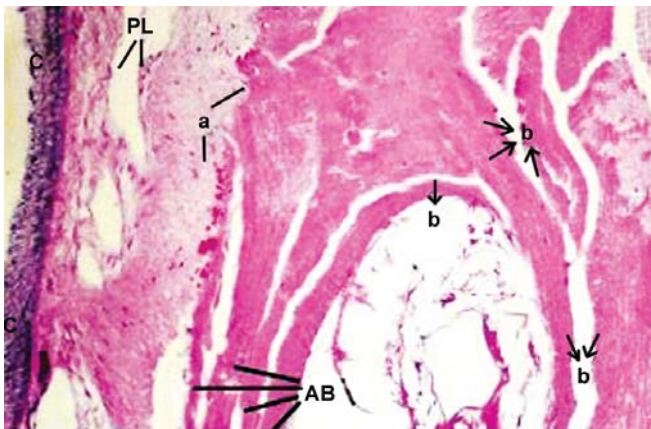


Figure 10.5: Alveolar bone: Alveolar bone (AB) at the periphery of periodontal ligament (PL) x 30 (a) Alveolar bone proper, (b) Supporting alveolar bone, (c) Cementum

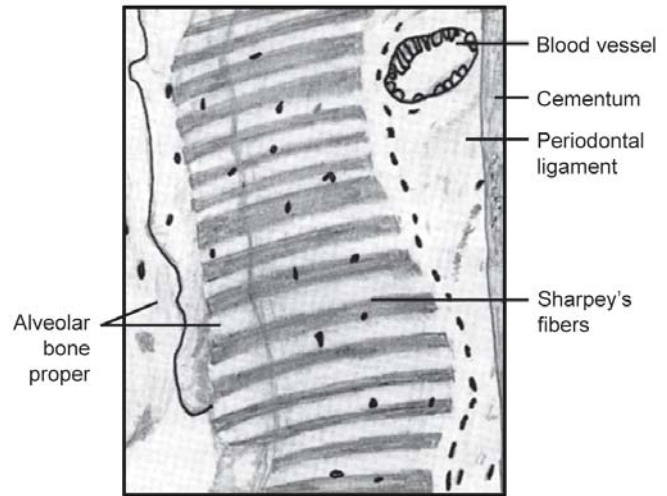


Figure 10.6: Histologic section showing continuation of bundles of principal fibers into the alveolar bone as Sharpey's fibers

portion is characterized by the thin bone lamellae which are arranged parallel to the root surface. It derives its name from the abundance of collagen fiber bundles (Sharpey's fibers) into it, and its presence is not peculiar to the jaws. Rather bundle bone is found wherever attachment of muscles and ligaments to the bone is present in the body. Hence it is evident that bundle bone forms the innermost portion of inner socket wall or lamina dura. Bundle bone is a part of alveolar bone proper and lamina dura.

Lamina Dura

The bundle bone is surrounded by dense, lamellated bone, and this dense bone along with bundle bone constitute the lamina dura or inner socket wall or alveolar bone proper. (Figs 10.7 A and B) depicts the perforated structure of lamina dura.

Supporting Alveolar Bone

Spongy or Cancellous Bone (Fig. 10.7C)

It is the cancellous bone with presence of cancellous bone trabeculae, lying between the cortical plates and alveolar bone proper, i.e. the spongy bone present between two compact layers of bone. Spongy bone or cancellous bone is not present always between these two layers at all areas of jaws, and its amount also varies considerably at different areas of alveolar processes of the jaws. Alveolar spongiosa (or spongy bone) is divided into two main types on the basis of radiographic studies.

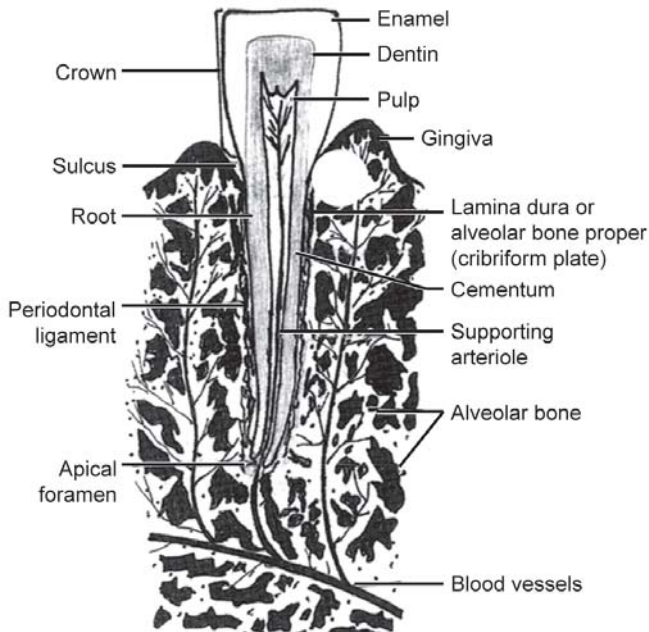


Figure 10.7A: Longitudinal section of the tooth and its environment. The bone lining the tooth socket (Lamina dura or alveolar bone proper) is known as cribriform plate because of its sievelike character



Figure 10.7B: Cribriform plate or alveolar bone proper (ABP) showing a foramen through which blood vessels are passing. Cementum (C), Periodontal ligament (PDL) Dentin (D) x 40

Cortical Plates

This is the external, compact covering of the alveolar process. It is thicker in mandible than maxilla and thickest in mandibular buccal region, more so posteriorly than anteriorly. It is composed of dense, compact, haversian bone. (Figs 10.8 and 10.9)

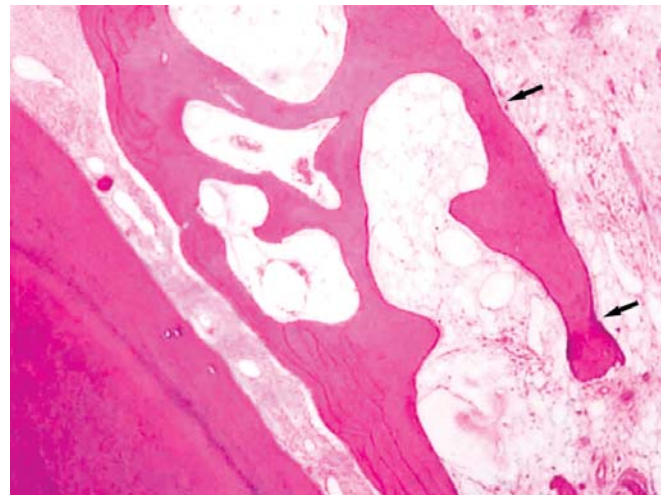


Figure 10.7C: Supporting trabeculae (shown by arrow) gives support to the alveolar bone proper

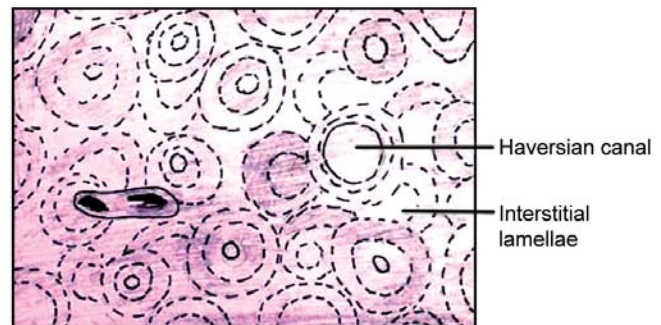


Figure 10.8: Ground section of compact bone

Histologically, cortical plates contain longitudinal lamellae and Haversian systems. In lower jaw, circumferential lamellae extend from body of the mandible into the cortical plates (Fig. 10.9).

Canals of Zuckerkindl and Hirschfeld (Nutrient canals) are present in the inter radicular and interdental septa. These canals carry the interdental and inter-radicular arteries, veins, lymph vessels and nerves (Fig. 10.10).

FUNCTIONS OF ALVEOLAR BONE

Important functions of alveolar bone are as follows.

1. Anchors the teeth with the help of Sharpey's fibers.
2. Protects the developing tooth bud of primary and permanent teeth.
3. Absorb and distribute the occlusal forces
4. Supply blood to the periodontal ligament.
5. Organizes and controls the eruption of tooth.

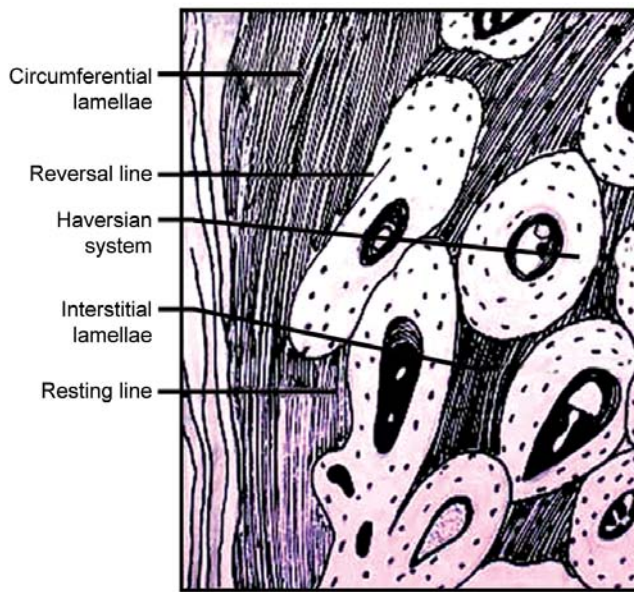


Figure 10.9: Formation of circumferential lamellae for appositional growth of mandible. These lamellae are formed and replaced by Haversian bone. Interstitial lamellae are the remnants of circumferential lamellae in the depth

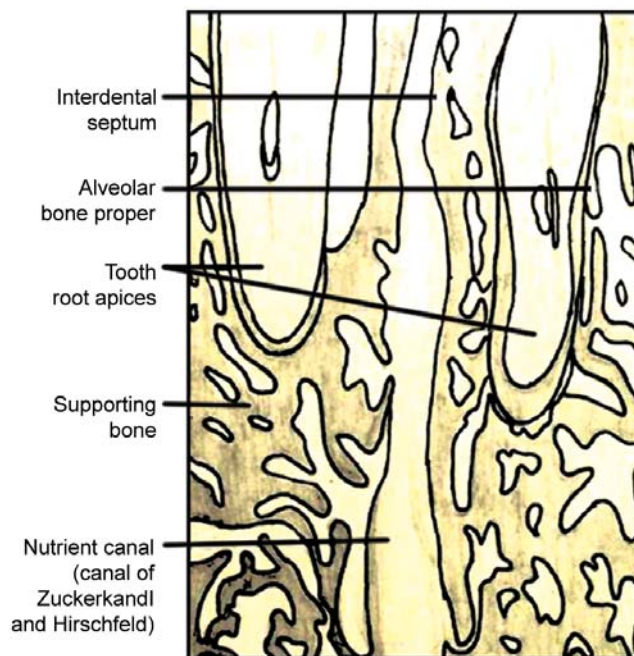


Figure 10.10: Section through jaw showing interdentary bony septum containing canal of Zuckerkandl and Hirschfeld or nutrient canal

6. Houses the roots of permanent and deciduous teeth.
7. Helps in the movement of teeth for proper occlusion.

Spongiosa of the alveolar process is divided into two main types on the basis of radiographic studies.

Type-I: The interdental and interradicular trabeculae are regular and horizontal in a ladder like form.

Type-II: The interdental and inter-radicular trabeculae are irregularly arranged and are numerous and delicate (Fig. 10.7B).

In both the types, there is variation in thickness of trabeculae and size of marrow spaces.

The type I architecture is mostly present in the mandible and is suitable for the trajectory pattern of spongy bone. In type II there is no distinct trajectory pattern but it is compensated by more number of the trabeculae per square centimeter. The type II arrangement is mostly present in maxilla. From the apex of mandibular molars trabeculae may sometimes radiate in a distal direction. Then radiating trabeculae are not prominent in maxilla as the maxillary sinus and the nasal cavity are close to the roots of the molars.

TYPES OF CELLS PRESENT IN FORMATION OF BONE

Five types of cells can be found in the formation of bone.

Osteoblasts

These are the bone forming cells and are found on the outer surface of the bone.

These are uninucleated cells and synthesize collagenous and noncollagenous proteins. These are derived from multipotent mesenchymal cells. Osteoblasts, under light microscope look like plump cells with open-faced nuclei and abundant basophilic cytoplasm. These have prominent Golgi bodies, rough endoplasmic reticulum, mitochondria, nucleoli, secretory vesicles and vacuoles (Fig 10.11). Periosteum is important reservoir of osteoblasts. Osteoblasts produce organic matrix of bone. The matrix is deposited around the cell bodies and the cytoplasmic processes of osteoblasts form the canaliculi. Osteoblasts contact one another by adherens and gap junction. Important functions of osteoblasts are as follows.

1. Formation of new bone

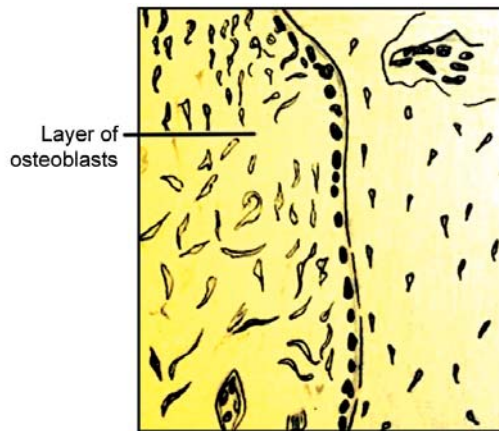


Figure 10.11: Alveolar bone lined by a layer of osteoblasts

2. Regulation and control of bone remodeling
3. Regulation and control of mineral metabolism
4. Important role in mineralization of osteoid

Osteocytes

Osteoblasts get trapped in their own secretion and subsequently become incorporated into the matrix as osteocytes. The number of osteocytes which are formed from osteoblasts depends upon the rapidity of bone formation. During their lifespan, osteocytes may resorb the surrounding matrix creating lacunae around it. Woven bone and repair bone contain more osteocytes. Narrow extensions from osteocytic lacuna are called as canaliculi. Canaliculi contain osteocytic processes and permit the diffusion of nutrients, gases and other waste products between the osteocytes and blood vessels. Canaliculi connect osteocytes with the osteoblasts and bone lining cells. This connection maintain the vitality and integrity of bone. Osteocytes also secrete matrix protein.

Osteoclasts

Large multinucleated giant cells responsible for resorbing bone are called osteoclasts. The cell is 40 to 100 micron in diameter and contains 15 to 20 closely packed nuclei. Cells with more nuclei resorb more bone than cells with less nuclei. The cell body is irregularly oval and may show branching processes. These are found in depressions called as Howship's lacunae. These have prominent mitochondria, lysosomes, vacuoles and little rough endoplasmic reticulum. Their nuclei have condensed chromatin and a single nucleolus. Osteoclasts are derived

from circulating blood cells called monocytes but can differentiate from the mesenchymal cells (Figs 10.12A & B and 10.13).

Factors affecting the activity of osteoclast are as follows.

1. Osteoprotegerin inhibit the formation of osteoclast and resorption of bone.
2. Estrogen suppress the activity of osteoclast.

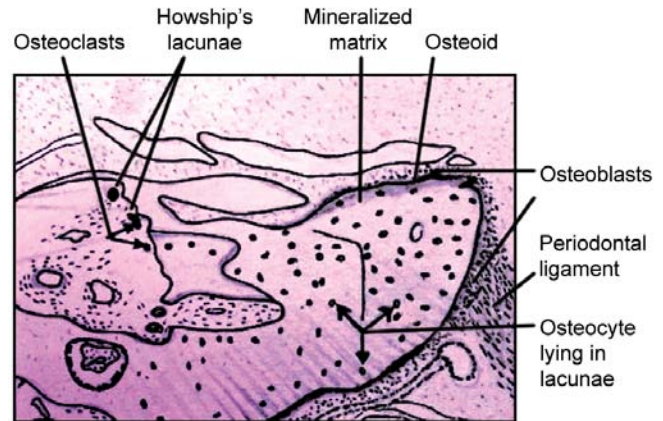


Figure 10.12A: Alveolar bone showing distribution of three main cell types osteoblasts, osteocytes and osteoclasts

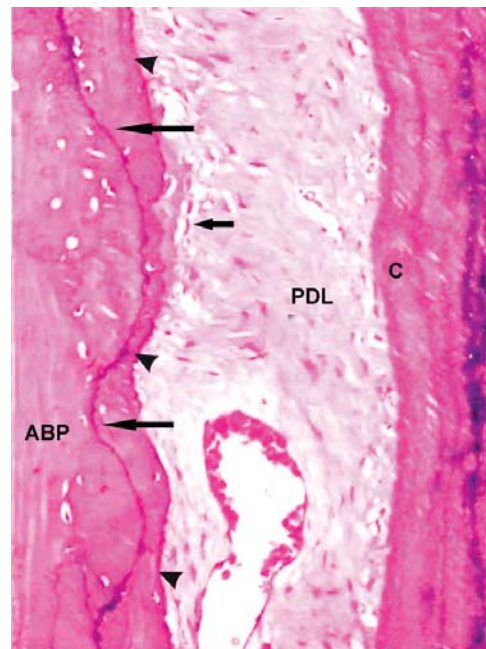


Figure 10.12B: Reversal line: Reversal line (indicated by arrow heads) is irregular and is located just adjacent to the osteoblastic layer. It has to be differentiated from line of apposition (indicated by long arrow). Osteoblasts (indicated by short arrows), Periodontal ligament (PDL), Alveolar Bone Proper (ABP), Cementum (C) x 100

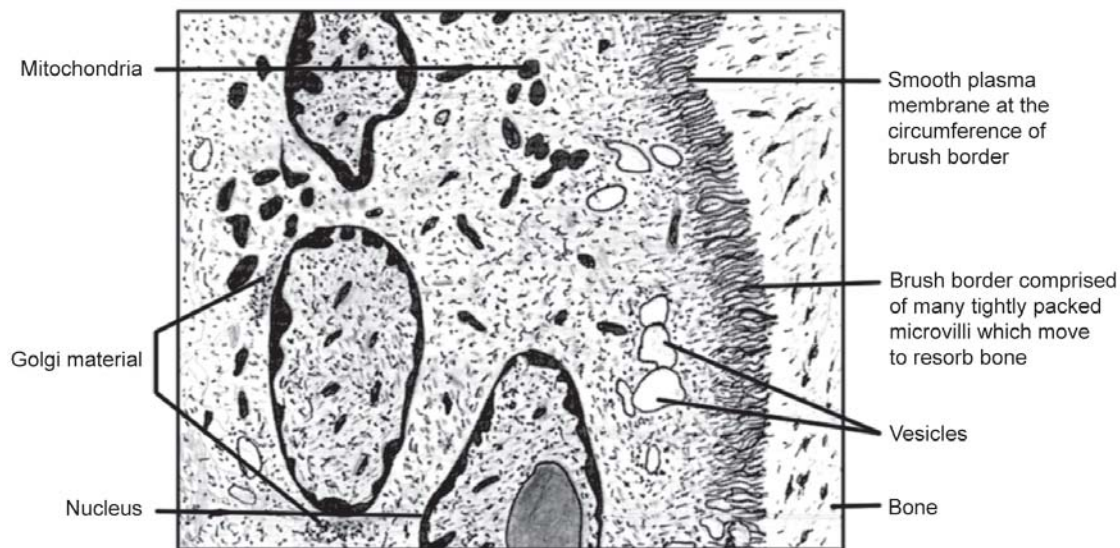


Figure 10.13: Features of a multinucleated osteoclast (as seen under electron microscope)

3. Vitamin D₃ and parathyroid hormone stimulate the activity of osteoclast.
4. Calcitonin inhibit the activity of osteocytes
5. TNF- β promote the differentiation of osteoclasts.
6. Osteoclast inhibitory lectin (OCIL) and interferon $\sim\gamma$ inhibit the formation of osteoclasts.
7. Biophosphonates inhibit the bone resorption by the osteoclasts.

Osteoprogenitor cells: These are mesenchymal, fibroblast-like cells, regarded as forming a stem cell population to generate osteoblasts.

Bone lining cells: They may represent inactive osteoblasts. Little is known about them. When alveolar bone is not deposited or resorbed by bone forming cells or bone resorbing cells, its surface is lined by undifferentiated, flattened cells termed bone-lining cells. These cells are vital, contain few organelles and retain gap junctions with osteocytes.

ALVEOLAR PROCESS OF MAXILLA AND MANDIBLE

In the alveolar process of maxilla, cortical plate is generally much thinner than in the mandible. It is thickest in the premolar and molar regions of the lower jaw, especially on the buccal side, than in the upper jaw. In the premolar and molar region of the maxilla, defect of the outer wall of the cortical plate is fairly common, because it is perforated by many small openings through which blood vessels and lymph vessels pass.

In radiographs, regular and horizontal trabeculae (Type I) are seen most often in the mandible. They give a general idea of a trajectory pattern of spongy bone. In the maxilla, irregularly arranged, numerous delicate interdental and interradicular trabeculae (Type II) are present. They do not give a general idea of a trajectory pattern of spongy bone (Figs 10.14A & B and 10.15).

PHYSICAL PROPERTIES OF ALVEOLAR BONE

Bone is a special mineralized connective tissue. It consists of 35 percent organic matrix and 65 percent of inorganic matrix. The inorganic material almost exclusively consists of calcium and inorganic orthophosphate in the form of needle-like crystals or thin plates about 8 micron thick and of variable length, that is hydroxyapatite.

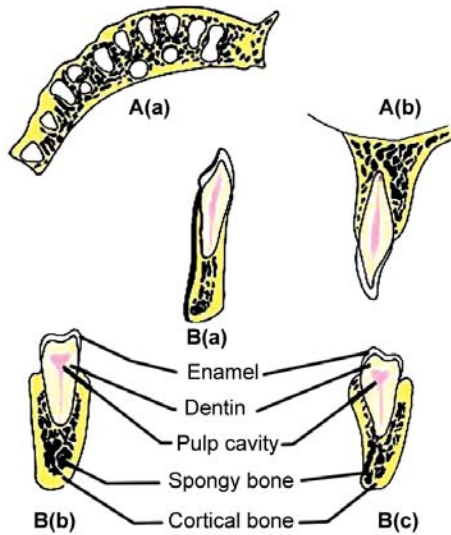
The organic material consists of about 88 to 89 percent Type I collagen. In addition, small amounts of other proteins such as osteocalcin, osteonectin, osteopontin, proteoglycans and glycoproteins are also present. The glycoproteins which are present in the ground substance consist of monosaccharides, disaccharides, polysaccharides or oligosaccharides. The proteoglycans are sulfated and nonsulfated glycosaminoglycans. The non collagen is 11 to 12 percent. It consist of following:

Glycoproteins = 6.5 to 10 percent

Proteoglycans = 0.8 percent

Sialoproteins = 0.35 percent

Lipids = 0.4 percent



Figures 10.14A and B: Sections of alveolar processes; A. Maxillary: (a) Horizontal, through alveolar process without teeth, (b) Labiolingual through permanent central incisor. B. Mandibular: (a) Labiolingual through permanent canine, (b) Buccolingual through permanent first molar (Distal root), (c) Buccolingual through second molar

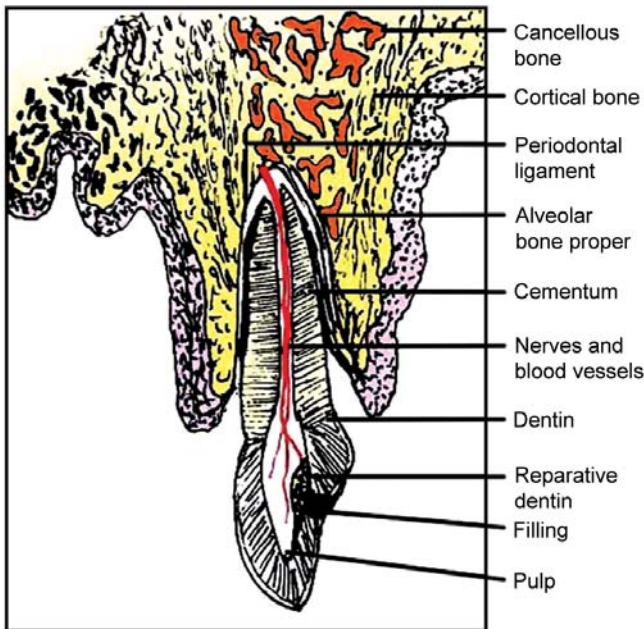


Figure 10.15: Labiolingual cross-section of the maxillary lateral incisor also showing the periradicular area and a filling and reparative dentin

PHYSIOLOGICAL CHANGES IN ALVEOLAR BONE

The inorganic crystals of bone are deposited on and inbetween the collagen molecules as well as in non-collagen material. Enzymes like alkaline phosphatase,

ATPase, and pyrophosphatase help in deposition of crystals. Because of any mechanical stress, the internal structure of bone changes. During growth and alteration of the functional stresses, it changes continuously.

Any mechanical stress results in change of internal structure of bone. The structural changes in jaw, e.g. growth, eruption, movements, wear and loss of teeth are made possible by certain destructive and formative activities. Osteoclasts remove overage bony tissue or bone while osteoblasts produce new bone (Fig. 10.16).

Osteoblasts secrete the type I collagen and noncollagen matrix of bone. The organic matrix is first devoid of mineral salts and is called as osteoid tissue. This tissue stains pink in hematoxylin and eosin stains. Some of the osteoblasts which become embedded in osteoid tissue form osteocytes. During bone formation, mineralization occurs after the production of bone matrix, so a layer of osteoid is seen. Being basophilic, mineralized bone can be distinguished from the osteoid tissue.

Osteoclasts are bone resorbing cells. These are multinucleated giant cells. At the site of great activity, the part of the cell in contact with the bone presents, a ruffled border. This is surrounded by a clear zone without organelles and with granular cytoplasm and micro-filaments. Rarely bone resorption occurs by osteocytes. This is known as osteocytic osteolysis. Bone resorption is a chemotactic phenomenon. This is initiated by the release of some soluble factors. These factors attract the blood monocytes to the target site. However, for bone resorption process genetic and functional influences are also important.

When aging osteocytes degenerate and die, they liberate certain substances, which cause the differentiation of the osteoclasts.

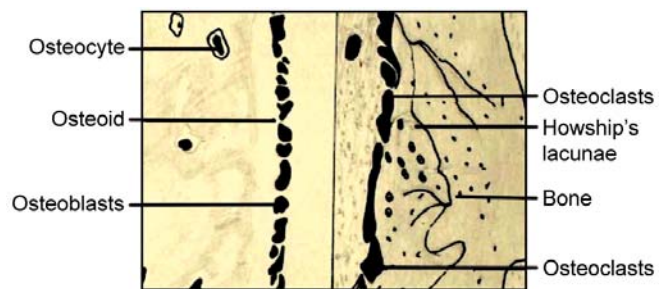
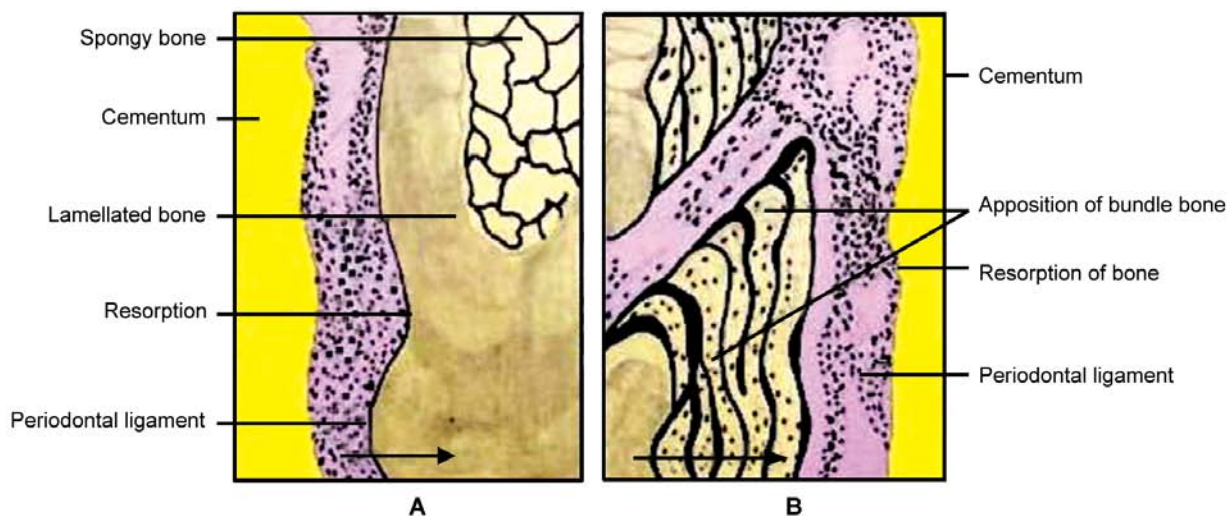


Figure 10.16: Apposition of bone at left and resorption of bone at right showing osteoblasts and osteoclasts respectively. Osteoblasts are present along bone trabeculae and osteoclasts are present in Howship's lacunae. Layer of osteoid tissue is a sign of bone formation



Figures 10.17A and B: Mesial drifting of tooth indicated by arrow. (A) Mesial alveolar wall showing resorption of bone, (B) Distal alveolar wall showing apposition of bundle bone

During bone resorption, the following three processes occur in rapid succession.

- i. Decalcification
- ii. Degradation of matrix and
- iii. Transport of soluble products to the extracellular fluid or the blood vascular system.

Bone is decalcified at the ruffled border of the osteoclasts by secretion of organic acid. These acids, e.g. citric and lactic acid chelate bone. The hydrogen ion increases the solubility of hydroxyapatite.

After decalcification, pieces of matrix are released by cathepsin B₁, lysosomal acid, protease and collagenase enzymes. Collagenase is secreted as a proenzyme that is activated by specific neutral proteases. This collagenolytic activity occurs on tropocollagen molecule. Broken fragments of collagen are further decalcified. Breakdown of collagen by proteases continue. After degradation of matrix, breakdown products of bone are transported to the extracellular fluid and to blood vascular system (Figs 10.17 to 10.19).

INTERNAL RECONSTRUCTION OF BONE

The bone forming the alveolar process is the same as bones present elsewhere in the body. The reconstruction depends upon the functional and nutritional demands of the bone. During the period of growth, bone is deposited on the outer surface of the cortical plates of maxilla and mandible.

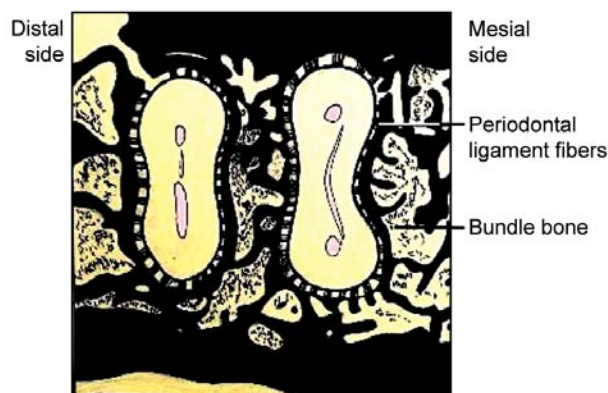


Figure 10.18: Transverse section of the root of a mandibular molar showing bundle bone adjacent to the periodontal ligament in the process of mesial migration

Mandible contains thick compact cortical plates and bone deposits in the form of circumferential lamellae. After a particular thickness of the lamellae, they are replaced from inside by haversian bone.

Osteoclast

The osteoclasts differentiate close to the surface of haversian canal and resorb the haversian lamellae and circumferential lamellae (Fig. 10.9). This area of resorption is called the cutting cone or the resorption tunnel. The bone which is resorbed is replaced by proliferating loose connective tissue.

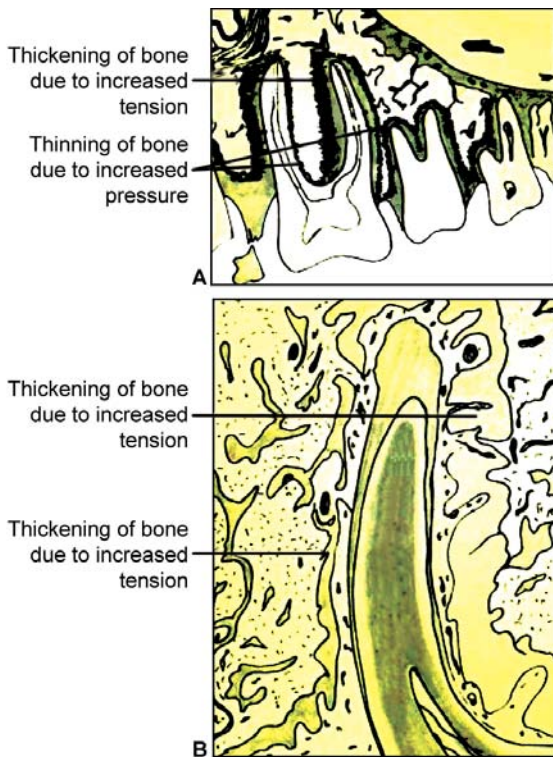


Figure 10.19: Bone response to increased occlusal forces. (A) Mesiodistal section through maxillary molar and premolars subjected to increased occlusal forces. (B) Root showing thickening of bone in response to increased tension and thinning of the bone caused by increased pressure

A reversal line (Fig. 10.12B) is seen when new bone is apposed on to the old bone, the scalloped outline of Howship's lacunae with their convexity present toward the old bone. A resting line corresponds to rest period in the otherwise continuous process of bone apposition. Resting and reversal lines are present between the layers of bone of varying age. A reversal line appears as a darkly stained cementing line.

Whenever there are physiologic eruptive movement of the teeth there is alteration in the structure of the alveolar bone. The eruptive movements of the teeth are directed mesio-occlusally. The continual apposition of the bone can be identified by resting lines, which separate parallel layers of the bundle bone. After reaching a particular thickness, the bundle bone is resorbed partly from the marrow spaces and these are replaced by lamellated bone or spongy trabeculae.

When mesial drifting of a tooth occurs, bone is deposited on the distal surface and resorbed on the mesial wall. Bundle bone is deposited on the distal wall. After

reaching a certain thickness, the osteoclasts in the marrow spaces remove part of the bundle bone. This is replaced by the lamellated bone.

On the mesial wall of a drifting tooth, there occurs active resorption, so there is the presence of the active lacunae with osteoclasts inside. Bundle bone here forms a thin layer at some places. This is because the resorption does not involve the whole of the mesial surface at the same time. The periods of resorption alternate with the periods of rest and repair (Figs 10.17 and 10.18).

During internal reconstruction of bone, compact bone may be replaced by spongy bone or spongy bone may change into compact bone. This type of change is seen in physiological mesial drift or in orthodontic mesial or distal movements of teeth. In these movements, an interdental septum shows apposition on one surface and resorption on the other surface.

If the alveolar bone proper is thickened by apposition of bundle bone, the interdental marrow spaces widen and advance in the direction of apposition. Conversely, if the alveolar bone proper is thinned by resorption, apposition of bone occurs on those surfaces that face the marrow spaces. In this way, reconstructive shift of the interdental septum of alveolar bone takes place.

CLINICAL AND THERAPEUTIC CONSIDERATIONS

Bone is one of the hard tissues of the human body. It is a biologically plastic tissue that enables the orthodontist to move the teeth without disrupting their relationship to the alveolar bone. Bone is resorbed on the pressure side because of an increase in the level of cyclic adenosine monophosphate (cAMP) in osteoclast cells. This may play an important role in bone resorption. The bone resorption is related to bacterial plaque and also to pocket formation. The gram-negative bacteria present in plaque produce endotoxin, which results in an increase in cAMP. The cAMP increases the osteoclastic activity.

Near periodontal pocket, osteoclast activating factor is present in lymphocytes. This factor increases cAMP and osteoclastic activity and reduces osteoblastic activity at the target site. Bone is apposed on tension side because tension acts as a stimulus for the production of new bone and resorbed on the side of pressure. As a result whole of the alveolus shifts with the tooth.

During healing of fractures or extraction wounds, an embryonic type of bone is formed called the embryonic

bone or immature or coarse fibrillar bone. It is characterised by a greater number, size and irregular arrangement of the osteocytes than is found in the mature bone. In radiographic appearance, the immature bone shows more radiolucency than mature bone because of the reduced volume of calcified intercellular substance and the greater number of cells present in the immature bone. This explains why a socket after an extraction wound appears to be empty in the radiograph, at a time when it is almost filled with immature bone. In radiographs, the healing of a fracture or a socket of extracted wound is seen after two or three weeks of actual formation of new bone (Fig. 10.19).

If teeth fail to develop in the jaw, the alveolar process fails to form. The most harmful change in the alveolar process is associated with periodontal disease. The bone resorption is almost universal, occurs more frequently in posterior teeth, and is difficult to repair or regenerate. Pattern of bone resorption after extraction is different in maxilla and mandible. In maxilla, ridge resorption is inward and upward while in mandible, resorption is outward and downward. After tooth loss, labial aspect of alveolar crest is the main site of resorption which first resorb in width followed by height.

The lamina dura is the most important diagnostic landmark in determining the health of the periapical tissues. Discontinuity and loss of density is usually due to infection in the periapical tissue.

On the whole, alveolar bone is well preserved as it continues to receive stimuli from tension of the periodontal ligament. Alveolar process of the maxilla and mandible develops and it supports the teeth. After loss of teeth it undergoes gradual atrophy. If during extraction of teeth, the root portion is retained in alveolar process the ridge does not undergo much reduction in height. The remaining root may be treated endodontically and retained to avoid further loss of alveolar bone. By preserving remaining roots by root canal treatment a good amount of alveolar bone is prevented from resorption and retained. This extra retained alveolar bone is very useful in obtaining better retention and stability to full denture prosthesis.

Osseous defects are successfully treated by the use of bone grafts. Bone grafts are of following types.

1. *Autograft*: Autograft is obtained from the same individual. This graft is best material for bone grafting because autograft is well accepted by the body and rate of bone growth is fastest than other graft materials.

Disadvantages of autograft are additional discomfort and secondary procedure. Sources of autograft from intraoral sites are bone from healing extraction wounds, from edentulous ridges, bone removed during osteoplasty and ostectomy and newly formed bone in wounds. Extraoral source of autograft is iliac bone.

2. *Allograft*: Graft obtained from the individual of same species but of different genotype is called allograft. Allograft may be non-demineralized or demineralized. Allograft is freeze-dried at ultra-low temperature and dried under high vacuum. The disadvantages of this graft are disease transmission and graft rejection. Allograft can be undecalcified freeze-dried bone or decalcified freeze-dried bone.
3. *Xenograft*: Xenograft is obtained from different species. Disadvantages are disease transmission and graft rejection. Processing reduce the risk of disease transmission and rejection. Boplant and Bio-Oss are the example of Xenograft.
4. *NON-BONE GRAFT MATERIALS*: Non-bone graft materials are also used in the treatment of periodontal disease. These materials are sclera, dura, cartilage, bioactive glass and coral derived materials.

BONE MORPHOGENETIC PROTEINS

Bone morphogenetic proteins like enamel matrix proteins are adjunct to the periodontal regeneration. These proteins initiate the differentiation of progenitor cells. They are used in the treatment of intrabony defects and furcations. Other proteins like growth factors and extracellular matrix provide osteogenic capacity and help in bone formation. Some synthetic materials are used to replaced bone tissue, which is lost through disease or injury. The non-resorbable hydroxyapatite and the resorbable tricalcium phosphate are used for the augmentation of the alveolar ridge. These synthetic inorganic materials are also used for filling bone defects produced by extraction wounds or periodontal diseases. Their use prolongs the life of the teeth and enhances their function.

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Salivary Glands

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INTRODUCTION

The environment of the oral cavity is moist. A film of fluid is constantly present in the mouth. This fluid is saliva, which is secreted by salivary glands. The main function of the salivary glands is the production and secretion of saliva. The saliva is very complex fluid. There are three pairs of major salivary glands, the parotid, the submandibular and sublingual. These are located outside the oral cavity, through the ducts they discharge the saliva in the oral cavity. The secretion of each of these glands is different. The parotid glands secrete watery serous saliva, which is rich in amylase. The submandibular gland produces a more mucinous saliva. The sublingual glands secrete more viscous saliva. Therefore, the saliva found in mouth is referred to as mixed saliva. There are minor salivary glands also which are situated just below or within the mucous membrane. These are uncapsulated and have very small ducts. They are grouped as labial, lingual, palatal, buccal, glossopalatine and retromolar glands.

SALIVA AND SALIVARY GLANDS

Saliva

Saliva is a complex fluid which is secreted in the oral cavity by various types of glands called salivary glands. Saliva is a balanced secretion, which results from the composition of secretion of the glands into oral cavity. Approximately 1000 to 1500 ml of saliva is secreted daily by a healthy adult person, of which 60 percent is produced by submandibular, 35 percent by parotid and 5 percent from lingual and 5 percent from the minor salivary glands.

Salivary Glands

Salivary glands are of two types:

- a. Major salivary glands
- b. Minor salivary glands

The major and minor salivary glands may be exocrine or merocrine in nature.

Major Salivary Glands

Major salivary glands are situated outside the oral cavity. These are three bilaterally present, paired glands and are named as (i) parotid, (ii) submandibular, (iii) sublingual. Major salivary glands have a long ductal system (Fig. 11.1).

Minor Salivary Glands

Minor salivary glands are present in groups beneath the oral epithelium and generally open by very small ductules. Both major and minor salivary glands are epithelial in origin which is evident by their parenchymatous structure supported by connective tissue.

FUNCTIONS OF SALIVA

The functions of the saliva, the mode of action and their active agents have been described in Table 11.1

Among the various functions of mixed saliva the main functions are as follows.

1. Protection

The function of protection is performed through many means, which are as follows.

a. Lubrication

Lubrication is the most important function of saliva. Saliva lubricates the oral cavity, which is necessary while masticating food. In this way, it protects the mucosa from friction by food during mastication. Saliva is mucilaginous because of its glycoprotein content. It forms a protective barrier against noxious stimuli, microbial toxins and minor trauma. Thus, it protects the lining mucosa of oral cavity.

b. Lavage

Saliva also acts to flush the oral cavity by mechanical washing action and prevents the collection of food particles and debris. Therefore, it delays the initiation of caries. Caries is very rampant in xerostomic patients. Saliva helps in removal of sugars from mouth so its availability to microorganisms is less. Pellicle is formed by calcium binding saliva. It protects the mucosa of the oral cavity.

c. Pellicle Formation

Shortly after cleansing the tooth surface, a thin layer is formed which is less than one micron thick and is called as pellicle. It contains glycoproteins and salivary proteins with a strong affinity for enamel. It is resistant to acids.

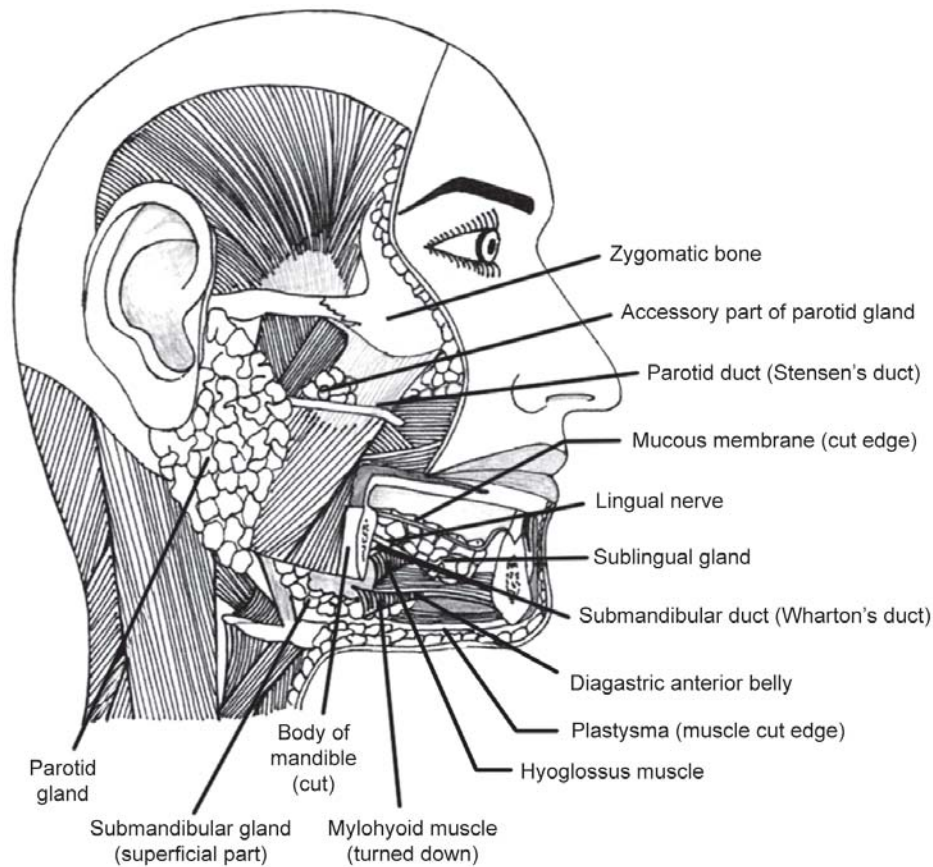


Figure 11.1: The excision of the cranial region of the superficial part of the submandibular gland has been done and the cut mylohyoid muscle has been turned down to expose a portion of the deeper part of the gland

d. Water Proofing

Due to the viscosity of saliva a somewhat water proof layer of saliva is formed on the tooth surface.

2. Digestion

Saliva helps in digestion by the following ways.

a. Bolus Formation

Saliva helps in digestion by bolus formation of food.

b. Digestion of Starch

Amylase and alpha amylase enzymes are present in saliva. These catalyze the hydrolysis of starch into smaller carbohydrate molecules. The saliva is watery in consistency and it dissolves food substances. The dissolved food can easily be acted upon by salivary enzymes amylase and alpha amylase. It also spreads over a large surface area to be easily tasted by tongue.

Salivary amylase breaks down the starch and converts it into maltose. After mastication, food is formed into a bolus by saliva, which can be easily swallowed and digested. Lingual serous glands produce lingual lipase. It helps in the digestion of dietary lipids. Triglycerides are hydrolysed to monoglycerides, diglycerides and fatty acids.

c. Neutralization of Esophageal Contents

Phosphate and bicarbonates present in saliva neutralize the esophageal contents.

3. Buffering

a. Maintenance of pH

The saliva maintains the pH of the oral cavity and thus creates an environment not favorable for growth of certain bacteria, thereby preventing formation of acids and toxins. Buffering capacity of saliva protects the oral cavity by two ways. Bacteria require specific pH conditions for

Table 11.1: Functions of saliva, their mode of action and active agents

Functions	Mode of action	Active agents
1. Protection	<ul style="list-style-type: none"> Lubrication Lavage Pellicle formation Water proofing 	<ul style="list-style-type: none"> Glycoproteins Water Salivary protein and glycoprotein Provide viscous somewhat waterproof layer on the surface of the teeth.
2. Digestion	<ul style="list-style-type: none"> Bolus formation. Digestion starch. Neutralization of esophageal contents 	<ul style="list-style-type: none"> Water Amylase and alpha amylase enzymes Phosphate and bicarbonate. Phosphate and bicarbonate
3. Buffering	<ul style="list-style-type: none"> Maintenance of pH unsuitable for colonization of micro-organisms. Neutralization acid. 	<ul style="list-style-type: none"> Phosphate and bicarbonate
4. Taste	<ul style="list-style-type: none"> Solution of molecules Taste buds growth and maturation. 	<ul style="list-style-type: none"> Water Gusten
5. Antimicrobial	<ul style="list-style-type: none"> Antibodies Hostile environment Barrier effect 	<ul style="list-style-type: none"> IgA, IgG and IgM Lysozyme and lactoferrin Glycoprotein
6. Tooth Integrity	<ul style="list-style-type: none"> Enamel maturation increases hardness, decreases permeability and increases resistance to caries. Remineralization of the enamel lesions. Decreased susceptibility: Repaired enamel lesions are less susceptible to future decay. 	<ul style="list-style-type: none"> Calcium and phosphate ions in saliva. Calcium and phosphate ions in saliva. When fluoride ions are also present in saliva, they get impregnated leading to acid resistant enamel.
7. Soft tissue repair	<ul style="list-style-type: none"> Reduces bleeding time. Rate of wound contraction is increased. Rate of wound healing enhances. Formation of blood clot. 	<ul style="list-style-type: none"> Active agent yet to be isolated-probably viscosity of saliva also helps in checking bleeding. Epidermal growth factor produced by submandibular glands.

maximal growth. Buffering capacity of saliva prevents colonization of pathogenic bacteria by not creating optimal environmental conditions. Secondly, the microorganisms, which produce acids from sugar, are removed by saliva, otherwise the acid can demineralize the tooth. Buffering capacity of saliva is due to bicarbonate and phosphate ions.

Sialin: It is a negatively charged residue of salivary protein. It is a peptide in saliva, which acts in raising the pH of plaque preventing demineralization and cavitation of enamel. Hence it acts as a buffer.

b. Neutralization of Acids

Buffering neutralizes acid on the surface of the tooth as soon as it is formed, thereby preventing dental caries.

4. Taste

a. Solution of Molecules

With the water of the saliva the molecules of the food are dissolved. Thus, saliva acts as a solvent for certain food materials and helps in their digestion. In this way the taste buds can taste the food.

b. Taste Bud Growth and Maturation

Saliva helps in the growth and maturation of the taste buds.

5. Antimicrobial

There are several defensive substances in the saliva which inhibit the growth of microorganisms and prevent infection.

Antibacterial and antifungal activities of saliva maintain the balance of microorganisms present in the oral cavity. The presence of certain bactericidal constituents also helps to inhibit the growth of microorganisms and prevent infection.

a. Antibodies

Many immunoglobulins are present in saliva. Among them important are IgA, IgG and IgM.

Main salivary antibody is IgA which is produced locally in salivary glands by plasma cells.

This antibody coagulates or agglutinates the bacteria by killing them. Secretory component is a glycoprotein, which is produced by parenchymal cells. This secretory component is a part of the IgA molecule. Secretory component helps in the transfer of IgA into the lumen. It also increases the resistance of the IgA molecule to denaturation or proteolysis.

b. Hostile Environment

There are many substances in saliva which make the oral environment hostile to pathogenic microorganisms. The main among them are.

Lysozyme: It is an enzyme (antibacterial protein) which hydrolyzes the polysaccharide of bacterial cell walls resulting in cell lysis.

Lactoferrin: It is an iron-binding protein, which is found in saliva. It has antibacterial properties.

Barrier effect: Glycoproteins form a barrier or protective covering and prevent adherence of bacteria on the oral surfaces and facilitate their removal from the oral cavity.

6. Tooth Integrity

Saliva is saturated with calcium and phosphate ions. As soon as the tooth erupts high ionic exchange with the tooth surface occurs and enamel maturation takes place, due to these calcium and phosphate.

a. Enamel Maturation

Enamel maturation increases hardness, decreases permeability and increases resistance to caries.

b. Remineralization

Due to high content of calcium and phosphate ions, remineralization of the lesion of enamel takes place.

c. Decreased Susceptibility

When fluoride ions are also present in saliva, repaired enamel lesion becomes less susceptible to future decay as the fluoride ions get impregnated leading to acid resistant enamel.

7. Soft Tissue Repair

a. Reduction in Bleeding Tissue

Saliva helps in the soft tissue healing as follows: Clinically it has been shown that the bleeding time of oral tissues even though they are highly vascular is shorter than other tissues of the body. The active agent is yet to be isolated. Probably viscosity of saliva helps in checking bleeding.

b. Increase in Rate of Wound Contraction

The rate of wound contraction is significantly increased in presence of saliva due to epidermal growth factor produced by submandibular salivary glands.

c. Rate of Wound Healing Enhances

In presence of saliva the rate of wound healing is enhanced.

d. Formation of Blood Clot

The wounds in the oral cavity can be repaired very easily by protective action of saliva which helps in the clotting of blood.

An additional specialised function of thermoregulation occurs in some mammals who lacks sweat glands. Certain structural and functional changes occur in the salivary glands due to thyroid and pituitary hormones. When patients with Addison's disease and Cushing's syndrome are given adrenocorticotrophic hormones or mineralocorticoids, an alteration of salivary $\text{Na}^+ : \text{K}^+$ ratio is seen.

STRUCTURE AND ANATOMY OF SALIVARY GLANDS

The parenchyma of the gland consists of a series of ducts, which end in terminal secretory end pieces like a bunch of grapes. The grapes represent the secretory end pieces. The stalks represent the duct system. The main secretory duct of the salivary gland breaks and divides into smaller and smaller ducts. The ultimate smaller duct opens into the blind terminal secretory end pieces.

The terminal end pieces greatly vary in size, shape and cell numbers. Their shapes may vary from simple circular outline to multilobed polygons. These cells enclose a central space between the cells, which is called as lumen, from here the ductal system starts.

The structure of these glands can be termed as compound racemose, as these glands are made up of numerous lobes, which in turn are divided, into lobules. The lobules consist of a series of duct endings in terminal secretory end pieces, which produce a primary secretion unit (Fig. 11.2).

There are four types of terminal end piece cells. These are as following.

- i. Serous
- ii. Mucous
- iii. Seromucous
- iv. Myoepithelial.

The first three (serous, mucous and seromucous cells) are of secretory variety and the myoepithelial cells are of nonsecretory variety. The main secretory duct of salivary glands are divided into smaller ducts or striated ducts which are then further divided into smaller intercalated ducts and open into terminal secretory end pieces. The secretion from these end pieces follows the route in which the ducts divide (Fig. 11.3).

Secretory end piece → Striated duct → Intercalated duct → Terminal excretory ducts.

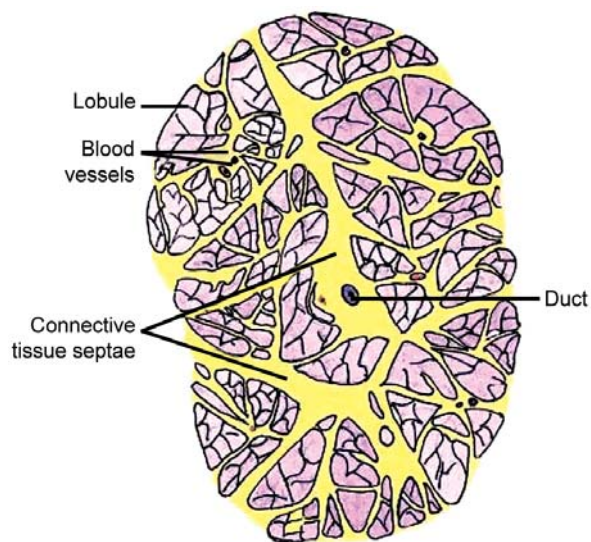


Figure 11.2: Salivary gland divided into successively smaller lobules by connective tissue septae in a low-powered photomicrograph

Types of Secretory Cells

- A. According to shape
 - a. Spheroidal or acinus or pyramidal
 - b. Tubular or cylindrical
 - c. Tubuloacinar that is intermediate in shape
- B. According to production of salivary contents
 - a. Serous
 - b. Mucous
 - c. Seromucous

Homocrine and Heterocrine Glands

- i. A gland containing only one type of secretory end piece cells is called homocrine, e.g. parotid- salivary gland.
- ii. A gland containing more than one type of secretory end piece is called heterocrine, e.g. Submandibular salivary gland.

Description of Secretory End Piece Cells

Serous Cells

Serous cells are specialized cells which synthesize and store proteinaceous material. They also synthesize polysaccharides. These are often called as seromucous cells. These cells are pyramidal in shape with their base resting on the basal lamina and apex toward the lumen. The nucleus is spherical in shape and is located in the basal third of the pyramid. Accumulation of secretory granules in the apical cytoplasm is the most prominent feature of this cell (Figs 11.4 and 11.5). Secretory granules are one to twenty micron in diameter and zymogen granules are formed by glycolated proteins.

These cells have a secretory function and contain all the specialized organelles related to secretion. These cells contain a closed system of membranous sacs or cisternae, rough endoplasmic reticulum is present on the basal portion of the cytoplasm. The ribosomes which are present in rough endoplasmic reticulum consist of ribonucleic acid and proteins. These are the basic units of protein synthesis. The messenger RNA from the nucleus direct the ribosomes to translate the encoded message. The ribosomes add the appropriate amino acids in their proper sequence in the protein which is being synthesized. Proteins are first synthesized as preproteins. These consist of 16 to 30 amino acids and an amino terminal extension. It is called a signal sequence. The signal sequence is recognised by a specific protein in the rough endoplasmic

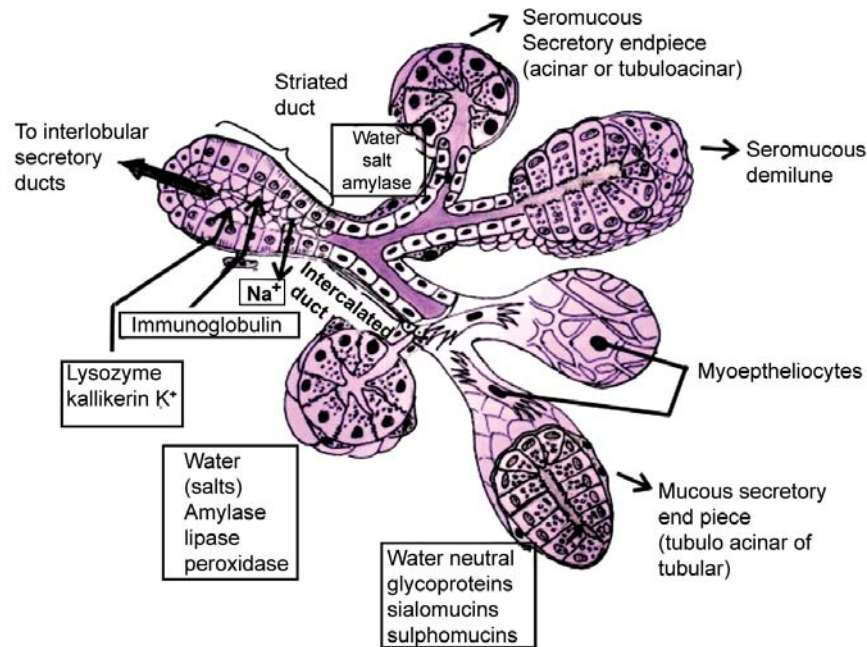


Figure 11.3: Generalized architecture of a salivary gland

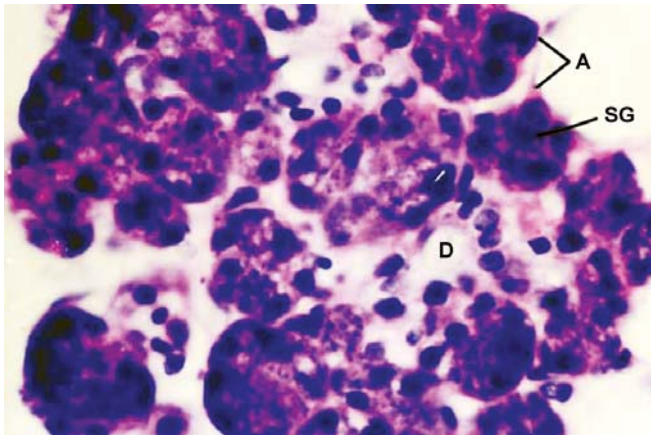


Figure 11.4: Serous cells: High power view of parotid gland showing acini lined by serous cells. Cells are pyramidal in shape with apex towards the center of acinus. Nuclei are round and are located near the base of cells. Apical portion of cells shows dark secretory granules in the cytoplasm. Intercalated ducts can be seen situated between the acini x 150. (A) Acinus, (SG) Secretory Granules, (D) Intercalated duct

reticulum. It directs attachment of the ribosomes to the membrane. The newly synthesized protein reaches the cisternal space of the rough endoplasmic reticulum. The sequence is removed by proteolytic enzyme known as signal peptidase and the protein assumes its three dimensional structure.

A second system of membranous cisternae, the Golgi apparatus is located apical or lateral to the nucleus. The Golgi apparatus consists of saccules. These are slightly curved or cup-shaped and their concavity or *trans* face is oriented toward the secretory surface of the cell. The Golgi apparatus is functionally connected to the rough endoplasmic reticulum through vesicles. Newly synthesized secretory proteins within the rough endoplasmic reticulum are transported to the Golgi apparatus through these vesicles.

The proteins move through the Golgi saccule toward the *trans* face of the Golgi apparatus. They are packed into vacuole of various size and density. These vacuoles are forming secretory granules, which are known as immature granules, prosecretory granules or condensing granules. The smaller immature granules increase in size as their content increases in density. These then acquire the size of the mature granule.

Before secretion of the secretory proteins, these undergo covalent structural modifications, most commonly glycosylation. In glycosylation, there is addition of a carbohydrate side chain to amino acids, which are asparagine, serine and threonine, these are present in the protein. The carbohydrates are mannose, fucose, glucosamine, galactosamine and sialic acid.

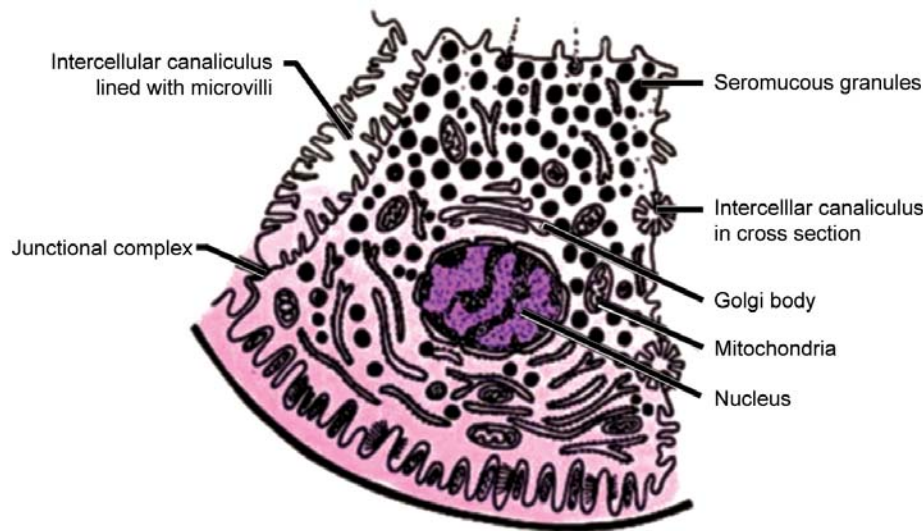


Figure 11.5: Serous cell showing intercellular canaliculi, various cell organelles and synthetic apparatus of the cells

Glycosylation begins in the rough endoplasmic reticulum and is completed in the Golgi apparatus. Other modification is the addition of phosphate or sulfate groups. The final secretory product is formed by proteolytic cleavage.

These secretory proteins are stored in secretory granules in the cell apex. These granules are discharged by the process of exocytosis. They secrete their contents into the lumen by fusing with the plasma membrane of the cell lining the lumen of the duct. By this process of exocytosis, the granule membrane become continuous with the plasma membrane.

During rapid secretion, compound exocytosis occurs. In this, a second granule may fuse with the membrane of the previously discharged granule. This results in a long string of interconnected granule profiles extending into the cytoplasm. Because of the addition of the granule membranes, there is an enlargement of the plasma membrane at the secretory surface. This excess membrane is removed by the cell as the small vesicles. Lysosome fuses with some portion of these vesicles and degrades the membrane. Some vesicles go back to Golgi apparatus and the membrane is reutilised for the formation of new secretory granules.

The parenchymatous cells are separated from the connective tissue by a basement membrane, which surrounds the end piece seromucous cells. The adjoining serous cells are joined by tight junctions and desmosomes and intercellular canaliculus or intercellular space passes between them.

Thus, a heavy amount of specialized secretory organelles do the function of:

- i. Protein synthesis
- ii. Glycosylation, which is the formation of linkages with glycosyl groups. In this, there is addition of a carbohydrate side chain to the amino acid side chain.
- iii. Production of salivary amylase.

The serous cell contains various cytoplasmic organelles. Free or unattached ribosomes are located in the cytoplasm throughout the cell. They help in the synthesis of nonsecretory cellular proteins. Mitochondria are found between rough endoplasmic reticulum cisternae, around Golgi apparatus and along lateral and basal plasma membrane. The mitochondria contain the enzyme of citric acid cycle, electron transport and oxidative phosphorylation. These are the sources of high-energy compounds. Lysosomes with hydrolytic enzyme are also present in the cell. They destroy foreign material taken by the cell. Cytochemical techniques demonstrate the presence of peroxisomes which contain the enzyme catalase and other oxidative enzymes. Tonofilament bundles with desmosomes and microfilaments are also present in the cytoplasm.

Mucous Cells

These cells also have functions similar to serous cells, as synthesis, transportation and storage of protein rich material.

These cells have a pyramidal structure similar to serous cells but have a narrow band of rough endoplasmic reticulum and larger Golgi apparatus involved in carbohydrate synthesis (Fig. 11.6). The nucleus is flat and situated in the basal region. The apex of mucous cells contains thin strands of cytoplasm forming a trabecular network. The secretory products, though proteinaceous, have a much higher carbohydrate content as compared to serous secretion, so carbohydrate metabolism is increased and there is presence of prominent Golgi complexes. It consists of several stacks of ten to twelve saccules.

The cells stain heavily for carbohydrate and are less marked for protein (with eosin and hematoxylin stains) as compared to serous cells. This shows that the ratio of carbohydrate to protein is greater.

The secretory product does not show any enzymatic activity and mainly helps in lubrication and protection of oral tissue. The mucous secretion is stored in droplets. Above the basal plasma membrane, an oval or flattened nucleus of mucous cell is present. The rough endoplasmic reticulum, mitochondria and other organelles are present in the cytoplasm along the base and lateral borders of the cell.

When secretion of mucous droplets occurs, its limiting membrane fuses with the apical plasma membrane. As a result a single membrane separates the droplets from the lumen. Now this separating membrane may fragment or may be lost with the membrane intact. When there is rapid

discharge of the droplet, the apical cytoplasm may not seal off itself and the whole mass of mucus may be spilled into the lumen.

The specific features of mucous secretion are as follows.

1. Mucous secretion contain large amount of sialic acid.
2. Ratio of carbohydrate to protein is greater than serous secretion.
3. Enzymatic activity of secretion is little or absent.
4. Important functions of mucous secretion are lubrication and protection of the oral tissues.

Myoepithelial Cells

Myoepithelial cells are contractile epithelial cells. These are also end piece cells belonging to another family having no secretory function. The myoepithelial cells are found in the vicinity of intercalated duct cells and the end piece secretory cells between the basal lamina and the basal plasma membrane. They are also called 'Basket cells' as their shape appears like a basket containing secretory cells. These cells are also abundantly found in mammary gland.

Myoepithelial cells, as the name suggests have muscle-like contractile function and these help the secretory acinus cells to expel their product. There is usually one myoepithelial cell per acini. These cells have a structure like octopus having a cell body situated in the intercalated duct region with processes extending backwards onto

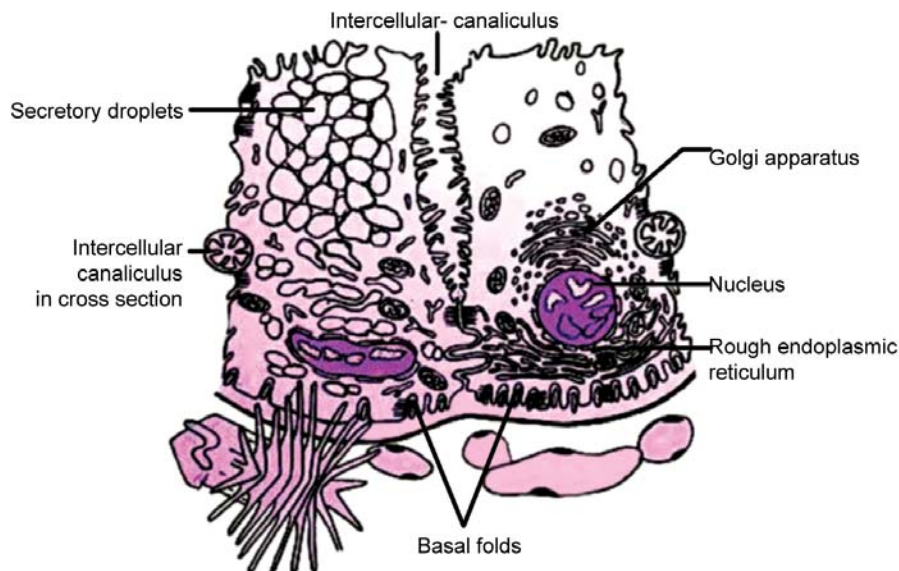


Figure 11.6: Two mucous cells at different stages of secretory cycle (in section)

the parts of the secretory end piece. The small body of the cell is filled mostly with the flattened nucleus. The myoepithelial cells have a smooth muscle-like action and structure having myofilaments containing actin and myosin. These cells contract causing pulsation and release of secretory products in spurts from the end cell. The morphology of myoepithelial cells depends upon their location. Nucleus is present in the central body of each cell. From the cell body four to eight processes radiate.

Between the myoepithelial cells and the underlying secretory cells desmosomal attachments are present. The cytoplasmic organelles are mainly located in the perinuclear region of the myoepithelial cell. Many micropinocytotic vesicles (caveolae) are present on the plasma membrane of myoepithelial cells.

Functions of myoepithelial cells are as follows.

1. Myoepithelial cells act as a support for secretory cells by preventing their overdistention, because secretory products accumulate within their cytoplasm.
2. Myoepithelial cells contract and widen the diameter of the intercalated ducts. Thus, they lower or increase their resistance to outflow.
3. Contraction of myoepithelial cells helps to rupture the acinar cells that are packed with mucous secretion (Fig.11.7).
4. The initial outflow of saliva from acini is accelerated by myoepithelial cells.
5. Myoepithelial cells help the salivary flow to overcome increase in peripheral resistance of the ducts.
6. Luminal volume is reduced by these cells.
7. These cells produce proteins, like antiangiogenesis factor and proteinase inhibitors. These proteins act as barriers against invasive epithelial neoplasms and suppress the activity of tumor.
8. Myoepithelial cells provide signals to the acinar cell to maintain the cell polarity and structural organization of the acinus.
9. Myoepithelial cells are also involved in the protection of salivary gland tissues.

DUCTAL SYSTEM OF SALIVARY GLANDS

Ductal system of salivary glands comprises of a network of small ducts which unite to form larger ducts. Ducts leading from the secretory end pieces are branched as follows (Fig. 11.8A).

Intercalated → Striated → Excretory or collecting duct.

Within a lobule, smallest ducts are the intercalated ducts. Intercalated duct connects the terminal secretory units to the larger ducts, the striated ducts. The striated ducts continue to join with the main excretory duct.

Intercalated Ducts

Intercalated ducts are the smallest ducts inserted or interposed between secretory cells. The secretion of the terminal end pieces pass first into the intercalated ducts. These ducts are lined by small cuboidal cells showing

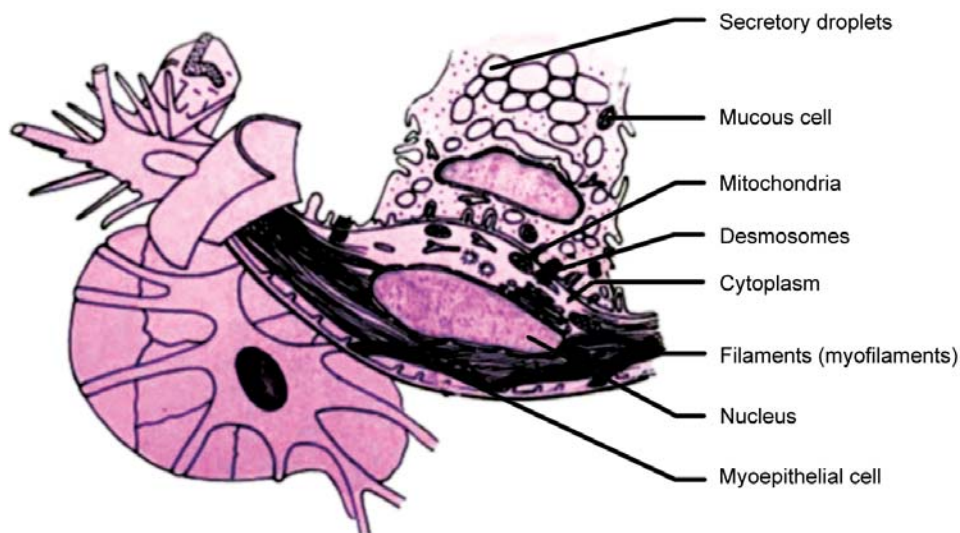


Figure 11.7: Myoepithelial cell showing myofilaments and perinuclear cytoplasm with cell organelles

characteristic of serous cells containing rough endoplasmic reticulum on the basal side and Golgi complex apically (Fig. 11.8B). These cells are attached laterally to each other by desmosomes or tight junctions and have microvilli projecting into the lumen of the duct. The cell bodies and processes of myoepithelial cells are present around the ductal cells surrounded by basement membrane. The ductal cells have the characteristic of serous cells and are frequently associated with parotid gland, which is a pure serous gland.

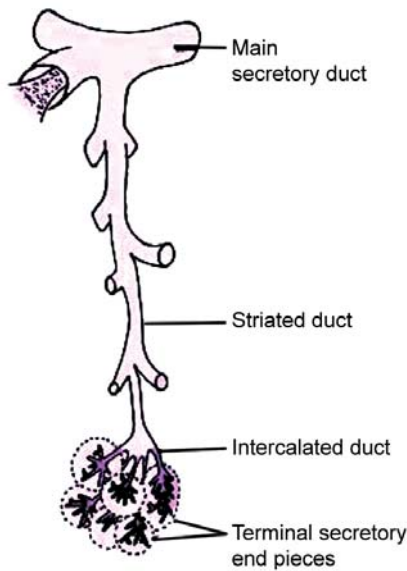


Figure 11.8A: Duct of a salivary gland

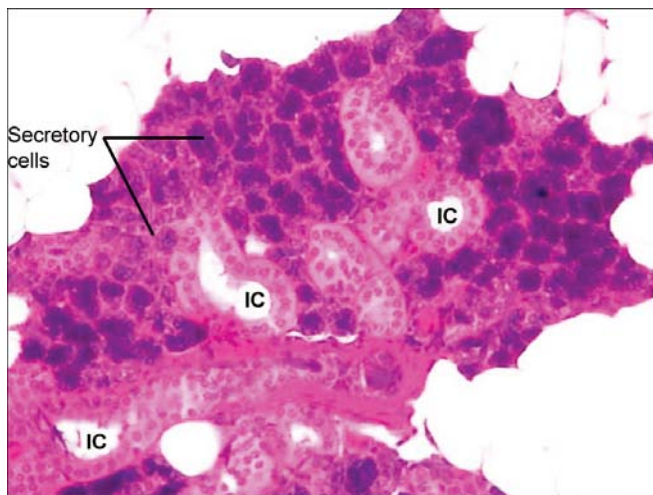


Figure 11.8B: Intercalated ducts: Between secretory cells (SC) Intercalated ducts (IC) are lined by low cuboidal cells

Functions of intercalated duct are as follows.

1. Duct modify the composition of saliva by secretory and resorptive process.
2. Secretory granules of ductal cells release lysozymes, lactoferrin and other component into the saliva.
3. Ducts act as house of undifferentiated cells which help in the repair of cells in the end piece or striated ducts.

Striated Ducts

Intercalated ducts pass into larger ducts called striated ducts. These are lined by tall columnar cells with centrally placed nuclei having abundant cytoplasm and is easily stained by eosin and hematoxylin (Fig. 11.8C).

The name striated duct is taken from its characteristic histological feature that is, presence of striations on the basal end of cells. The basal striation is caused due to deep indentations of the basal plasma membrane into the cell and a column of packed elongated mitochondria. In a small amount, rough endoplasmic reticulum and Golgi bodies are also present in the cell. Under light microscope, the infoldings and mitochondria seem like striations. Many lysosomes, numerous small peroxisomes, bundles of cytoplasmic filaments, free ribosomes and moderate amount of glycogen are present. The single layered epithelium of striated duct contains 8 and 18 simple cytokeratin intermediate filaments. Presence of large number of mitochondria indicate active transport by the cells.

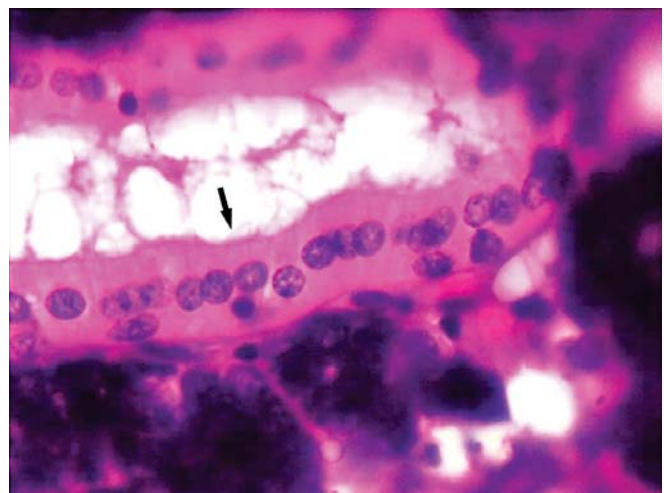


Figure 11.8C: Striated duct: Striated ducts are lined by tall columnar epithelial cells with centrally placed nuclei (shown by arrow)

The cell surface facing the lumen have short microvilli. Junctional complexes and desmosomes join adjacent cells laterally to each other. The striated cells have the important function of altering the composition of serous fluid.

In striated ducts the epithelium becomes pseudo-stratified. The smaller basal cell is situated in between the tall columnar cells. As the duct increases in size, the characteristics of the striated cells are maintained to a variable degree and become less pronounced. The epithelium of the main duct gradually becomes stratified as it gets merged with the epithelium of the oral cavity.

As the serous fluid passes through striated ducts, isotonic sodium rich fluid changes to hypotonic low sodium fluid. This occurs through the sodium pump, which establishes a concentration gradient between luminal fluid and cell.

Functions of striated ductal cells are as follows:

1. Electrolyte reabsorption
2. Ductal cells synthesize and secrete the glycoproteins like epidermal growth factor and kallikrein.
3. Protein reabsorption

Ductal cells resorb the protein by endocytic mechanisms.

Terminal Secretory (Collecting) Ducts

Terminal secretory ducts are the ducts through which salivary fluid from the glands is secreted into the oral cavity. As the duct runs to the exterior from the striated duct, its size and cell character changes. The size increases from starting to exit and cell layer changes from pseudostratified columnar epithelium to true stratified squamous epithelium. Lymphocytes, macrophages and dendritic cells are also present.

These ducts also function to alter the salivary fluid composition by altering electrolyte concentration. Towards the end of the duct mucous goblet cells are found which add mucin content to salivary secretion.

Functions of Salivary Ducts

Primary saliva secreted by terminal secretory units is conveyed by salivary ducts to the oral cavity. These ducts modify the saliva by secretion and resorption of electrolytes and secretion of proteins.

Two antibacterial proteins, lysozyme and lactoferrin are present in the cytoplasm of the intercalated duct cells. The striated duct cells, contain, Kallikrein, which is an

enzyme found in saliva and synthesizes secretory glycoproteins. The glycoproteins are stored in apical granules. The striated duct cells are involved in water and electrolyte transport.

The primary secretion of intercalated duct is isotonic or slightly hypertonic to plasma in which sodium ion and chloride ion concentrations are approximately equal to those in plasma. Potassium ion concentration is low compared to that of sodium ion but it is higher than potassium ion concentration of plasma. Bicarbonate ion concentration is variable.

The fluid of excretory duct is hypotonic, with low sodium ion and chloride ion and high potassium ion concentrations. With increasing flow of saliva, sodium ion (Na^+) and chloride ion (Cl^-) increase, bicarbonate ion (HCO_3^-) also increases while potassium ion (K^+) decreases. The striated ducts actively resorb sodium ion from the primary secretion and secrete potassium ion and bicarbonate ion, chloride ion follow the electrochemical gradient.

When there is an increased flow of saliva, sodium ion resorption becomes less as the secretion is in contact with the epithelium for a shorter time. So the sodium ion concentration of the saliva increases. The water enters saliva by terminal secretory units, the striated and excretory ducts are relatively impermeable to water. The re-absorption of potassium ion and chloride ion in the duct exceeds the secretion of potassium ion and bicarbonate ion, so the saliva becomes hypotonic in luminal fluid.

CONTROL OF SALIVARY SECRETIONS

The flow of saliva is controlled by nervous stimulation mainly through the activity of autonomic nervous system. Hormones also exert varying levels of control over salivary gland function. There is no direct inhibitory innervation of the salivary glands. Hormones can cause modification of the salivary constituents but they cannot themselves initiate flow of saliva.

Hormonal control: This does not evoke the release of salivary secretion but alters the response to neural stimuli through local hormones.

Neural control: Salivary glands chiefly depend on the autonomic nervous system to evoke secretions. The nervous system involved in secretion are cholinergic

(Parasympathetic) and adrenergic (Sympathetic). Smaller mucosal glands also show spontaneous secretion which is independent of neural control.

Secretomotor nerves from both sympathetic and parasympathetic system innervate the glands and interact with alpha and beta adrenergic and cholinergic receptors (Fig.11.9). The neurotransmitters are released from the vesicles in the nerve terminals near the parenchymal cells. As a result secretory granules are discharged which secrete water and electrolytes. The neurotransmitters interact with specific receptors located on the plasma membrane of the acinar cell.

Nor-epinephrine is a sympathetic transmitter. It interacts with both α and β adrenergic receptors. Acetylcholine interacts with the cholinergic receptors. Protein secretion is mediated through the β adrenergic receptors. Stimulation of the α adrenergic and cholinergic receptors causes low levels of protein secretion. These two receptors are also responsible for secretion of water and electrolytes.

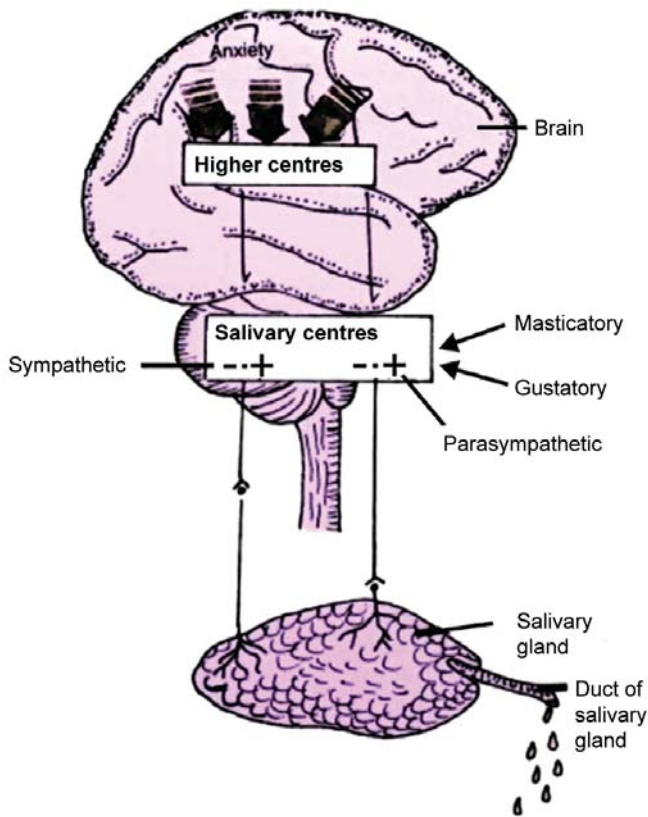


Figure 11.9: Nerve pathways involved in the secretion of saliva

Substance P is present on the salivary gland cells. These are the receptors for peptide transmission. Substance P stimulates secretion similar to that caused by α adrenergic and cholinergic agonists.

Vasoactive intestinal polypeptide (VIP) is present in nerve endings in the salivary glands. It induces secretion by some glands. Stimulation of the receptors results in an increase in the intracellular concentration of second messengers. This results in the cellular response.

After the stimulation of α -adrenergic, cholinergic and substance P receptors, the membrane permeability to calcium (Ca^{++}) is increased and there is influx of calcium (Ca^{++}) into the cells. This increased cytoplasmic calcium concentration causes potassium efflux, water and electrolyte secretion and a low level of exocytosis.

After the stimulation of β adrenergic receptors, there is activation of the plasma membrane enzyme adenyl cyclase. It catalyzes the formation of 3', 5' cyclic adenosine monophosphate from adenosine triphosphate. There is increase in the intracellular concentration of cyclic AMP which activates cyclic AMP dependent protein kinase. The protein kinase is an enzyme that phosphorylates other proteins which may be involved in exocytosis.

Adjacent secretory cells are joined to one another by specialized intercellular gap junctions. These are permeable to ions and small molecules. So, if there is any change in intracellular concentration of any substance in one cell, the same change is reflected in the adjacent cells.

Parasympathetic and sympathetic stimulation:

- i. Evokes fluid secretion.
- ii. Causes contraction of myoepithelial cells.
- iii. Controls the composition of saliva and its movement from glands.

CONNECTIVE TISSUE ELEMENTS OF THE SALIVARY GLAND

Connective tissue of the gland are surrounded by capsule. Septa from the capsule divide the gland into lobes and lobules and contain blood vessels and nerves. The ground substance is composed of glycoproteins and proteoglycans. Fibroblasts, macrophages, mast cells, leukocytes, fat cells and plasma cells are also found in the connective tissues. Immunoglobulins produced by the plasma cells are secreted into saliva by the process called as transcytosis.

NERVE SUPPLY

Salivary glands are supplied by postganglionic secretomotor nerves from sympathetic and parasympathetic sources. The nerves which enter the salivary glands follow the blood vessels. They break up into smaller bundles and form a plexus near the terminal parenchyma. Nerve plexus consists of unmyelinated axons which are embedded in the Schwann's cell cytoplasm. From here the nerves are distributed to the muscles of arterioles, to the secretory cells of the terminal end piece, to myoepithelial cells and to cells of intercalated and striated ducts.

In the parenchyma, two different morphologic patterns of innervation occur. In the first, epilemmal or subepithelial type, axons remain in connective tissue separated from the secretory cells by the basement membrane. It loses its Schwann's cell covering while approximating a secretory cell. When a nerve impulse occurs, a neurotransmitter is released and it diffuses approximately 100 to 200 nanometers across the basement membrane. This space is present between the axon and the secretory cells. In the second, hypolemmal or intraepithelial type nerve axons penetrate the basement

membrane. They lose their Schwann's cell covering and run between the secretory cells. A gap of 10 to 20 nanometers exists between the axon and the secretory cells (Fig. 11.10).

Both divisions of the autonomic nervous system may participate in the innervation of the secretory cells. Glands respond to sympathetic and parasympathetic stimulation by the change in their membrane potential. Large amount of watery saliva is secreted in response to parasympathetic stimulation while thicker, high in organic content and less in quantity of saliva is produced by sympathetic stimulation. Around the ducts, the cholinergic and adrenergic nerves are found in the connective tissue. Ductal system is responsive to autonomic stimulation or administration of autonomic drugs.

BLOOD SUPPLY

For rapid secretion of saliva an extensive blood supply is required. One or more arteries enter the gland and give rise to numerous arterioles. These branch into a dense distribution of capillaries. Large veins drain in to the periphery of the glands. Mainly the capillary network is found around the striated

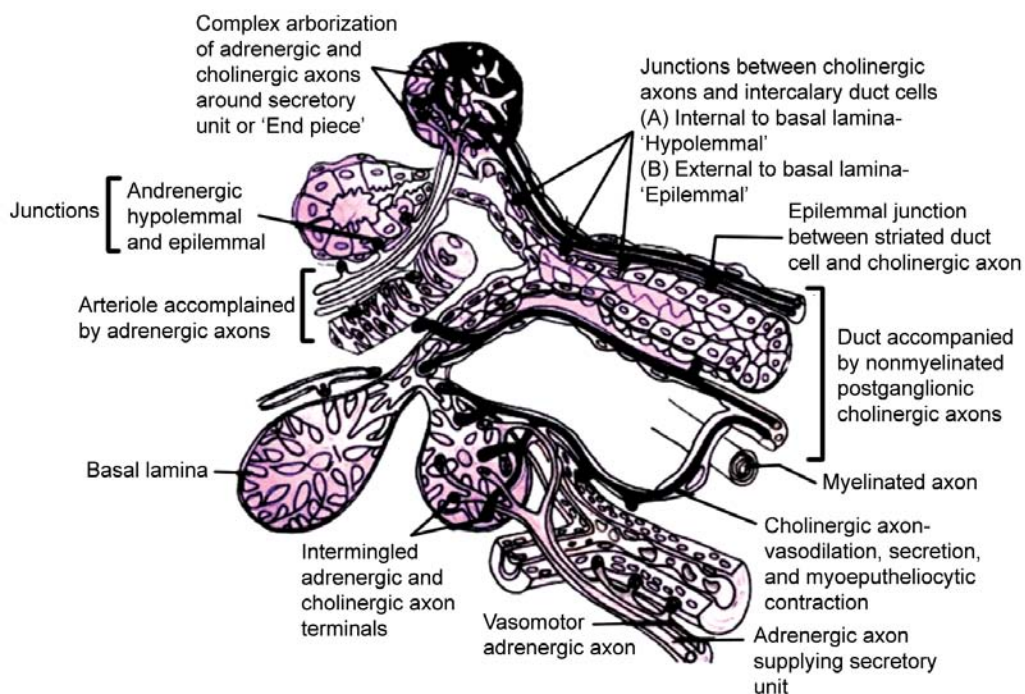


Figure 11.10: A generalized salivary gland showing innervation of the ducts, arterioles and secretory units

ducts, where ionic exchange occurs. With increase in salivary secretion, there is increase in blood flow of the gland.

CLASSIFICATION AND DESCRIPTION OF DIFFERENT SALIVARY GLANDS

Salivary glands are classified according to the following factors given in Table 11.2.

Table 11.2: Classification of salivary glands

- | |
|--|
| <p>A. According to size</p> <ol style="list-style-type: none"> i. Major (Fig. 11.1) <ol style="list-style-type: none"> a. Parotid, b. Submandibular, c. Sublingual ii. Minor (Glands of lips, cheek, hard and soft palate) <p>B. According to branching of ducts</p> <ol style="list-style-type: none"> i. Simple non-branching → minor glands ii. Compound racemose → major glands <p>C. According to damage of secreting cells</p> <ol style="list-style-type: none"> i. Apocrine glands—exhibit the type of glandular secretion in which the free end of the secreting cell is cast off along with the secretory products accumulated therein. ii. Holocrine glands—exhibit glandular secretion in which the entire secretory cell laden with its secretory products is cast off. iii. Merocrine glands—discharge only the secretory products and leave the secretory cell intact, e.g. salivary glands fall into this group. <p>D. According to secretion</p> <ol style="list-style-type: none"> i. Serous glands <ol style="list-style-type: none"> a. Parotid b. von Ebner ii. Mucous glands <ol style="list-style-type: none"> a. Palatine b. Posterior lingual c. Glossopalatine iii. Mixed glands <ol style="list-style-type: none"> a. Submandibular (mainly serous) b. Anterior lingual (mainly mucous) c. Sublingual (mainly mucous) d. Buccal (predominantly mucous) e. Labial (mainly mucous) |
|--|

Major Salivary Glands

Parotid Gland

Parotid gland is a pure serous gland. It is the largest of all the major salivary glands having the shape of a flattened inverted three-sided pyramid. Parotid glands are located on the sides of the face in front of the ears. Their superficial portion lies in front of the external ear and deeper part fills the retromandibular fossa. A well-formed connective tissue encloses the parotid gland. Stensen's duct is the main secretory duct of the gland. The duct opens into the oral cavity, on the buccal mucosa opposite the maxillary

second molar. A small papilla is present at the opening of the duct. The gland is an irregular lobulated yellowish mass, which lies below the external acoustic meatus between the ramus of mandible and the sternocleidomastoid muscle. The dimensions of gland craniocaudally is 5.8 cm and ventrodorsally is 3.4 cm. The weight is between 14 and 28 gm. Dimensions of Stensen's duct are 4 to 6 cm in length and 5 mm in diameter. There are two types of flow rates in salivary glands, (a) Active or stimulated i.e. when chewing, (b) Resting or unstimulated i.e. when not chewing like during sleep etc. Active flow is many times more than resting. Parotids gland secrete about 35 percent of whole saliva and resting flow rate is 0.4 ml per minute. External carotid artery supply the blood to the gland. Lymphatic drainage occurs into the superficial and deep cervical lymph nodes.

Parasympathetic nerve supply is from ninth cranial nerve. Sympathetic innervation is by the postganglionic fibers from the superior cervical ganglion.

Histological Structure The end secreting acinar cells are mostly serous with few cells which are seromucous in nature. Secretory granules can be seen in well-stained and fixed sections.

There is presence of numerous intercalated ducts. Long and branched striated ducts are also numerous which stain pink with eosin dyes and appear as "pink necklaces" in stained sections in contrast to more deeply stained acinar cells. Many fat cells are present in the connective tissue septa. These increase in number with age. In histologic sections, these are seen as the empty spaces (Figs 11.11A and B).

Submandibular Gland

Submandibular gland is a mixed gland that is both serous and mucous but mainly serous. Main secretory duct of the submandibular gland is the 'Wharton's duct' which opens on the floor of the mouth at the 'caruncula sublingualis'. This gland is well-encapsulated and is of the size of a walnut. The gland is located in the submandibular triangle behind the free border of the mylohyoid muscle. Mylohyoid muscle divides the submandibular gland into a smaller superficial part and a larger deep part, which are continuous with each other at the posterior border of the mylohyoid. Due to the curved path of the duct of submandibular gland and a higher mineral content in the saliva secreted by it, salivary calculi may develop in the duct of the submandibular gland.

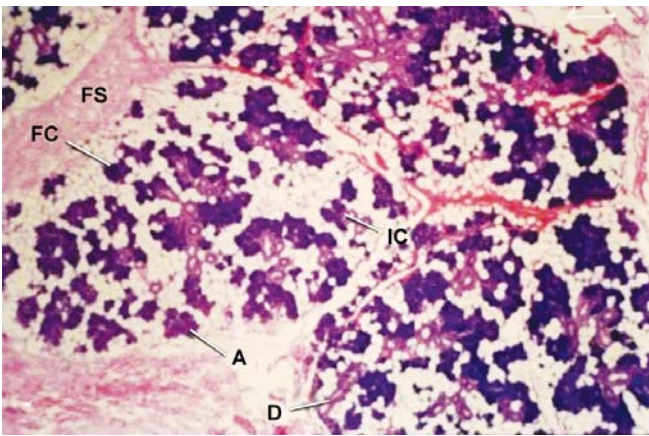


Figure 11.11A: Parotid gland: Low power view of parotid gland in adult. Lobular architecture of the gland is evident. Lobules are divided by fibrous septae (FS). Lobules consist of groups of serous acini (A) separated from each other by fat cells (FC). Many intercalated ducts (IC) are also present x 30 (D) Striated duct

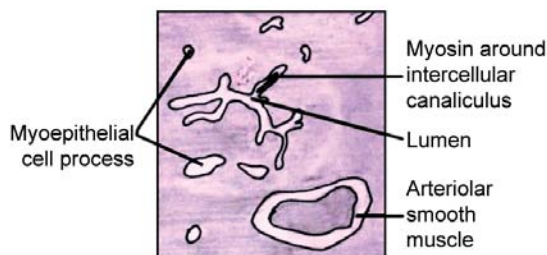


Figure 11.11B: Parotid gland showing myosin in apical cytoplasm of acinar cells, outlining lumen and intercellular canaliculi, cut in cross section and in myoepithelial cell process. (as seen fluorescence microscope)

Submandibular glands secrete about 60 percent of whole saliva and resting flow rate is 0.1 ml per minute. Lingual and facial arteries supply the blood. Parasympathetic innervation is from the seventh cervical nerve. Lymphatic drainage is into the deep cervical and jugular chain of nodes.

Histologically, (Figs 11.12 and 11.13) submandibular gland can be differentiated from the parotid gland by (i) Mixed acini (about 80 percent serous and 20 percent mucus), (ii) Intercalated ducts are few and lesser-developed and striated ducts are longer and more developed as compared to the parotid glands. The demilunes of serous cells are present over the mucous terminal portion.

Sublingual Glands

Sublingual glands are the smallest of all the major salivary glands. They are situated in the sublingual region that is

between the floor of the mouth and the mylohyoid muscle in contact with the sublingual fossa. It is a mixed gland but mainly mucous. It consists of one main gland and several smaller glands. Main secretory duct of the sublingual gland is called the 'Bartholins duct' which opens near the submandibular duct and eight to ten smaller duct openings are present separately along with the sublingual fold. Intercalated and striated ducts of sublingual glands are poorly developed and may be absent. Sublingual glands secrete about 5 percent of whole saliva. Sublingual and submental arteries supply the blood. Parasympathetic nerve supply is from seventh cranial nerve. Lymphatic drainage is into submandibular lymph nodes.

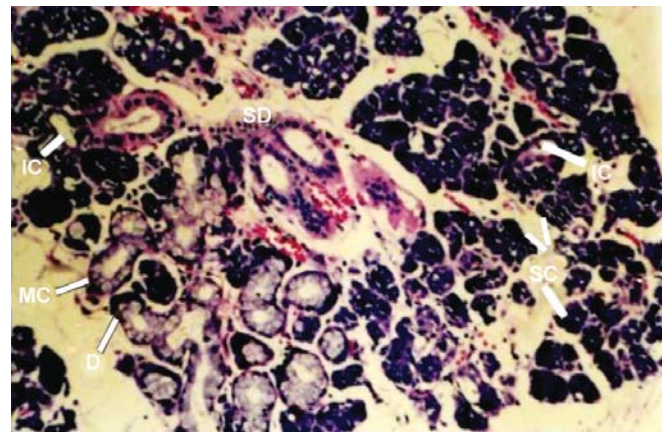


Figure 11.12: Submandibular gland shows presence of serous as well as mucus acini. Some mucous acini are covered with a cap of serous cells (SC) near the base called demilunes (D) x 150. (SC) Serous cells, (MC) Mucous cells, (IC) Intercalated ducts, (SD) Striated ducts

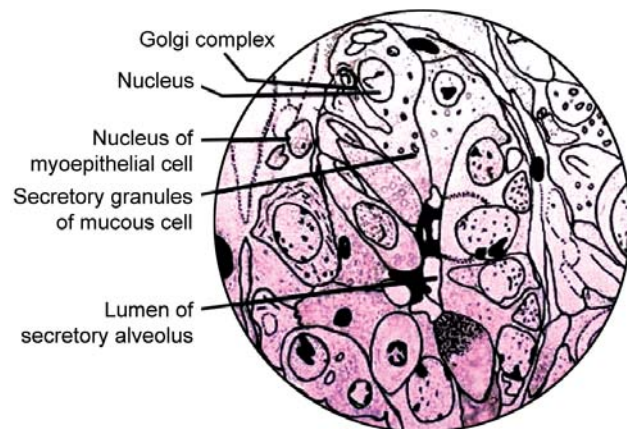


Figure 11.13: Transmission electron micrograph of developing submandibular gland (in fetus) showing mixed alveolus

Capsule is not well-developed and the connective tissue septa are prominent within the gland. Serous demilunes may be present at the blind end of the tubule. Sublingual glands are made up of mucin secreting acini, which are lined, by mucous cells. Nucleus of mucous cells is flattened or ovoid, compressed against the basal surface of the cell (Fig. 11.14). Cytoplasmic membrane looks distinct.

Minor Salivary Glands

Minor salivary glands are scattered and distributed in groups beneath the epithelium having very small and narrow ducts. These glands are present almost on all parts of the oral cavity. Separate capsule is not present on these glands. They get mixed with the connective tissue of the submucosa or muscle fibers of the tongue or cheek.

Labial and Buccal Glands

Labial and buccal glands are situated under the epithelium of lips and cheeks and are mixed in nature. These consist of mucous tubules with serous demi lunes. Intercalated ducts are variable in length.

Palatine Glands

Palatine glands are present in the glandular region of the hard palate, that is the posterolateral part of hard palate, soft palate and uvula. These are pure mucous glands. The openings of these ducts on the palatal mucosa are large and can be easily seen.

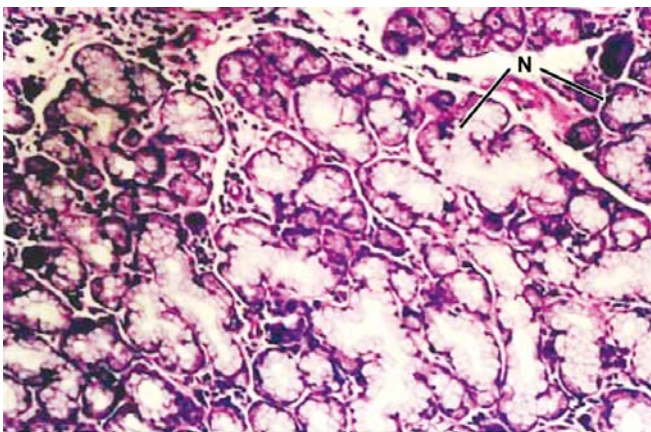


Figure 11.14: Mucous gland: Sublingual gland is made of mucin secreting acini which are lined by mucous cells. Nucleus of mucous cells is flattened or ovoid, compressed against the basal surface of the cell. Cytoplasm is empty looking. Cytoplasmic membranes are distinct x 150(N) Nuclei

Glands of Tongue

Several groups of the glands are present on the tongue.

Anterior glands Near the apex of the tongue anterior lingual glands are present. These are the glands of Blandin and Nuhn. Anteriorly the glands are mucous while posteriorly they are mixed in nature. The ducts of the glands open on the ventral surface of the tongue near the lingual frenum.

Posterior glands These are found in the posterior part of the tongue near the vallate papilla or posterior to the sulcus terminalis. These are purely mucous glands. Their ducts open into the dorsal surface of tongue.

von Ebner's gland These are purely serous glands opening into the trough of vallate papilla. The secretion of von Ebner's glands have specific lipolytic activity.

Lipolytic activity is very important when fat intake is high and levels of pancreatic lipase are low. Important functions of the secretion of von Ebner's gland are as follows.

1. Taste sensation-Washing out the trough of circumvallate papillae and make ready the taste receptors for new stimulus.
2. Digestive function- Antibacterial enzyme like peroxidase, lysozyme and lipolytic activity of secretion have digestive functions.
3. Protection – Secretion helps in the lubrication of the oral cavity.

AGE CHANGES IN SALIVARY GLANDS

Functioning of salivary glands reduces slowly with age, which may be due to the following histologic changes in the glands.

- i. Fibrosis
- ii. Fatty changes
- iii. Alteration in mitochondrial structure
- iv. Accumulation of lymphocytes.

DEVELOPMENT OF SALIVARY GLANDS

All salivary glands develop by the same process. The formation of the salivary gland initiates with the growth of the epithelial bud or cord of cells from the oral epithelium into the underlying ectomesenchyme. The growth of the bud is stimulated by the underlying mesenchyme. This cord of cells branches several times

and extends deeply into the ectomesenchyme. Ductal system and terminal secretory end pieces are developed by degeneration of their control cells. The elements formed from such an epithelial ingrowth form the parenchyma of a salivary gland. The ectomesenchyme forms the connective tissue component of the glands, which supports the parenchyma. It consists of septa and fibrous capsule. The primordia or buds of different glands appear at different periods.

- Buds of parotid and submandibular glands appear at fifth to sixth weeks of intrauterine life whereas primodium of sublingual glands appears after seven to eight weeks of intrauterine life. Minor salivary glands start developing at the third month (twelfth week) of intrauterine life.
- Epithelial buds develop into chord and chord grows into mesenchyme (Figs 11.15A and B). Chords divide and form a branched network which is solid earlier but gradually a lumen develops and changes into ducts (Fig 11.15C).
- Secretory portion develops later by the further budding and branching of chord cells.
- The connective tissue or ectomesenchyme surrounding the chord cells form the capsule around the developing gland parenchymatous structure. Capsule is the last component of the gland to differentiate. Septa divide the gland into lobes and lobules (Fig. 11.15D).
- The process of branching and the formation of hollow, tubular glands (Figs 10.15E and F), is because of the presence of the microfilaments in the epithelium cell.
- As arteries and collecting veins are formed, branches from the parasympathetic and the sympathetic nerves migrate into the gland.
- Increased stimulation can cause an increase in size of the gland.

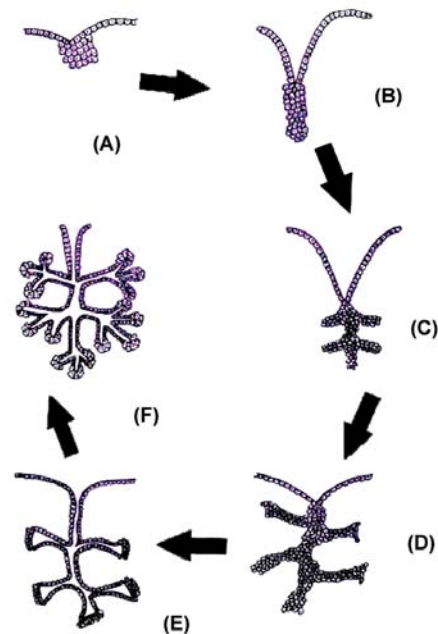
COMPOSITION OF SALIVA

Saliva consists of 99 percent or more of water.

Organic Constituents

Organic constituents of saliva are as follows.

- Enzymes-Amylase, ribonuclease, kallikrein, esterase, hystatin, cystatin, peroxidase, lysozymes, lactoferrin, acid phosphatase etc.
- Immunoglobulins-Ig G and Ig M.
- Other factors like blood clotting factors, aminoacids, urea, uric acid, glucose are also present.



Figures 11.15A to F: Stages in the development of salivary glands

- Formation of bud
- Formation of chord
- Branching of chords
- Formation of lobule
- Canalization of chords
- Cytodifferentiation.

Electrolytes

Important electrolytes of saliva are Na, K, Ca, Cl, HCO₃ and HPO₄. Other electrolytes like Mg, SO₄, F, SCN and I are also present in smaller concentration.

Saliva also contains squamated oral epithelial cells, microorganisms, leukocytes, serum constituents, food remnants and fluid from gingival crevice. pH of whole saliva is 6.4 to 7.4. Total volume of saliva is 1000 to 1500 ml daily. Resting flow rate of whole saliva is 0.2 to 0.4 ml per minute.

CLINICAL CONSIDERATIONS

Importance of Brushing Teeth at Night before Sleeping

On an average out of 700 milliliters of saliva produced daily only a few milliliters are produced at night. Due to decreased production of saliva and increase in its viscosity, food accumulation takes place rapidly at night, which increases the rate of tooth decay. Availability and the quantity of saliva has a profound effect on the incidence of caries.

This is the most important reason to brush after a meal at night before going to bed.

Parotid Enlargement in Systemic Conditions

Under the following chronic systemic conditions parotid enlargement takes place.

- i. Alcoholism
- ii. Protein deficiency
- iii. Diabetes mellitus
- iv. Chronic liver disease
- v. Starvation

These may sometimes be associated with parotid gland diseases.

Xerostomia

Certain inflammatory and neoplastic conditions, irradiation effects or the use of chemotherapy may reduce the flow of saliva from major or minor salivary glands, leading to dryness of mouth. Drugs like anticholinergics, antidepressants, antipsychotics, antihypertensives and anorectics can cause the dry mouth. Flushing action of saliva on food particles is reduced causing their accumulation and increase in dental caries. Inflammation and infection of oral mucosa occurs when salivary flow is decreased. Decreased salivary secretion cause painful swallowing, difficulty in speech, mastication and taste perception.

Formation of Salivary Calculi

Salivary calculi are the formation of stones in the salivary duct, resulting in reduced or stopped flow of saliva. This leads to inflammation of glands, pain and ultimately causes its atrophy. Due to curvature in the submandibular salivary duct and increased mineral content of its secretion, salivary calculi may easily form in the submandibular gland (Fig. 11.16).

Atrophy of Salivary Glands

With increase in age, atrophy of salivary glands occurs. The decreasing order of resistance to atrophy is in sublingual, submandibular and parotid glands, that is, sublingual gland is the most resistant and parotid is the least resistant to atrophy. Salivary glands are present almost everywhere in the oral cavity except most



Figure 11.16: Oval-shaped calculus or stone removed from the secretory duct of submandibular gland

anterior portions of the hard palate. Developmental coincidences may occur. In the mandible, this occurs posterior to the third molar teeth. In the maxilla, salivary glands may be present in the nasopalatine canal.

Lesions of salivary glands can occur almost anywhere in the mouth. So while differentiating any lesion of the oral cavity, origin of salivary glands should be checked.

In an autoimmune disease, e.g. Sjögren's syndrome, there is reduction of the salivary flow. Ulceration and oral infections are common features of these patients.

Mucocoele, a common surface lesion of the oral mucosa is a vesicular elevation. There is severance of the ducts of minor salivary glands resulting in pooling of the saliva in the tissues. If blockage of salivary gland duct occurs in major salivary glands by a calcified plug or mucous plug, it causes pain and requires surgical treatment.

Some drugs like, barbiturates, tranquilizers and antihistamines reduce the salivary flow. Hence, such drugs should be avoided in patients who have deficient salivary flow. Otherwise it may result in xerostomia.

SALIVA AS DIAGNOSTIC HELP

Saliva helps in diagnosis in the following ways.

- a. Blood type of individuals can be determined from salivary samples.
- b. Levels of hormones can be monitored from salivary secretions.

- c. Information about fetal growth can be obtained from the levels of estrion and estradiol hormones in saliva.
- d. Concentration of salivary electrolytes may aid in the diagnosis of systemic diseases like, Addison's disease and Cushing's syndrome.

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Eruption of Teeth and Physiologic Teeth Movements

- Introduction
 - Physiologic tooth movements
- Pattern and histology of various stages of eruption
 - Posteruptive stage
- Mechanism of tooth movements
 - Bone remodeling
 - Growth of root
 - Hydrostatic pressure
 - Periodontal ligament traction theory
- Cellular and molecular events in the eruption of tooth
- The important events in the eruption of teeth
- Clinical considerations
 - Deficiency of vitamin C
 - Orthodontic tooth movement
 - Teething problems
 - Abnormal tooth movements and eruption

INTRODUCTION

The jaws of an infant can only accommodate small teeth. These teeth, once formed cannot increase in size. But the jaw bones continually grow in size from infancy to childhood to adulthood. The teeth which erupt in infancy and childhood are too few and too small to serve the human masticatory needs in adult life. Therefore, the larger jaws of adults need not only more, but also bigger teeth. This is accomplished in humans by having two set of dentitions. The first is known as the primary or deciduous dentition and second as the permanent or secondary dentition (Fig.12.1).

For teeth to become functional, considerable movement is needed to bring them into occlusal plane. These movements are complex and are described under the following headings.

- a. Pre-eruptive tooth movement
- b. Eruptive tooth movement
- c. Posteruptive tooth movement

Tooth eruption can be defined as a physiologic process of the axial or occlusal movement of the tooth within the jaw from its developmental position to its functional position in the occlusal plane, or its clinical position.

It means that eruption is only a part of the total pattern of physiologic tooth movement. This tooth movement occurs because teeth undergo complex movements in order to maintain their position in the growing jaw and compensate for masticatory wear.

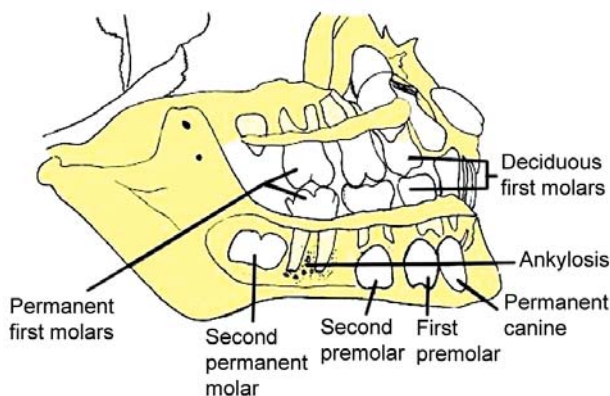


Figure 12.1: Mixed dentition present in a dried skull of seven years old child. The outer cortical plate is cut to show the developing permanent teeth

Physiologic Tooth Movements

These are of various types:

1. Axial or vertical movement - Primary eruption movement along the axis of tooth.
2. Drifting -It is a bodily movement in a direction perpendicular to the long axis of tooth like.
 - A. Mesial drift
 - B. Distal drift
3. Torsion - It is a rotatory movement around the longitudinal axis of tooth.
4. Tipping - Movement around the transverse axis of tooth is called tipping.

PATTERN AND HISTOLOGY OF VARIOUS STAGES OF ERUPTION

Pre-eruptive Tooth Movements

These are the movements made by both the deciduous and permanent tooth germs within the tissues of jaws before they began to erupt.

Pattern of Tooth Movements

These movements occur in association with growth of the jaws, and help in placement of the tooth germs at the correct positions of eruption.

Deciduous tooth germs during the early stage of differentiation are very small and a lot of space is present between them. This space is utilized with the growth of tooth germs, which is more than the growth of bone and thus, crowding occurs in the incisors and canine region. With the further growth of jaws, crowding is relieved, which allows the anterior germs to move forward and the deciduous molar germs to move backward.

Together with the movement at its own place, tooth germ also moves bodily outward, upward or downward with the growth of jaws in length, width and height.

The permanent teeth with their deciduous predecessors apart from moving about in their own bony crypt also show bodily movement before they reach the position from which they will erupt (Fig.12.1). In the first 25 months there is a lot of change in position between the permanent incisor tooth germ and its deciduous predecessor. These tooth germs develop on the lingual aspect of their deciduous predecessors in the same bony crypt. As jaws develop, the tooth germ shifts from this position. So the incisors and canines occupy a position in their own bony

crypts, on the lingual side of the roots of their deciduous predecessors (Figs 12.2A to G and 12.3). These changes in the relative position are basically because of growth of the permanent tooth and eruptive movement of the deciduous tooth. The same holds true for the permanent molars. The developing premolars are also present in their own bony crypt, but are eventually present in between the divergent roots of deciduous molars (Fig.12.4). The succession permanent teeth first occupy the same bony crypt as the deciduous teeth, but later occupy their own bony crypt.

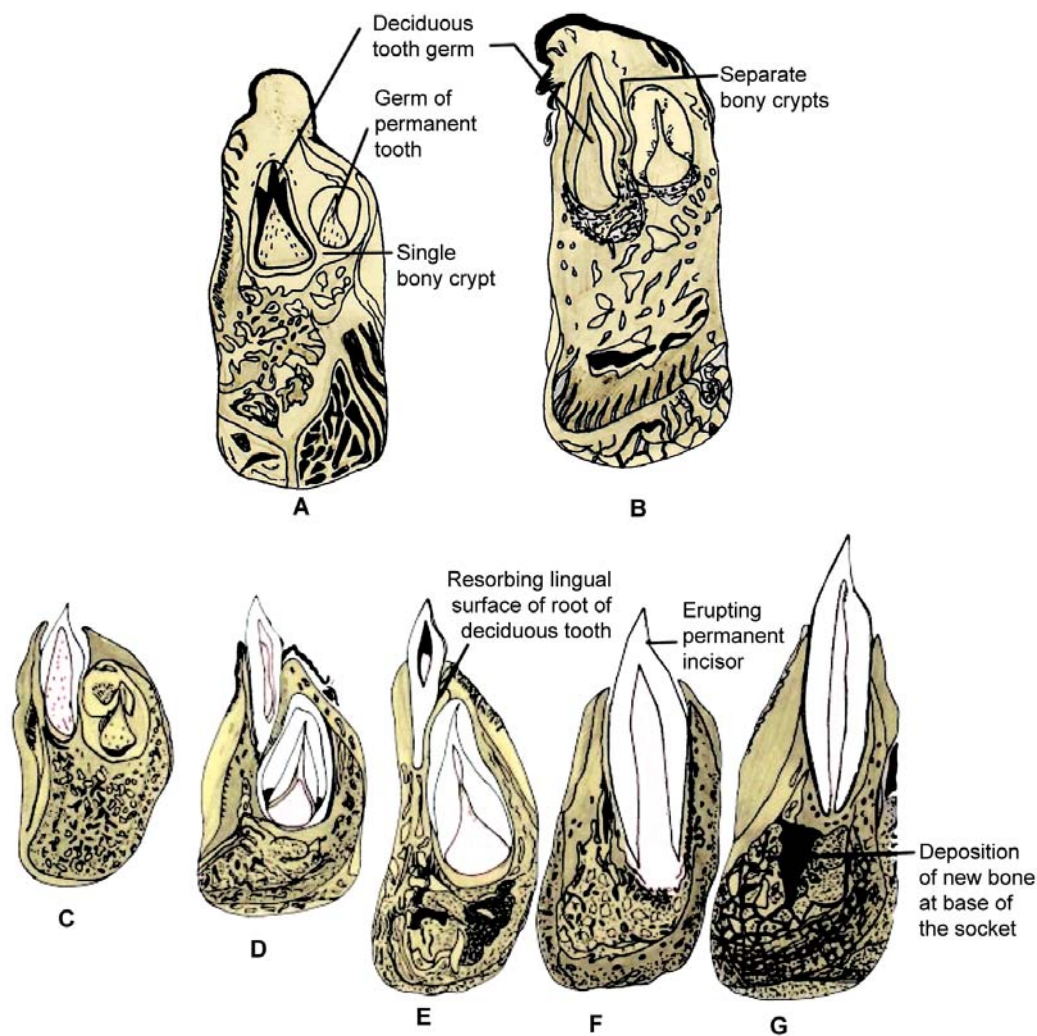
The permanent teeth with no deciduous predecessors, like permanent molars, during the early stage of their development have very little space to develop, so that the

maxillary permanent molars which develop in the maxillary tuberosity first develop with their occlusal surface facing distally and the mandibular molars develop with their occlusal surface facing mesially. The molars swing and become upright with the growth of the maxillary and mandibular jaws when space becomes available for the growing teeth.

The bony crypt remodels according to the movements of the developing tooth, thus pre-eruptive tooth movements can be described as movements to adjust and position the tooth with its crypt within the growing jaw.

These pre-eruptive movements of both deciduous and permanent tooth germs are a combination of two factors.

- i. Total bodily movement of the germs.



Figures 12.2 A to G: Labio-lingual sections of mandible through central incisors. (A) At birth (B) At the age of four months (C) Ten months (D) Twenty-five months (E) Fifty-five months (F) seventy-five months (G) One hundred and ten months

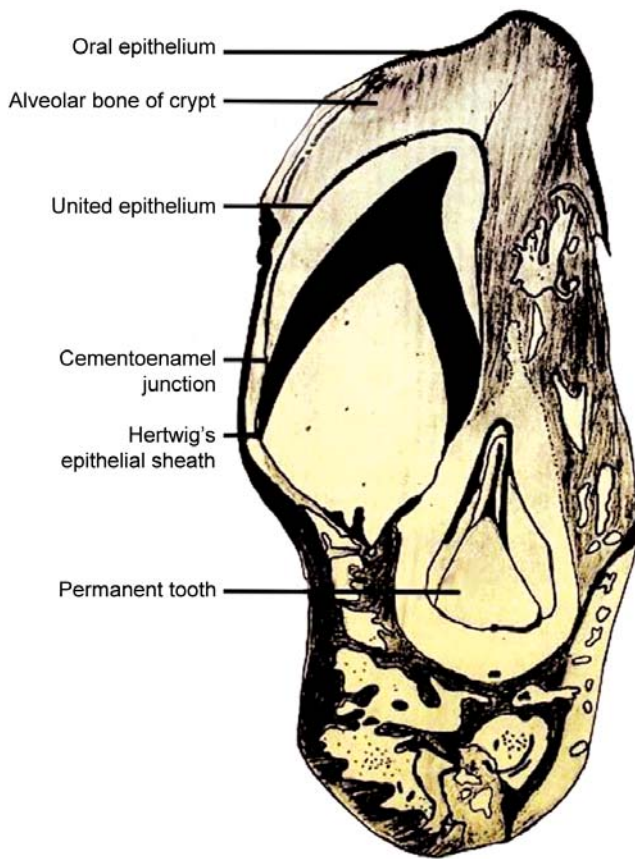


Figure 12.3: Unerupted deciduous canine and its permanent successor located lingually and apically to it, in a section (bucco-lingual) of mandible at the age of six months

- ii. Eccentric growth, in which one part of the tooth germ remains fixed while the rest continues to grow, causing a shift in the center of the tooth germ.

Histology of Tooth Movements during Pre-eruptive Phase

Histologically, pre-eruptive movements are reflected in the patterns of bone remodeling within the crypt wall. This occurs by selective deposition and removal of bone by osteoblastic and osteoclastic activity. As for example, during bodily movement in a mesial direction, bone resorption occurs on the mesial surface of crypt wall, and bone deposition occurs on the distal surface as part of a "filling-in" process (Fig.12.5). But during eccentric growth, only bone resorption occurs, which changes the shape of the crypt to accommodate the changing shape of tooth germ.

Marrow spaces develop in the bone of appendicular skeleton; bones grow in length and width by balanced resorption and deposition. Therefore, bones can be considered as moving in three-dimensional space.

It has been proved experimentally that physiologic activity of bone remodeling requires the presence of dental follicle and also, if the developing tooth germ is removed but follicle remains, the eruptive pathway through the bone will form, and if the follicle is removed, there will be no eruption, whether or not the tooth germ is present.

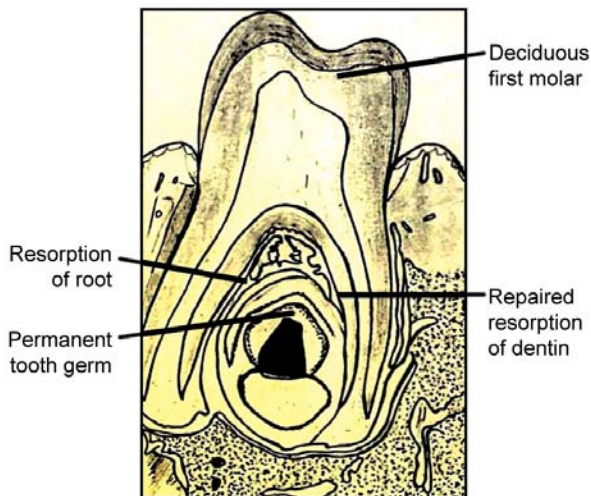


Figure 12.4: Deciduous first molar showing germ of permanent first premolar between its roots at the age of about two years (seen after removing buccal plate of mandible)

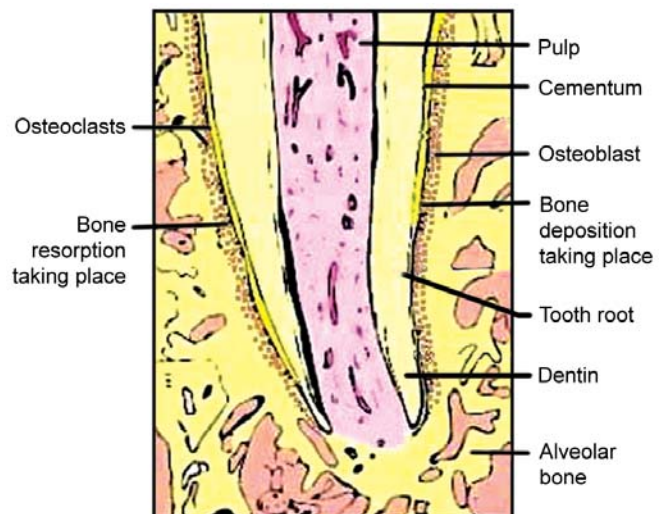


Figure 12.5: Bone remodeling within the crypt wall during physiologic tooth movement. At right, osteoblasts are present for bone deposition and are seen as a continuous row of minute spots. At left, osteoclasts are present for bone resorption and are seen as dark dots

Eruptive Tooth Movements

Pattern

These are the axial or occlusal movement of teeth from their developmental position in the jaw to their functional position in the occlusal plane. The actual eruption of the tooth, when it comes out of the gum is only one phase of eruption (Fig.12.6). During eruptive tooth movement, the jaw growth is still occurring and most of the teeth are erupting, so any movement other than axial dominate the eruptive movement.

Histology of eruptive movements

This phase is associated with several important histologic changes associated with tooth which are the following:

- Root formation
- Formation of periodontal ligament
- Formation of dentogingival junction

Root Formation

Eruptive tooth movement begins nearly with the onset of root formation, which is initiated by growth of the Hertwig's epithelial root sheath. After this, the differentiation of odontoblasts from dental papilla takes place. The odontoblasts then form root dentin so there is overall increase in length of the tooth.

The developing root initially grows towards the floor of bony crypt. This causes resorption of bone in this

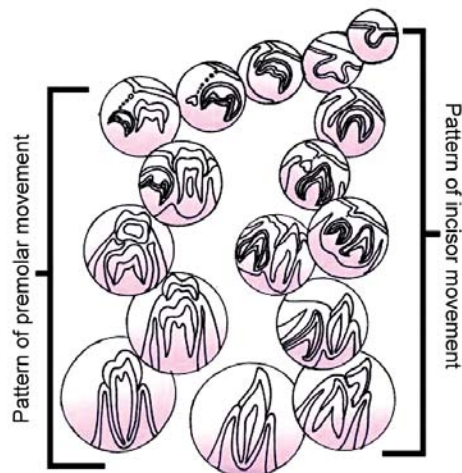


Figure 12.6: Summary of physiologic tooth movements: Pre-eruptive and eruptive, along with pattern of tooth resorption. The left stream shows pattern of premolar movement whereas the right stream shows pattern of incisor movement

location to provide room for the advancing root tip. But with the beginning of eruptive tooth movement (possibly coincident with periodontal ligament formation), space is created for the forming root, and resorption no longer occurs on the floor of crypt. In some cases, the distance moved by tooth exceeds the rate of root formation and bone deposition occurring on the crypt floor.

As root formation continues, important changes associated with the development of supporting apparatus of the tooth occur in the dental follicle. The changes, which lag behind root formation are the following.

- Cementum deposition on the newly formed root surface.
- Bone deposition on the crypt wall.
- Organization of a periodontal ligament from the dental follicle.

Formation of Periodontal Ligament

Periodontium consists of cementum, periodontal fibers and bone forming the crypt.

The periodontal ligament forms only after root formation has been initiated. Once the periodontal ligament has been established, it must be remodeled to allow continued eruptive tooth movement. A number of structural changes are seen within the periodontal ligament, which may be responsible for tooth movement.

There are many histologic features of the periodontal ligament, which help in explaining eruptive tooth movements:

- Active ingestion and synthesis of collagen by fibroblasts allows remodeling of the principal fiber bundles of the periodontal ligament.
- Presence of complex intracellular system of tubules and filaments, the contractile elements, some of which help the cell to contract.
- Occurrence of cell to cell contacts of the adherence type between the periodontal ligament fibroblasts.
- Occurrence of a structure called the fibronexus. The fibronexus has a morphologic relationship between intracellular microfilament of the fibroblast, region of dense cell membrane, extracellular filaments and fibronectin. Fibronectin is a sticky glycoprotein, which sticks to a number of extracellular components including collagen (Fig.12.7).

Formation of the Dentogingival Junction

Formation of the dentogingival junction has already been explained in Chapter 9, hence, only a brief outline is given here.

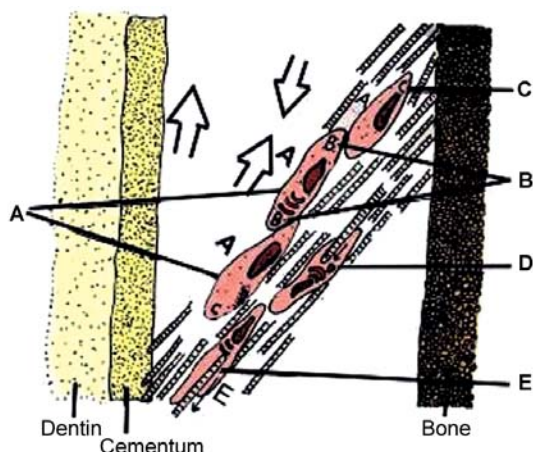


Figure 12.7: Structures present in periodontal ligament to cause tooth movement. (A) Fibroblasts (B) Adherence type intercellular junctions (C) Fibronexus, attached to collagen by fibronectin. They transmit the force to the collagen fiber bundle on contraction of fibroblasts (D) Deposition of collagen (E) Resorption of collagen

A number of changes occur in the tissues, which cover the erupting tooth that is in the intervening connective tissue present between the reduced enamel epithelium covering the crown of the tooth and the overlying oral epithelium. Because of this loss the two epithelia proliferate and form a solid plug of cells in advance of the erupting tooth. The central cells of this epithelial cell mass degenerate and form a canal lined with epithelium through which the tooth erupts without any hemorrhage. This epithelial cell mass is also involved in the formation of the dentogingival junction.

Rate of tooth eruption

Once the tooth is visible into oral cavity, it continues to erupt at a rate of about 1 mm every one to two months and this rate slows only when the erupting tooth comes in contact with its antagonist of opposite arch. At this time the rapid eruptive movement stops, but root formation is not yet complete. As further occlusal tooth movement is prevented additional root growth is accommodated by removal of bone on the socket floor.

Determination of final position of tooth in oral cavity

This is determined by the environmental factors of oral cavity. As soon as the teeth erupt in the oral cavity, they encounter forces of muscle pull from all sides by tongue, cheek and lips. These muscle forces determine the final position of tooth. Some habits like tongue thrusting, thumb sucking or mouth breathing may disturb the muscle

balance and cause the tooth to finally erupt in an abnormal direction and position. A sustained muscle force of 4 to 5 grams is enough to move a tooth. The above description holds true for all teeth. But successional teeth possess an additional anatomic feature called the gubernacular canal and its content the gubernacular cord, which might influence the eruptive tooth movement.

With the eruption of deciduous teeth the permanent tooth germ (which earlier shared a common crypt with the deciduous tooth) shown in figure (12.2A) acquires its own bony crypt and is completely covered by bone, except for a narrow and small canal called the gubernacular canal which is filled with connective tissue and often contains epithelial remnants of the dental lamina. This connective tissue mass is called the "gubernacular chord." This guides the permanent tooth as it erupts in the oral cavity. In the dried skull, holes of gubernacular canals can be seen in the jaws on the lingual aspect of deciduous teeth.

After removal of the bone overlying the developing tooth germ, there is a loss of soft connective tissue between the reduced enamel epithelium covering the crown of tooth and the overlying oral epithelium. This happens because the erupting tooth causes local ischemia and necrosis.

Posteruptive Stage

Prefunctional eruptive tooth movement: The movement of the tooth after its appearance in the oral cavity till it attains the functional position is known as prefunctional eruptive tooth movement.

Pattern

Posteruptive stage refers to the movements made by the tooth generally after the age of 14 to 15 years, that is, the period after the tooth has acquired its functional position.

Teeth are required to perform these movements for two main reasons, these are as follows.

- A. To accommodate for the continuously growing jaw and maintain position of the erupted tooth, which usually occurs upto the age of 18 years.
- B. For occlusal and interproximal wear, which occurs continuously.

The first movement mainly occurs in an axial direction to keep pace with an increase in height of the jaws. It involves both the tooth and its socket and stops when jaw growth is completed. The movement which compensates

for occlusal and proximal wear continues throughout life. It occurs by axial and mesial migration. Final position of the tooth in the oral cavity is determined by the following factors.

1. Pressure exerted by the tongue and cheeks.
2. Pressure exerted by the adjacent teeth.
3. Abnormal habits like tongue thrusting, thumb sucking and lip biting.

Histology

Tooth movement to accommodate condylar growth and growth of jaws: Between 14 to 18 years of life, rapid condylar growth takes place, which separates the jaws and teeth, creating space for further eruptive movements and adjustment of position of tooth socket.

Remodeling of socket takes place by new bone formation at alveolar crest and at the base of socket to increase the alveolar bone height. This increase in bone height causes the tooth to move away 2 to 3 mm coronally in an axial direction from the base of socket (Fig.12.8). However, this bone remodeling is not responsible for tooth movement. This posteruptive tooth movement maintains the position of erupted tooth with the growth of the jaws.

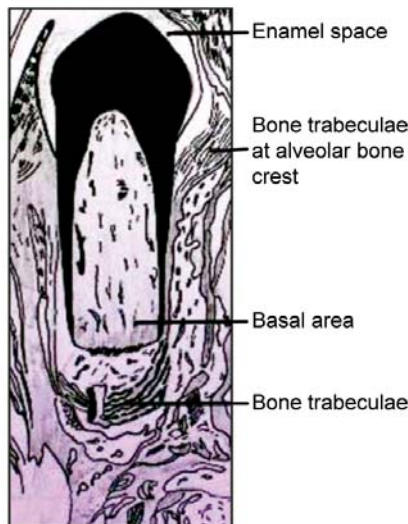


Figure 12.8: Erupting mandibular deciduous canine

Movements to compensate occlusal and interproximal wear: A tooth makes the axial movement to compensate for occlusal wear, which is achieved by the same mechanism as eruptive tooth movement. These axial posteruptive movements are made when the apices of the lower

permanent molars are formed completely and those of the second premolar and second molar are almost complete. This clearly indicates that root growth is not a factor responsible for axial eruptive tooth movement. It was thought that the continuous deposition of cementum around the apices of the roots of teeth is sufficient to compensate for occlusal wear. It lays stress on role of the periodontal ligament indicating that it is very likely that the forces causing tooth eruption are still available to bring about sufficient axial movement to compensate for occlusal wear. The axial movement at this time does not require bone remodeling. These are very slow movements made to adjust and replenish lost tooth height due to occlusal wear.

Interproximal wear occurs at the contact points of tooth slowly with age. The extent of this interproximal wear is considerable (more than 7 mm in the mandible). To compensate for this wear and maintenance of contact points, a phenomenon called mesial drift or proximal drift takes place. This occurs by resorption of bone mesially by osteoclasts and deposition of bone distally by osteoblasts and remodeling of collagenous periodontal and transseptal fibers.

MECHANISM OF TOOTH MOVEMENTS

There are certain factors, which are responsible for the eruption of teeth, though the exact factor responsible for eruption is still debatable.

Four important factors considered to be responsible for tooth movement are as follows:

1. Bone remodeling (deposition).
2. Root growth.
3. Hydrostatic vascular pressure.
4. Periodontal ligament traction.

Bone Remodeling

According to this factor, the selective resorption and deposition of bone takes place during both eruptive and pre-eruptive phases of tooth movement around tooth germs. Intense metabolic activity in alveolar bone, which takes place around the moving tooth requires the presence of dental follicle. Hence, dental follicle determines the position of tooth. Dental follicle has important role in coordinating bony changes around the erupting tooth. But it does not conclusively prove that only the follicle is responsible for tooth eruption. Bone remodeling around

the developing tooth germ carries it axially. This theory is still recognized, as the persons genetically lacking in osteoclasts have retarded tooth eruption.

Dental follicle is considered to have an important role in bone remodeling, as it is the stimulating factor for osteoblasts and osteoclasts. Absence of follicle hampers bone remodeling and stops tooth eruption. Many experiments have shown that axial movement of tooth towards the oral cavity is only possible when dental follicle is intact, no matter whether a tooth bud is present in the follicle or not. Dental follicle provides the source for new bone forming cells.

Growth of Root

Root growth theory considers growth in root length to be responsible for tooth eruption, as the growing root presses against the socket base and opposite force pushes the tooth to erupt. After crown formation, there is formation of the root. It involves cellular proliferation and formation of new tissues. These new tissues which are formed, are accommodated by either movement of crown or resorption at the base of the socket.

The basis of this theory is questionable due to certain facts :

- a. If root formation results in an eruptive force, there is the requirement of a fixed base but the bone at the base of root cannot act as a fixed base as pressure on the bone results in its resorption.
- b. Sometimes the distance travelled by an erupting tooth is more compared to the length of root formed during the same period.
- c. Rootless teeth also erupt for example natal teeth which are occasionally present in oral cavity at birth are always without root.
- d. Eruption can occur even after root formation is complete.
- e. Hammock ligament - this structure was considered to provide a fixed base for the growing root, so as to create an eruptive force and it was thought to be located below the root, like a sling attached to bone on all sides. But now, it is recognized that this structure either does not exist or it separates pulp from the dental sac and has no bony insertion.
- f. After resecting root, it prevents further root formation but eruptive tooth movement continues. Root growth theory is now considered to be unimportant for tooth eruption, but it is true that root growth is necessary for eruption as it gives attachment to periodontal fibers.

Hydrostatic Pressure

Hydrostatic pressure theory is based on the fact that dental papilla is highly vascular. Vascular (arterial) pressure in the blood vessels of papilla and fluid pressure due to retained water in periodontal ligament causes teeth to erupt. Teeth move in their socket according to arterial pressure. This local volume change produces limited movements. By retaining water, ground substance may also swell, so pressure is created. But such pressures are not primary in tooth movement because if root and periapical vasculature are removed surgically, tooth eruption is not prevented. This theory is not recommended now as in the absence of blood vessels (in periapical region) also, tooth eruption occurs.

Periodontal Ligament Traction Theory

Among all four theories periodontal ligament traction theory has strongest evidences in its favour. The periodontal ligament has an important role in maintaining tooth position. The transseptal fibers running between adjacent teeth across the alveolar process draw neighbouring teeth together and maintain them in contact. As long as the periodontal ligament is viable, tooth movement occurs.

The fibroblasts by virtue of their contractile forces exhibit fibronexus. It is through these fibronexus that the fibroblasts transmit forces to the collagen bundles. These collagen bundles apart from undergoing remodeling are also inclined at the correct angle to bring about eruptive movement. The orientation of the fiber bundles, which is established by the developing root, is a basic requirement for tooth movement. The follicle, before it becomes the periodontal ligament, also plays an important role in tooth eruption, although it does not provide the actual eruptive force.

Basically, eruptive movement is brought about by a combination of events involving a force initiated by the fibroblast. This force is transmitted to the extracellular compartment and to the collagen fiber bundles via fibronexus. These fiber bundles in proper alignment (brought about by root formation), bring about tooth movement by virtue of the ability to remodel. The removal of bone to create an eruptive pathway is also determined by the tissues.

It is concluded that the force for tooth eruption is most probably generated by the contractile property of fibroblast of ligament along with other conditions, which

must be present so that this contraction can be used for tooth eruption. Hence, eruption is a multifactorial phenomenon.

Mesial or Proximal Drift

Mesial or proximal drift involves a combination of two distinct forces resulting from occlusal contact of teeth and contraction of the transseptal ligaments between the teeth. When the jaws are clenched bringing teeth into contact, force is generated in a mesial direction as of the summation of cuspal planes as many teeth have a mesial inclination. This can be shown in a number of ways. When opposing teeth are removed, the rate of mesial drift is slowed, but not eliminated. These observations indicate that although an anterior component of occlusal force is responsible for mesial drift, it is not solely responsible.

Transseptal ligament runs across the alveolar process in between the teeth and it maintains the teeth in position. If a tooth is bisected, the two halves move away from each other, but if the transseptal ligament is previously removed, this separation does not occur. If the approximal contacts are disked to make room for mesial drift, the teeth begin moving to reestablish contact. But if the teeth are ground out of occlusal contact, the rate of drift is slowed. Therefore, it can be concluded that mesial drift is achieved by contraction of transseptal fibers and increased by occlusal forces.

The rate of eruption of permanent teeth is as follows (a) one micron per day when the tooth is in the bony crypt, (b) the eruption rate increases to 7.5 micron per day when the tooth comes out of bony socket and then (c) eruption rate may accelerate upto 11.11 to 16.67 micron per day (about one mm in every one to two months or 500 micron to 1 mm per month) when the tooth appear in the oral cavity and yet to occlude with opponent teeth. In anterior teeth eruption rate is slightly higher than in posterior teeth. In deciduous teeth eruption rate is about double the permanent teeth.

CELLULAR AND MOLECULAR EVENTS IN THE ERUPTION OF TOOTH

Eruption of tooth is a localized genetically controlled and programmed event. Genes related with the eruption are in dental follicle. Series of signaling interactions between the cells of bony crypt like osteoblasts and osteoclasts and

cells of dental follicle are involved and are regulated by the genes. These are (a) the recruitment of the mononuclear cells to the dental follicle, (b) its differentiation into osteoclast (c) activation of osteoclast (d) bone resorption at the coronal half of the dental follicle and (e) bone formation at the basal end.

THE IMPORTANT EVENTS IN THE ERUPTION OF TEETH

Dental follicle serves as a target tissue for mononuclear cells and regulate the cellular events of the eruption. Tooth eruption is regulated by various molecular events. These molecules having similar and overlapping functions. Important factors which increase the rate of eruption are CSF-1, MCP-1, TGF α and EGF.

Enamel organ function as a biological clock and regulates the timing of the tooth eruption. The cellular events of tooth eruption consist of complex interrelationship between the various factors.

CLINICAL CONSIDERATIONS

Deficiency of Vitamin C

Vitamin C is necessary for the formation of collagen fibers. It causes cross-linkages to occur between collagen molecules. In deficiency of vitamin C, collagen fibers are, poorly formed in periodontal ligament and tooth fails to erupt or erupts slowly and very late.

Orthodontic Tooth Movement

The plasticity of the alveolar bone allows it to react favorably or unfavorably to its surrounding environment. This plasticity of alveolar bone allows it to get resorbed and deposited even when very light forces are applied. This plasticity of bone and its characteristic to remodel is used by the orthodontist to change the position of teeth and alignment of dental arches.

Teething Problems

Teething problems are most common problem faced during eruption of teeth in children when teeth tear the gum and just erupt and appear into the oral cavity. The gum adjacent to the just erupted teeth shows acute inflammatory reaction and pain, and child may also show systemic manifestations as general malaise, fever and sometimes diarrhea. These problems faced by children during tooth eruption are called Teething problems.

Abnormal Tooth Movements and Eruption

Table 12.1 gives the time of tooth emergence; it is important to note that there is a considerable variation in these timings. However, only teeth emerging significantly outside these ranges should be considered abnormal and indicative of some fault in eruptive movement. Most of the time, the greatest number of aberrations in eruption timings are delayed eruptive movements (Tables 12.1 and 12.2).

Premature Eruption (Occurs Infrequently)

Deciduous teeth start appearing in the oral cavity at the age of about 6 to 6½ months but sometimes, very rarely teeth appear very early in postnatal life or at times they are even present at birth. Teeth, which are associated most frequently with premature eruption, are predeciduous lower central incisors.

A. *Natal teeth*: These are predeciduous teeth, hornified epithelial structures without roots present in infants even at the time of birth.

B. *Neonatal teeth*: These teeth erupt in first thirty days of postnatal life.

Premature eruption of permanent teeth usually is a result of premature loss of deciduous teeth.

Delayed Eruption (More Common)

Systemic conditions like rickets, cretinism, and cleidocranial dysplasia are associated with delayed eruption of teeth. Sometimes, local factors work as a barrier in the path of erupting tooth and prevent its eruption. Such local factors may be retained deciduous tooth or fibrosis of gingiva which prevents eruption of tooth or delays it.

Embedded or Impacted Teeth

Embedded teeth are unerupted because of lack of or insufficient eruptive forces. Impacted teeth are unerupted due to lack of space to erupt, some physical barrier in the path of eruption; barrier may be bone or adjacent teeth. Mandibular third molar are most commonly impacted

Table 12.1: Chronology of deciduous dentition

Primary tooth	First evidence of calcification (weeks in utero)	Amount of enamel formed at birth	Enamel Completion (months after birth)	Eruption Mean age in months	Root completion (yrs)
Maxillary					
Central incisor	14 (12 to 16)	Five-sixths	1 ½	10 (8 to 12)	1 ½
Lateral incisor	16 (15 to 16 ½)	Two-thirds	2 ½	11 (9 to 13)	2
Canine	17 (15 to 18)	One-third	9	19 (16 to 22)	3 ¼
First molar	15 ½ (14 ½ to 17)	Cusps united, occlusal surface completely calcified plus a half to three-fourths crown height	6	16 (13 to 19) - Boys (14 to 19) - Girls	2 ½
Second molar	19 (16 to 23 ½)	Cusps united, Occlusal surface incompletely calcified tissue covers a fifth to a fourth crown height	11	29 (24 to 33)	3
Mandibular					
Central incisor	14 (13 to 16)	Three-fifths	2 ½	8 (6 to 10)	1 ½ yrs.
Lateral incisor	16 (15 to 17)	Three-fifths	3	13 (10 to 16) months	1 ½ yrs
Canine	17 (16 to 18)	One-third	9	20 (17 to 23) months	3 ¼ yrs.
First molar	15 ½ (14 ½ to 17)	Cusps united, occlusal surface completely calcified	5 ½	16 (14 to 18) months	2 ¼ yrs.
Second molar	18 (17 to 19 ½)	Cusps united, occlusal surface incompletely calcified	10	27 (23 to 31) Boys (24 to 30) months Girls	3 yrs.

Table 12.2: Chronology of permanent dentition

Tooth	First evidence of calcification	Enamel completed (in years)	Eruption (in years)	Touching tooth / teeth of opposite arch (in years)	Root Completed (in years)
MAXILLARY					
Central incisor	3 to 4 months	4 to 5	7 to 8	8 to 9	10
Lateral incisor	1 year	4 to 5	8 to 9	9 to 10	11
Canine	4 to 5 months	6 to 7	11 to 12	12 to 13	13 to 15
First premolar	1½ to 2 years	5 to 6	10 to 11	11 to 12	12 to 13
Second premolar	2 to 2½ years	6 to 7	10 to 12	10 to 12	12 to 14
First molar	At birth	2½ to 3	6 to 7	8 to 9	9 to 10
Second molar	2½ to 3 years	7 to 8	12 to 13	13 to 14	14 to 16
Third molar	7 to 9 years	12 to 16	17 to 21	18 to 23	18 to 25
MANDIBULAR					
Central incisor	3 to 4 months	4 to 5	6 to 7	7 to 8	9
Lateral incisor	3 to 4 months	4 to 5	7 to 8	8 to 9	10
Canine	4 to 5 months	6 to 7	9 to 10	10 to 11	12 to 14
First premolar	1½ to 2 years	5 to 6	10 to 12	12 to 13	12 to 13
Second premolar	2¼ to 2½ years	6 to 7	11 to 12	12 to 13	13 to 14
First premolar	At birth	2½ to 3	6 to 7	8 to 9	9 to 10
Second molar	2½ to 3 years	7 to 8	11 to 13	12 to 13	14 to 15
Third molar	8 to 10 years	12 to 16	17 to 21	18 to 23	18 to 25

teeth followed by maxillary third molars and maxillary canines.

Submerged Tooth

Submerged tooth is found in a condition in which tooth shows loss of periodontal ligament and the root gets directly attached to the bone and fuses with it. This condition is generally found in the deciduous mandibular second molar. These deciduous teeth do not exfoliate at their scheduled time and if situated between the two fully developed permanent teeth, may appear submerged between the two in the jaw.

Ankylosed Tooth

Ankylosis of teeth in permanent dentition generally occurs if deciduous teeth get severe trauma and underlying developing permanent dentition is affected by this trauma. A dental follicle may get destroyed and periodontal ligament formation is prevented and the developing tooth fuses directly with the jaw bone causing ankylosis (Fig. 12.9).

Premature Loss of Deciduous Tooth

Sometimes premature loss of deciduous tooth occurs. This may lead to early eruption of its successor. More

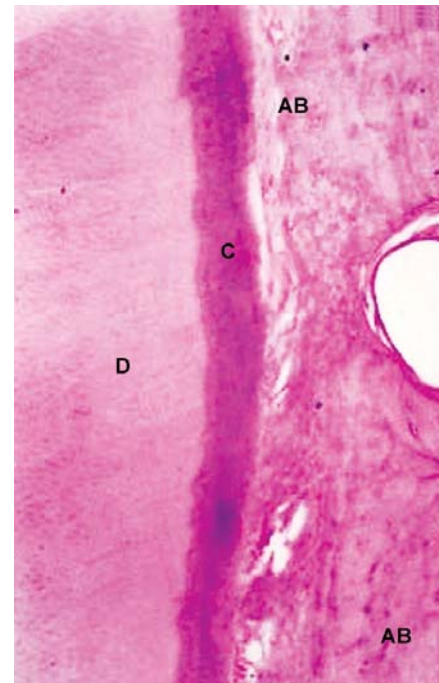


Figure 12.9: Ankylosis: The tooth is adherent to the bone. There is practically no soft tissue between the tooth and the bone of the socket. Cementum (C) is in direct contact with alveolar bone (AB) with no intervening periodontal ligament (D) Dentin (H and E x 200)

commonly delayed eruption occurs because of local or systemic factors. Systemic factors are nutrition, genetic and endocrine deficiencies.

Local factors are situations such as loss of deciduous tooth and drifting of opposing teeth to block the eruptive pathway.

Supraerupted or Overerupted Teeth

When opposing teeth are absent then teeth erupt beyond the occlusal plane and is known as supraerupted or overerupted teeth. In case of supraeruption, the gingival margin may follow the tooth or gingival margin may stay at original level and root gets exposed.

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Shedding of Deciduous Teeth

- Introduction
- Definition
- Pattern of root resorption
- Histology of resorption
 - ◻ Odontoclasts (dentinoclasts)
- Mechanism of resorption and shedding
- Clinical considerations
 - ◻ Remnants of deciduous teeth
 - ◻ Retained deciduous teeth
 - ◻ Submerged deciduous teeth



INTRODUCTION

The complete life-cycle of human beings consists of two types of dentition, first deciduous (primary) and then permanent. The pressure from the erupting successional permanent tooth helps to determine the pattern of deciduous tooth resorption. It means that the eruptive pathway of the permanent teeth is according to the shedding or exfoliation of the deciduous teeth. Because teeth, once formed cannot increase in size, so a second dentition, which consists of larger and more teeth, is required for the larger jaws of adult.

DEFINITION

The physiological process responsible for the removal of deciduous or primary teeth is known as shedding. The presence of two generations of teeth in a life-cycle have special importance, which are as follows:

1. The jaws of infants are smaller, thus they can bear only smaller teeth and also lesser in number, like the teeth of deciduous dentition. With development of the jaws, the teeth of deciduous dentition exfoliate and are taken over by teeth of permanent dentition which are larger and more in number. This mechanism can be understood as an adaptation to the growth of jaws.
2. Deciduous teeth provide space for the development of permanent teeth and guidance for their eruption. Deciduous teeth are present for about 8 years in human life. During this time they play a very important role in reserving space for the maintenance of proper alignment, spacing, and occlusion of permanent teeth. Early loss of deciduous teeth or infection around its root causes complications in the permanent tooth arch. In these conditions, permanent teeth may be either impacted, hypoplastic, stained or maloccluded.
3. Deciduous dentition helps in the development of speech in a growing child. Malaligned deciduous teeth may cause defective speech in a child, which may have a permanent impact on his or her speech clarity.
4. A developing child requires various types of nutritious meals for proper growth. With the emergence of deciduous teeth, child changes his/her diet from liquid to semisolid and then gradually to more solid food. Thus, deciduous teeth help in faster growth of child by maintaining proper diet and efficient mastication of food.

PATTERN OF ROOT RESORPTION

The word deciduous is taken from a latin word which means to 'fall off'. Thus, deciduous teeth can be compared to the leaves of a tree which fall off when the season goes. The shedding of deciduous teeth occurs as a result of gradual resorption of the roots of teeth and their supporting tissues, the periodontal ligament. The pattern of resorption of deciduous teeth is determined by the position of successional tooth in relation to its predecessor. The successional tooth in turn exerts pressure which is initially directed against the root surface of the deciduous tooth.

In case of the deciduous incisors and canines, resorption occurs on the lingual surfaces of their roots. This occurs as their successors develop lingually and erupt in an occlusal and vestibular direction. Exfoliation of these teeth occurs with most of their pulp chamber intact (Fig.13.1). Finally, the developing tooth germs occupy a position directly apical to the deciduous tooth, which allows them to erupt in a position occupied by the deciduous tooth (Fig.13.2). Sometimes, however, this apical positioning of the tooth germs does not occur and the permanent tooth erupts lingual to the still functioning deciduous tooth (Fig.13.3). Generally this occurs in case of permanent mandibular incisors.

The tooth germs of the developing premolars are found between the divergent flared roots of the deciduous molars (Fig.13.4). With the development of the successional tooth germ, the resorption begins on the inner surface of roots

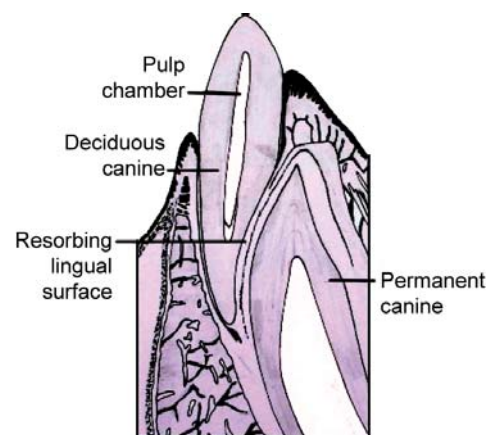


Figure 13.1: The deciduous and permanent canines in their relative position. The lingual aspect of the deciduous canine showing resorption before exfoliation with most of the part of the labial surface of the root being intact

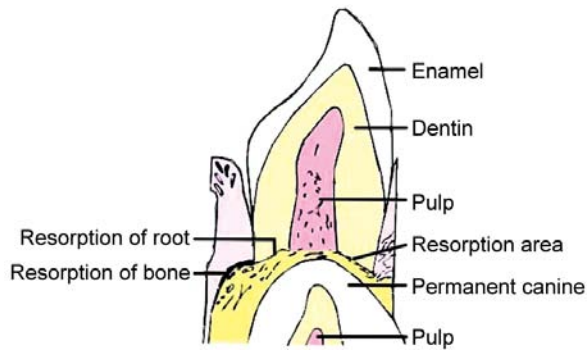


Figure 13.2: Root of deciduous canine showing resorption

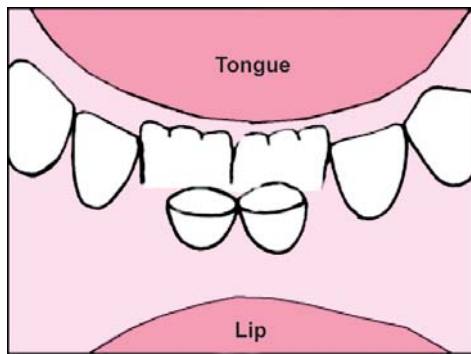


Figure 13.3: Eruption of permanent mandibular central incisors lingual to the deciduous incisors before their shedding

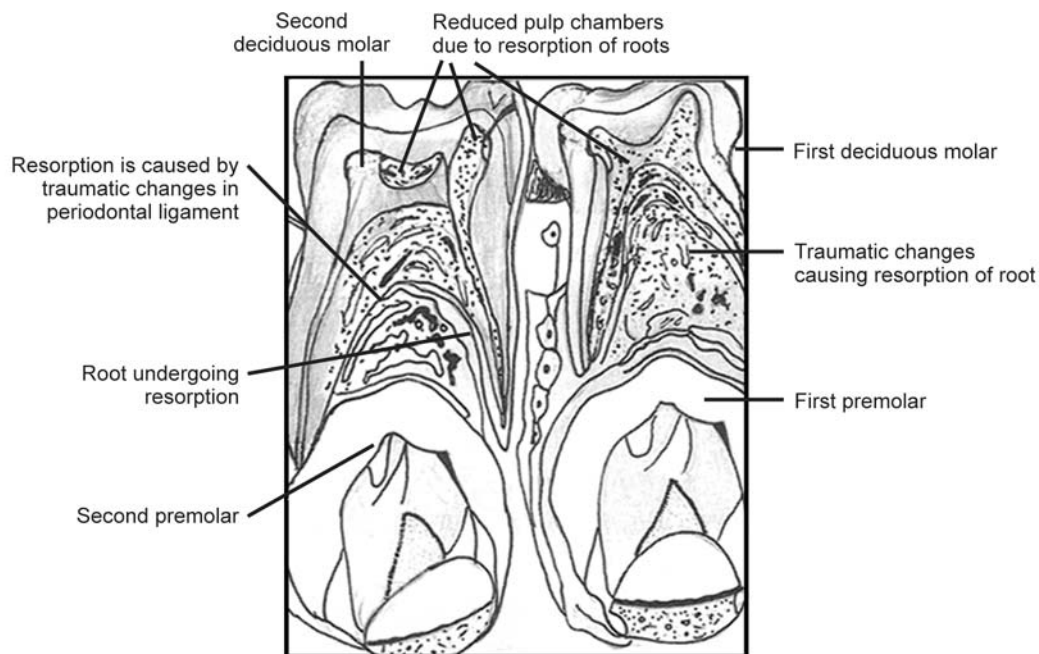


Figure 13.4: Deciduous first and second molars with developing first and second premolars below their roots (at the age of 4 years and 6 months)

of the deciduous molars. This resorption starts much before the deciduous molars are shed and it reflects the expansion of their growing permanent successors. But, ultimately, the successional tooth germs come to lie apical to the deciduous molars. This happens, because of the continuous growth of the jaws and occlusal movement of the deciduous molars. Space is provided for the continued development of the growing bicuspid, by this change in position. This also relieves pressure on the roots of the overlying deciduous molars. Deposition of a cementum like substance repairs the areas of early resorption. The resorption of the deciduous molars is again started, when the premolars begun to erupt and this time resorption continues until the roots are completely lost and the tooth is shed (Fig. 13.5).

The pattern of shedding is symmetrical for the right and left sides of the mouth. The mandibular deciduous teeth are always shed before their counterparts in the maxilla. The exception being the second molars. All four deciduous second molars are lost at approximately the same time. The teeth of the girls are shed little earlier than boys. The least discrepancy between the sexes is seen for the maxillary central incisors and maximum for the mandibular canines. The sequence of shedding in the mandible follows the anterior to posterior order of the teeth in the jaw. In the maxilla, loss of the first molar before the canine disrupts this sequence.

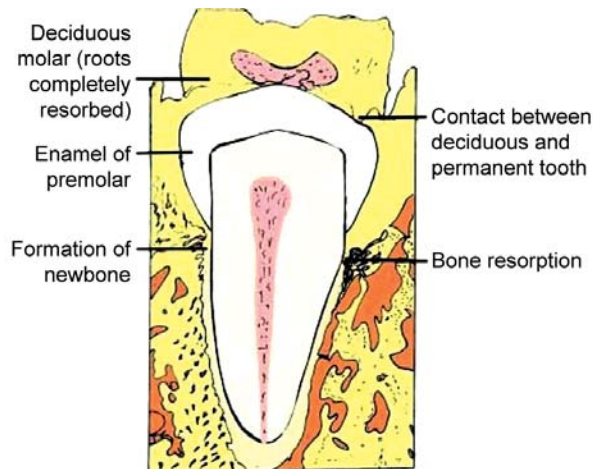


Figure 13.5: Root of deciduous molar are completely resorbed. Dentin of deciduous tooth is in contact with enamel of premolar. Resorption of bone on one side and formation of new bone on opposite side of premolar is caused by transmitted eccentric pressure to premolar

Briefly, the pre-eruptive, eruptive and posteruptive tooth movements are responsible for the initial placement of tooth in its functional position and its maintenance thereafter. Along with these movements, is the progression from the deciduous dentition through mixed dentition to the permanent dentition, which involves the shedding of primary teeth (Fig. 13.6).

HISTOLOGY OF RESORPTION

Local resorption of the deciduous tooth occurs at all sites where successional teeth contact them. The most important histological factor to be considered for the resorption of roots and hence exfoliation of deciduous teeth are the odontoclasts.

Odontoclasts (Dentinoclasts)

The odontoclasts, are giant multinucleated cells, which occupy resorption bays on the dental hard tissue surface. Their cytoplasm is vacuolated and they show a ruffled border on the surface, which is adjacent to the resorbing hard tissue (Figs 13.6 and 13.7). The ruffled border can be seen as an extensive folding of the cell membrane into a series of invaginations. These invaginations are 2 to 3 microns deep and contain mineral crystallites within their depth.

Contractile proteins, actin and myosin are present in the 'clear zone' (Fig. 13.8), which lies peripheral to the ruffled border. This clear zone does not contain any cell

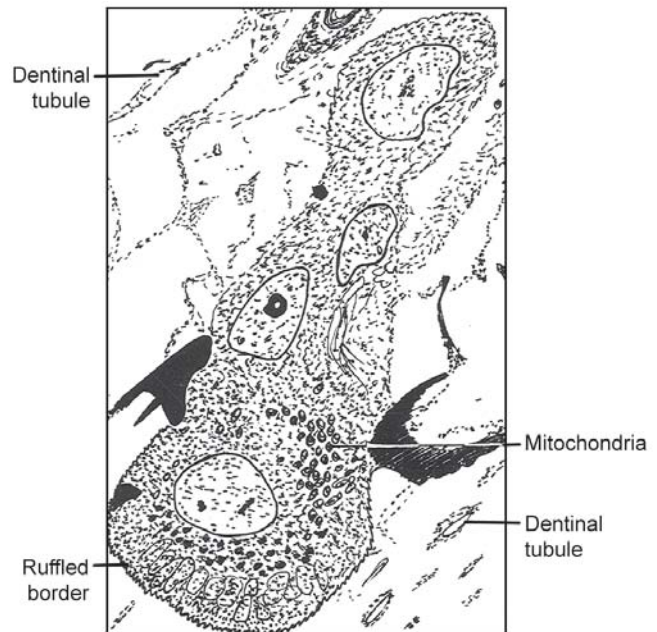


Figure 13.6: Multinucleated odontoclast cell with ruffled border which is well adapted to resorption lacuna in root dentin. Dense mitochondria are collected toward "Resorption" (lower) pole of cell and most of the cytoplasm is highly vacuolated

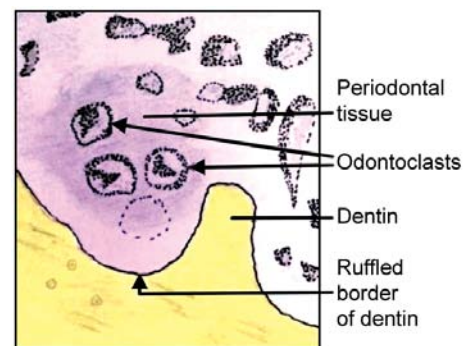


Figure 13.7: Resorption of dentin by odontoclast

organelles and it depicts the attachment apparatus of the odontoclast. Odontoclasts are derived from tartrate-resistant acid phosphatase (TRAP)- positive circulating monocytes. Osteoclasts are formed from the circulating monocytes.

Odontoclasts contain many vacuoles and high content of mitochondria (adjacent to the ruffled border) which shows increased acid phosphatase activity. Odontoclasts can resorb all the dental hard tissues, including enamel. During the resorption of dentin (Fig. 13.9), a pathway for the extension of odontoclast processes is provided by the dentinal tubules.

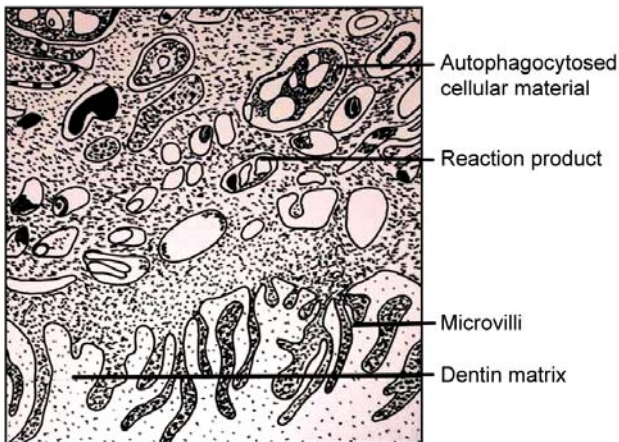


Figure 13.8: Interface between odontoclast ruffled border region (showing microvilli) and disintegrated dentin matrix of root surface undergoing resorption. Many vacuoles are especially concentrated adjacent to ruffled border showing varied contents of autophagocytosed cellular material and dense patches of reaction product. Acid phosphatase activity is seen within these vacuoles. (as seen under electron microscope)

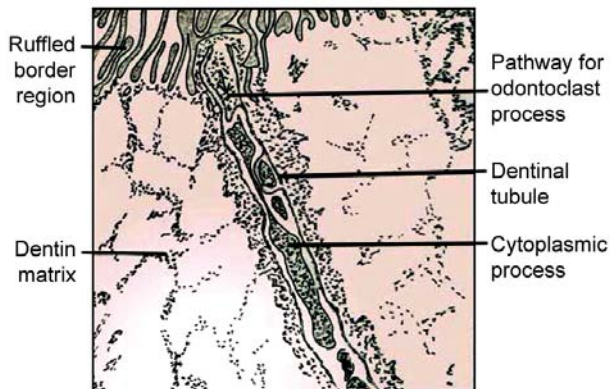


Figure 13.9: Cytoplasmic process occupying dentinal tubule emanating from ruffled border region of odontoclast. The bulk of field is occupied by dentin matrix. (As seen under electron microscope)

Odontoclasts have the same origin as osteoblasts. They are commonly found on root surfaces lying adjacent to the erupting successor, where they resorb cementum and dentin. They are generally not found in the pulp chambers of single rooted teeth, as these usually exfoliate before root resorption is complete (Fig.13.10). Therefore, the odontoblast layer remains intact.

In multirooted teeth, the odontoblast layer is replaced by the odontoclasts, which resorbs both the primary and secondary dentin (Fig.13.11). So, the roots are completely resorbed and the crown is also partially resorbed, prior to exfoliation.

Periods of resorption are interspersed with those of repair and rest but ultimately resorption predominates. Cells resembling cementoblasts lay down a collagenous matrix showing small foci of mineralization which helps in repair. The final repair tissue is similar to cellular cementum but is less mineralized (Figs 13.12 and 13.13). Resorbed enamel and dentinal surfaces are coated with cellular cementum like tissue secreted by mononuclear cells. Smaller and shallow resorption bays were covered with acellular cementum like tissue. Larger and deeper resorption bays were covered with cellular cementum. Cementum like tissue play important role in retention of deciduous teeth.

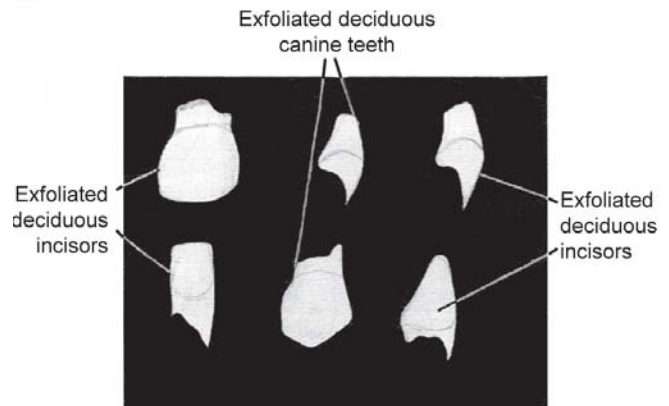


Figure 13.10: Random selection of exfoliated deciduous incisor and canine teeth showing shedding of these teeth before root resorption is complete, considerable amount of root dentin remains at the time of exfoliation

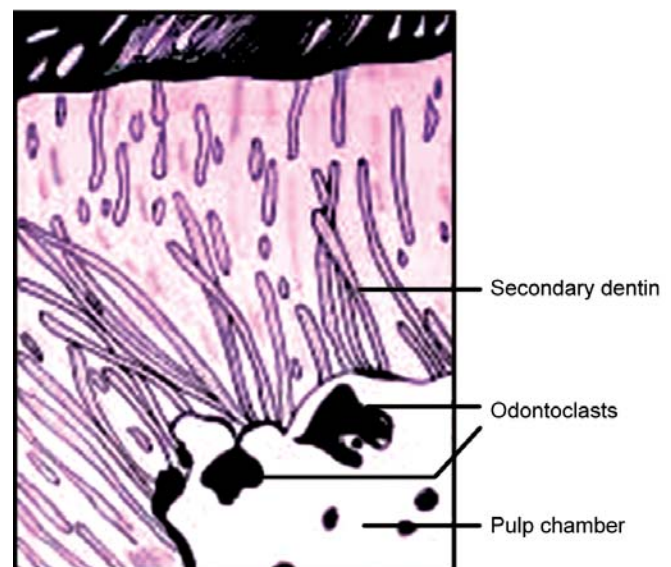


Figure 13.11: Resorption of secondary dentin by odontoclasts

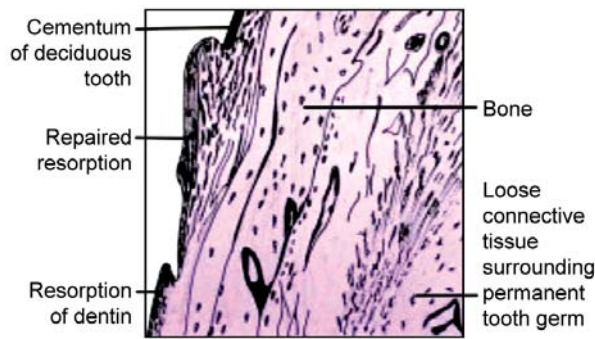


Figure 13.12: Repair of previous resorption seen in high magnification. The final repair tissue resembles cellular cementum but is less mineralized



Figure 13.14: Periodontal ligament fibroblast exhibiting intracellular collagen (as seen under electron microscope)



Figure 13.13: Repair of cementum in resorption lacunae, newly deposited repair tissue is not as electron dense as underlying cementum. (as seen under electron microscope)



Figure 13.15: A degenerated fibroblast in periodontal ligament exhibiting characteristic appearance of physiologic cell death

Gingival epithelium holds the tooth in the cervical area and tearing of this epithelium occurs in the final stage of shedding.

Not much is clear about the resorption of soft tissues, such as the pulp and periodontal ligament. It is assumed that loss of the periodontal ligament is sudden. Cell death in this area shows two distinct patterns

- i. The fibroblasts accumulate intracellular collagen, which indicates interference with normal collagen secretory mechanisms (Fig. 13.14).

- ii. The ligament fibroblasts show features characteristic of 'apoptotic cell death'. This involves condensation of the cell and its phagocytosis by adjacent macrophages (Fig. 13.15). The finding of apoptotic cell death in the resorbing periodontal ligament indicates that the exfoliation of teeth is a programmed event.

MECHANISM OF RESORPTION AND SHEDDING

Pressure from the erupting successional tooth plays an important role in shedding, as the odontoclasts always appears at the predicted sites of pressure. Pathologic root resorption may be caused by pressure exerted by tumors and cysts. Before resorption, cementoblasts covering the root are damaged by inflammatory process, because they are not responsive to cytokines and hormones. It is suggested that inflammatory process is initiated by the reduced enamel epithelium of the erupting permanent teeth.

In bone resorption, osteoblast first degrade the osteoid and expose the mineralized bone on which osteoclast can attach. In dentin resorption, same mechanism is followed. Predentin resists the resorption more than any other dental tissue.

A sealed space, lined by the ruffled border of odontoclast is created by attachment of the odontoclasts via the clear zone to hard tissue surface. Mineral dissolution occurs as hydrogen ions are constantly added to the extracellular environment via the membrane of the ruffled border, which acts as a proton pump. Additions of the hydrogen ion acidify it so that mineral dissolution occurs. Enzymatic contents are secreted into the same environment by the lysosomes. This causes degradation of the organic matrix.

If successional tooth germ is congenitally missing, shedding of deciduous tooth is delayed but may not be prevented. This clearly indicates that apart from pressure, other key factors are involved in initiating resorption. Growth of the jaws and face occurs with the corresponding enlargement in the size and strength of the muscles of mastication. This in turn increases the forces applied to the deciduous tooth to the point that the tooth's supporting apparatus, mainly the periodontal ligament, is damaged, initiating resorption. Less information is available about the resorption of pulp, periodontal ligament and dental soft tissue. Apoptotic cell death is supposed to be involved in the resorption of periodontal ligament.

Briefly, the superimposition of both local pressure and masticatory forces on physiologic tooth resorption decides the pattern and rate of the shedding of deciduous teeth.

Pressure from the erupting permanent tooth → some root loss → decreased tooth support → therefore tooth is less able to bear the increasing masticatory forces → thereby the process of exfoliation is accelerated.

CLINICAL CONSIDERATIONS

Remnants of Deciduous Teeth

In some cases, parts of root of deciduous teeth are not in the path of erupting permanent teeth. These remnants of root may not resorb and consists of cementum and dentin. These may remain in jaw for considerable time (Fig.13.16). They are most commonly found associated with the lower second premolars. The second deciduous molars have much flared and curved roots. The mesiodistal diameter of the second premolars is smaller than the greatest distance between the roots of the second deciduous molar. Therefore, the roots of the second deciduous molars are not in the path of their erupting successors and hence escape resorption. These remnants, consisting of cementum and dentin may remain buried in the jaw,

completely surrounded by and ankylosed to the bone (Fig. 13.17). When they come close to the jaw surface, they are eventually lost. The gradual resorption of these remnants and replacement by bone may cause their disappearance.

Retained Deciduous Teeth

Most commonly, the retained deciduous teeth are the maxillary lateral incisors, followed by the lower second premolar and rarely the lower central incisor. Deciduous teeth are retained beyond their shedding schedule usually when their permanent successors are missing or impacted.

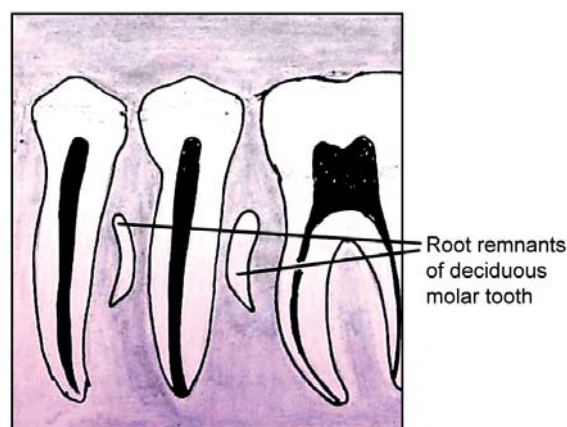


Figure 13.16: Remnants of roots of deciduous molar are embedded in interdental septa

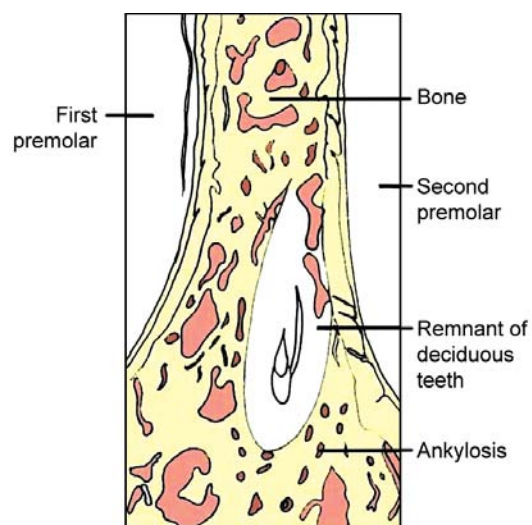


Figure 13.17: Root remnant of deciduous tooth is seen deep in the bone, completely surrounded by and ankylosed to the bone

If a permanent lateral incisor is missing, the deciduous tooth is resorbed under the pressure of the erupting permanent canine. This resorption occurs simultaneously with the resorption of the deciduous canine (Figs 13.18 A and B). Sometimes the permanent canine causes resorption of the deciduous lateral incisor only and erupts in its place. In such conditions, the deciduous canine is retained distally to the permanent canine.

Most commonly the deciduous and permanent canine teeth are retained. This occurs when the permanent tooth is impacted resulting in the retention of its deciduous predecessor (Figs 13.19 A to D). Sometimes, a supernumerary tooth or an odontogenic tumor may be present. These prevent the eruption of one or more than

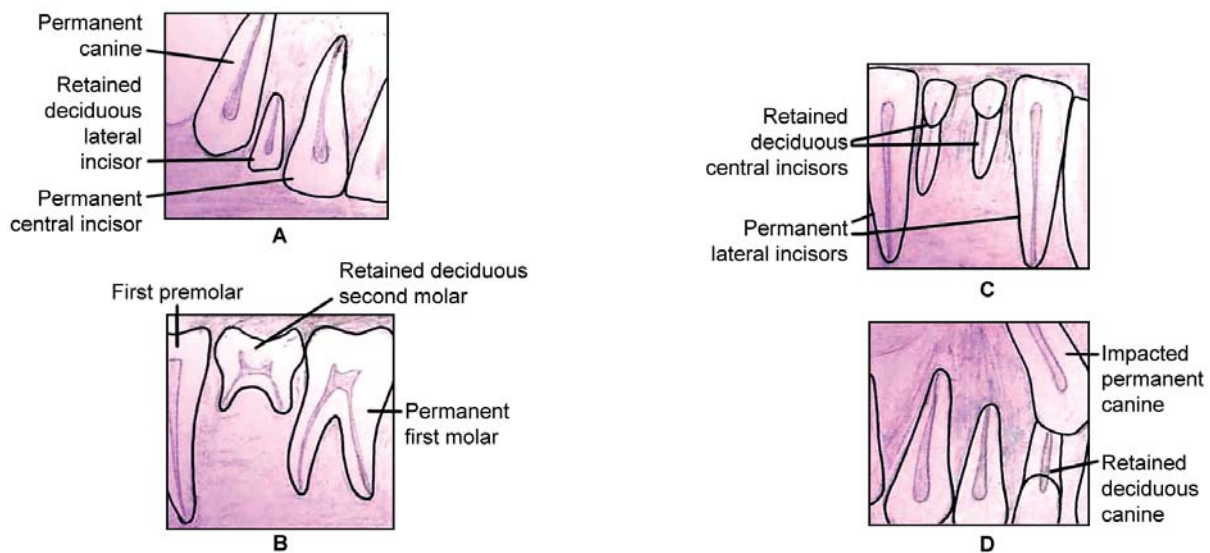
one permanent teeth. In such cases, ankylosis of the deciduous tooth may take place.

Submerged Deciduous Teeth

Trauma may damage the developing periodontal ligament causing prevention of eruption of the tooth and its subsequent ankylosis to the bone of the jaw. There is continued eruption of the neighbouring teeth along with increase in height of the alveolar bone. This causes the ankylosed tooth to become 'shortened' or 'submerged'. These teeth prevent the eruption of their permanent successors. Therefore, they should be extracted as soon as possible (Fig. 13.20).



Figures 13.18A and B: In the absence of permanent upper lateral incisor, resorption of deciduous lateral incisor and deciduous canine occurring due to pressure of erupting permanent canine; (A) At the age of 11 years. (B) At the age of 13 years



Figures 13.19A to D: Retained deciduous teeth. A. Retained deciduous maxillary lateral incisor and missing permanent lateral incisor. (At age of 56 years). B. Retained deciduous mandibular second molar with roots partly resorbed and missing mandibular second premolar. C. Retained deciduous mandibular central incisors and missing permanent central incisors. D. Retained deciduous maxillary canine and impacted permanent canine



Figures 13.20: Deciduous mandibular second molar submerged in the alveolar bone. Second premolar is absent. Deciduous second molar has tilted distally and permanent first molar has tilted mesially. No space for the eruption of second molar has been left

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Temporomandibular Joint

- Introduction
- Type of joint and its evolution
- Temporomandibular joint as synovial in nature
- Anatomy
- Histology of joint structures
 - Bony structures
 - Fibrous covering of articular surface
 - Articular disk
 - Synovial membrane
- Embryological development
- Ligaments associated with the joints
- Movements
- Clinical considerations
 - Developmental disturbances
 - Myofascial pain dysfunction syndrome (MPDS)
 - Fracture of articular fossa
 - Structural changes in TMJ
 - Trauma at TMJ
 - Arthritis

INTRODUCTION

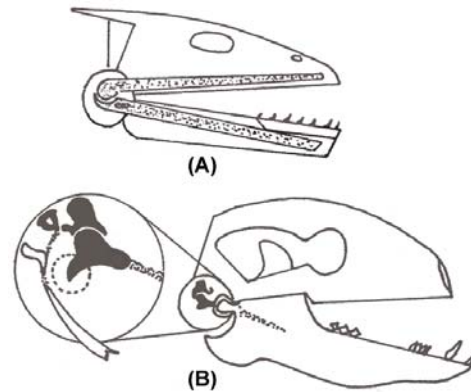
The temporomandibular joint (TMJ) is a complex and precisely integrated bilateral joint structure. It is formed by the articulation of lower jaw with the cranium and the upper facial skeleton. The bones taking part are the mandible and the temporal bones, that is why this joint is called temporomandibular joint. The head of the condyle of the mandible articulates with the articular fossa and articular eminence of the temporal bone. It functions in speech, mastication and deglutition. The downward and forward sliding action of the condylar heads can be felt by placing the fingers on them while opening and closing the mouth. It can also be felt during symmetric protrusion and retrusion and asymmetric lateral shift.

TYPE OF JOINT AND ITS EVOLUTION

Temporomandibular joint, is a synovial, bicondylar diarthrodial joint. It is further classified as ginglymus or ginglymoarthrodial joint that is, showing both sliding and hinge movements. The two condylar joints on each side always act in collaboration.

Among vertebrates, presence of the temporomandibular joint is specific only to mammals and is evolved in the manner given below. In other vertebrates the lower jaw is made up of several bones, which forms from the posterior part of the Meckel's cartilage. The lower jaw articulates with the quadrate bone of the skull (Fig. 14.1).

1. According to the figure, amphibian skull has dentary portion, which contains teeth and the articular portion, which is in articulation with the quadrate bone.
2. In evolutionary hierarchy next come the reptiles in which the articular portion is same as the amphibians that is the posterior portion of Meckel's cartilage is in articulation with the quadrate bone but the length of dentary or tooth-bearing portion is increased.
3. The jaws of mammalian reptilian show the development of coronoid process but the articulation is between the same two bones.
4. As mammals evolved, the compound lower jaw was reduced to a single bone, the mandible. In mammals, Meckel's cartilage does not form lower jaw and is only left as remnant. Now the articulation is formed only by dentary or mandible and with temporal bone forming temporomandibular joint (Fig. 14.2).



Figures 14.1A and B: Evolution of temporomandibular joint from amphibian to mammals; A. Amphibian skull: Teeth in dentary bone. An articulation between Meckel's cartilage and palatoquadrate bar; B. Mammalian skull: Dentary formed an articulation with temporal bone, inner ear formed by original joint

TEMPOROMANDIBULAR JOINT AS SYNOVIAL IN NATURE

Synovial joints generally consist of two articulating surfaces covered by hyaline cartilage. These joints show good amount of movement and are surrounded by a fibrous capsule, which forms joint cavity. The joint cavity is filled with synovial fluid, which is formed by synovial membrane. This synovial membrane lines the nonarticular surface of the joint.

Synovial fluid provides lubrication to the joint and helps in its movement. The ligaments associated with the joint helps to strengthen the articulation and check excess movement of the joint. Therefore TMJ has almost all the properties of the synovial joint.

ANATOMY

Temporomandibular joint is an articulation between the mandibular condyle and the temporomandibular fossa of the temporal bone. This joint is divided into two compartments by a disk. Joint is surrounded by a capsule which is lined with synovial membrane.

The temporomandibular joint is formed by; (i) temporal bone above, by involving anterior region of mandibular and articular tubercle (ii) Condyle of the mandible below (Fig. 14.2).

The articulating surfaces are lined by white fibrous tissue or fibrocartilage and not by hyaline cartilage which is present in other synovial joints.

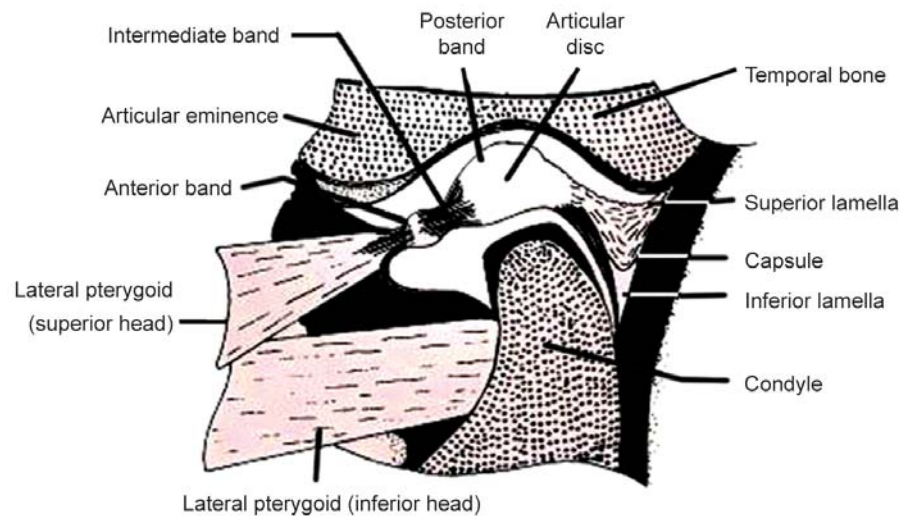


Figure 14.2: Diagrammatic representation of temporomandibular joint (in a sagittal section)

Joint Capsule

The whole of TMJ is enclosed in a fibrous capsule, which is attached to the articular tubercle in front and to the lips of squamous tympanic fissure posteriorly. In between these two attachments, it is attached to the borders of articulating surfaces of mandibular fossa and below the neck of the mandible. The joint capsule is a fibroelastic sac. The inner surface of capsule is lined by synovial membrane, which is smooth and glistening. Temporomandibular ligament attaches at the lateral aspect of the capsule and strengthens it.

Interarticular Disk

Interarticular disk is also called meniscus. It is fibrous in nature. It is situated between the articular surfaces of the joint and divides it into two, upper and lower compartments. This disk is shaped according to the shape of condyle to adjust it and in accordance with the concavity of mandibular fossa. The disk acts as a shock absorber and provides smooth surface for gliding motion. It is biconcave sagittally thicker anteriorly and thinner posteriorly, oval, avascular and non-innervated in the middle. It is firmly attached to the medial and lateral poles of condyle by medial and lateral collateral ligaments. Superior head of lateral pterygoid muscle attaches to anterior band of articular disk. The disk fits like a cap over the condyle. Posteriorly its undersurface has a concavity and upper surface has a convexity and

anteriorly it is saddleshaped. The head of the condyle and disk move in unison.

Anteriorly the disk splits into two lamellae, the upper sheet attached to the anterior edge of the articular eminence and the lower one to the anterior surface of the condylar head. In between these two lamellae, muscle fibers of the lateral pterygoid muscles are inserted into the disk. It also shows that movements of the disk and condyles are unified.

Posteriorly, the disk divides into inferior and superior lamellae. The inferior lamella is a thin extension of the fibrous disk. It runs over the posterior surface of the condylar head and fuses with the periosteum of the neck of the condyle. The upper component is extensive. A loosely textured mass of tissue is present in fibrous disk. It contains blood vessels and elastic fibers. This is confined posteriorly by the capsular ligament and superiorly gets mixed with capsular ligament and gets attached to squamotympanic fissure.

The disk also acts as a third bone, thus providing a movable articulation for the condyle. In the lower half of joint space rotational movement occurs which permits opening of jaw. In the upper joint space disk is firmly attached to the lateral and medial parts of the condyle.

Condyle

The condyle is convex and oval, wider lateromedially (13 to 25 mm) than anterioposteriorly (5.5 to 16 mm). The

long axis of condyle is oriented posteromedially, being angled backward at between 13 to 15° to frontal plane.

The heads of mandibular condyles articulate with glenoid fossae. At birth, the heads of the condyles are round. During development the condyle grows in a lateral direction. This results in a change in shape of the condyle, which becomes ovoid by maturity. The condyle has an ability to gradually change the shape, which allows adaptation to functional stress.

Articular Fossa

The articular fossa is the part of temporal bone which articulates with condyle. It is composed of an anterior part and a posterior part. Anterior part is in the form of eminence while posterior part is in the form of a depression or cavity in inferior part of temporal bone. Petrotympanic fissure is the junction of the temporal and petrous part of temporal bone. It is present on the posterior wall of fossa.

The articular fossa is oval in shape, situated anterior to the auditory canal. Its anterior boundary is formed by articular tubercle or eminentia articularis, externally it is bounded by zygomatic arch and posteriorly by tympanic plate which is the part of petrous portion of temporal bone.

Upper and Lower Compartments

Articular disk divides the temporomandibular joint cavity into upper and lower compartments. The upper one is called as temporodiskal compartment and the lower one is known condylodiskal compartment. The upper compartment is present between the articular fossa and the disk. The lower compartment is present between the disk and the head of condyle.

In the lower joint space, there occurs rotational movement about an axis through the heads of the condyles. It is a hinge movement and it permits opening of the jaw.

In the upper joint space, disk is firmly attached to the lateral and medial poles of the condyle and there is contraction of the inferior head of the lateral pterygoid muscle. Because of this the translatory movement occurs as the disks and condyle traverse anteriorly thus producing anterior and inferior movement of the mandible. In a healthy joint the superior and inferior joint spaces are reduced to the thickness of a synovial film except small spaces at the most anterior, posterior, medial and lateral limits of the joint spaces.

Blood Supply

Four arteries supply blood to temporomandibular joint. These are; (i) branches of the superficial temporal, (ii) deep auricular, (iii) anterior tympanic, (iv) ascending pharyngeal. These blood vessels approach the joint and penetrate its capsule and give off branches to the periphery of the disk and also to posterior area of the joint (Fig. 14.3).

Nerve Supply

The nerves which supply to temporomandibular joint arise from the branches of the mandibular division of the trigeminal nerve, mainly auriculotemporal, masseteric and deep temporal branches. Large myelinated and smaller non-myelinated nerves enter the capsule and disk. They supply all surfaces of the head, fossa, disk and the capsule. Nerve terminals of pain, pressure, touch and temperature are found within the joint. (Fig. 14.4).

Temporomandibular joint contains receptors present in the capsule. These receptors help in mandibular positioning in space. Important receptors are as follows.

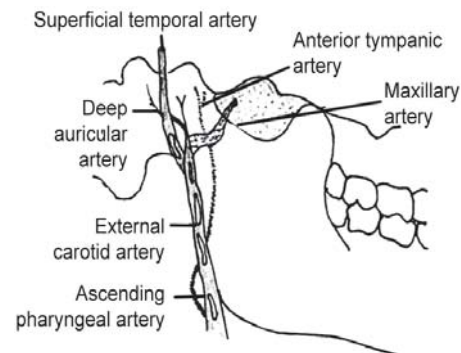


Figure 14.3: Blood supply of temporomandibular joint

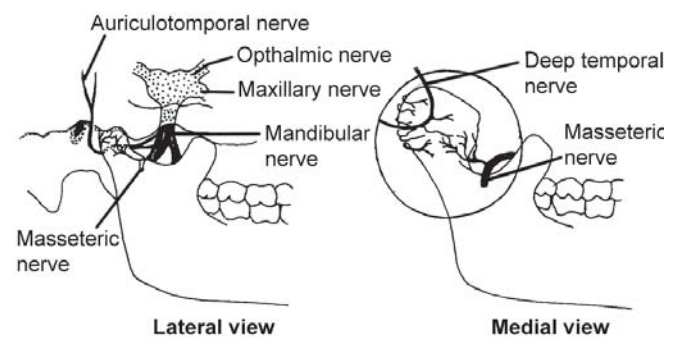


Figure 14.4: Nerve supply to temporomandibular joint

Proprioceptors—proprioceptors like Ruffini's corpuscles are present in the capsule and sense the changes in the joint in static condition.

Mechanoreceptors—Pacinian corpuscles in capsule, signal the slowness and rapidity of the joint movements. Golgi tendon in the TMJ ligament protect the joint when joint movements are excessive.

Nociceptors—Most numerous and widely distributed free nerve endings causing pain and curtailing the movement for the protection of joint from the excessive movements.

HISTOLOGY OF JOINT STRUCTURES

Bony Structures

The condyle of mandible and glenoid fossa of squamous part of temporal bone form the bony structure of temporomandibular joint (Fig.14.5).

Condyle

Condyle of the mandible is made up of inner core of cancellous bone and outer core of compact bone (Fig. 14.6).

The cancellous bone consists of trabeculae which have large marrow spaces in between them. These marrow spaces contain red marrow. In older individuals, the marrow spaces reduce in size because of the thickening of the trabeculae and red cellular marrow or myeloid type of marrow changes to fatty and yellow marrow.

The articulating surface of condyle is covered by thick dense fibrous perichondrium. Hyaline cartilage lies between the fibrous connective tissue and the compact bone of condyle. Sometimes, remnants of hyaline cartilage persist into old age. The hyaline cartilage of the condyle

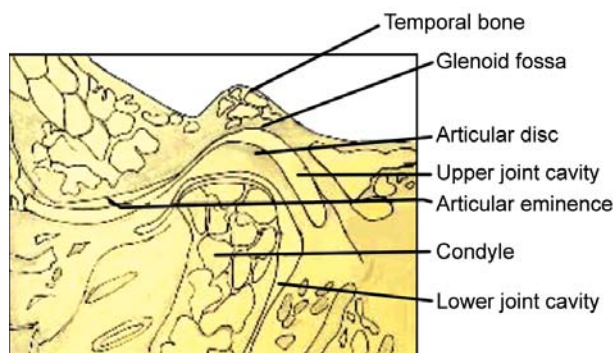


Figure 14.5: Histologic appearance of the temporomandibular joint in sagittal section (microscopic view in low power)

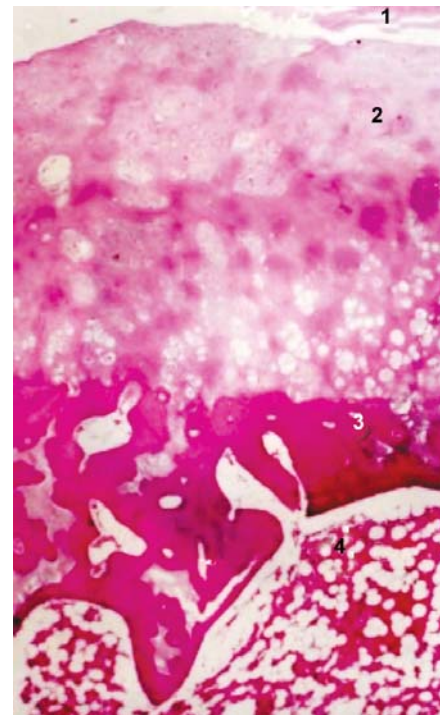


Figure 14.6: Histological structure of condylar head of mandible in a young adult; 1. Fibrous layer 2. Cartilage 3. Bone 4. Bone marrow (H & E x 100)

is referred to as the secondary cartilage. At the interface between forming bone and the cartilage, the hyaline cartilage of the condyle is not organized into parallel rows of cells. Along with appositional growth and subperiosteal mandibular bone growth, the growth of secondary cartilage and its replacement with bone occurs. This results in downward and outward growth of the mandible. This growth can be stimulated intrinsically (internally) by growth hormones and extrinsically (externally) by force, e.g. functional appliances.

Glenoid Fossa

The roof of glenoid or mandibular fossa is made up of a thin plate of compact bone. The articular tubercle is made of inner core of cancellous bone covered by compact bone. In articular eminence, areas of chondroid bone and islands of hyaline cartilage are also seen.

Fibrous Covering of Articular Surface

Both the articulating surfaces are covered by dense, fibrous tissue, which contains fibroblasts and chondrocytes. It is

fibroelastic in nature (Fig.14.7). The fibrous covering consists of network of strong collagenous fibers. The fibrous lining on the articular surface of temporal bone is double layered, divided into inner and outer layer, fibers of inner layer have orientation perpendicular to the bone surface and the fibers of outer layer are parallel to the surface.

Superficial layer of the fibrous lining covering the articulating surface of mandibular fossa is uniformly thin but rapidly thickens on the posterior slope of articular tubercle.

The fibrous covering of condyle is of uniform thickness and occasionally consists of chondrocytes in the superficial layer. These stain deeply with basic dyes. Small undifferentiated cells are present in the deep layer of cartilage. Few thin collagenous fibers are present in the deep layer. This is known as reserve cell zone. In this zone, appositional growth of the hyaline cartilage of the condyle occurs. Fibrocartilage bear the mechanical stress and to some extent, resists aging process.

The articulating surface of the temporal bone is covered by fibrous layer. This layer is thin in the articular fossa and gets thickened on the posterior slope of the articular eminence. Here the fibrous tissue shows arrangement of two layers with a small transitional zone between them. The course of fibers of the inner zone is at right angles to the bony surface. The fibers in the outer zone run parallel to the surface. The chondrocytes may be found in the tissues on the temporal surface. The deepest layer may show a thin zone of calcification in adults.

Articular Disk

The articular disk is composed of dense collagen fibers, which are straight and tightly packed. Few fibroblast cells are found in between which are elongated, having flat cytoplasmic processes. Sometimes, chondroid like cells are identified in areas of disk, which are subjected to excessive mechanical stress. The fibrous collagenous structure provides flexibility to the disk, which helps in translatory and rotatory movements of the condyle. The presence of chondrocytes increases the resistance and resilience of the fibrous tissue.

The central area of meniscus has no blood and nerve supply, thus despite continuous pressure from moving condyle, the articular disc does not undergo resorption or cause pain, and once damaged, it has a poor reparative capacity.

Synovial Membrane

Synovial membrane, at the time of birth, lines all the internal surfaces of the joint including the articular disc. After birth as the joint starts functioning, synovial lining is lost from all the articulating surfaces and covers only the inner aspect of articular capsule.

Synovial membrane is thrown into the folds which form synovial villi that project into the joint spaces. The villi increase the flexibility of inner surface of capsule which enables the easy change of shape of disc and sulcus to occur during normal movement.

The synovial membrane consists of two layers, which are intima and subintima.

The subintima is made up of loose connective tissue rich in blood supply.

The intimal layer which is present directly adjacent to the joint cavity contains cells of three principal types (Fig. 14.8).

- i. B cell type or S cell type: This cell type is rich in rough endoplasmic reticulum and its main function is secretion of synovial fluid. It is also called as fibroblast like or secretory S cell sometimes .
- ii. A cell types: Rich in Golgi complex and lysosomes and contains less or no rough endoplasmic reticulum. It has macrophage like phagocytic activity and it helps to debride the joint cavity. These are also known as macrophage like cells.
- iii. This type lies in between A cell and B cell type in morphological aspects. Regeneration ability is seen in synovial membrane.

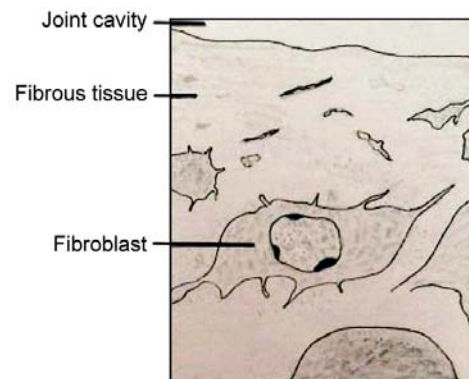


Figure 14.7: The fibrous articular tissue covering the mandibular condyle (as seen under electron micrograph)

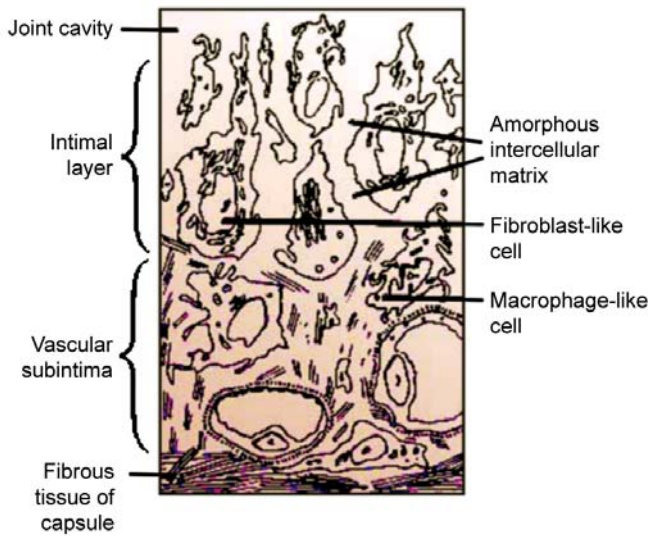


Figure 14.8: Synovial membrane of temporomandibular joint (as seen under electron micrograph)

Synovial Fluid

Synovial fluid is a clear, straw colored fluid found in joint spaces. It is a viscous fluid, which functions to lubricate the joint. It also provides nutrition to avascular fibrous tissue, which covers the condyle and the articular eminence and the disk.

It is diffused from the rich capillary network of synovial membrane in association with B cells (described above) which augment the secretion of synovial fluid. The amount of synovial fluid decreases as the age increases.

EMBRYOLOGICAL DEVELOPMENT

The temporomandibular joint is a secondary development, which means that it is developed from primary reptilian type. In the reptilian type, the TMJ is formed at dorsal end of Meckel's cartilage, which in humans appears as joint between malleus and incus bones of the middle ear showing adaptation of bones of primitive jaw to sound conduction. Meckel's cartilage forms and provide skeletal support for developing lower jaw. The cartilage extends backward and dorsally and terminates as malleus.

The TMJ develops by approximately 10th week of intrauterine (IU) life from two widely separated centers that grow toward one another. Appearance of two regions of mesenchymal condensation is the first evidence of the presumptive joint development. These are the temporal and condylar blastemas. The temporal blastema develops first and condylar blastema afterward. Temporal and

condylar blastema are present at some distance from each other. Ossification begins first in temporal blastema. By the 12th week, a pair of clefts appears in the mesenchyme between developing squamous portion of temporal bone and develop into upper and lower joint cavities thus defining intervening articular disc (Fig. 14.9).

The joint capsule is formed from condensation of mesenchyme, which progressively isolates the joint with its lining synovial membrane. At the time of birth, the glenoid fossa and articular eminence are poorly developed and the fibrous capsule is the only means of providing stability to the joint.

After the 12th week, *in-utero* development of condyle takes place very rapidly (Fig. 14.10). Condyle develops by endochondral growth that is by growth of cartilage. The condylar cartilage is slowly replaced by membranous bone leaving small posterior part which remains as cartilage till the end of 2nd decade of life and helps in active growth and change in condylar morphology in response to functional and environmental effect.

LIGAMENTS ASSOCIATED WITH THE JOINTS

Three ligaments are associated with the temporomandibular joint, which stabilize the joint. These are as follows (Figs 14.11 and 14.12):

- a. Temporomandibular ligament
- b. Sphenomandibular ligament
- c. Stylomandibular ligament

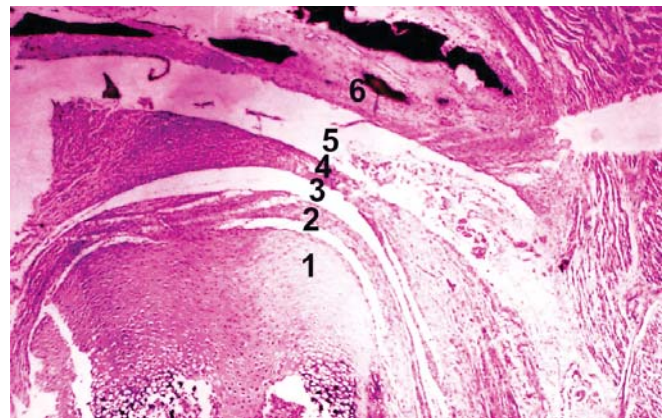


Figure 14.9: Section of temporomandibular joint in a 16-week-old fetus 1. Condylar head of the mandible, 2. Fibrous capsule 3. Lower articular cavity, 4. Articular disc, 5. Upper articular cavity, 6. Articular tubercle of temporal bone (H and E x 50)

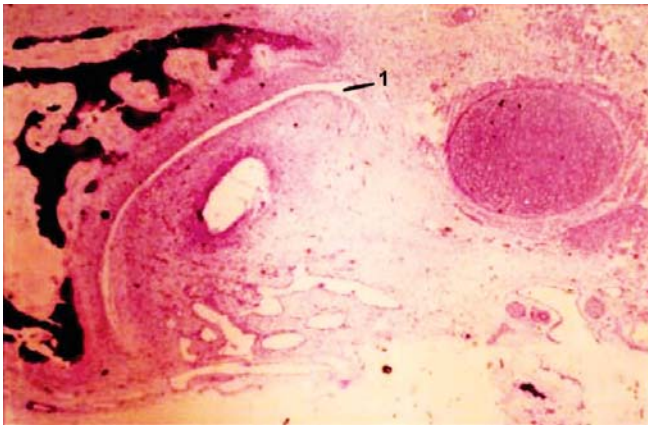


Figure 14.10: Section of temporomandibular joint: Fetus 18- week-old: Joint cavity (1) surrounded by articular surface made of immature mesenchymal tissue. Ossification of the articular surfaces has not yet taken place (H & E x 30)

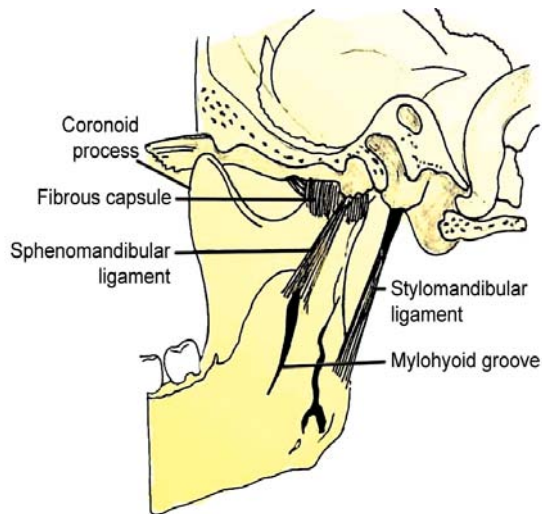


Figure 14.11: Medial aspect of right temporomandibular joint

Out of these three ligaments the temporomandibular ligament is functionally most significant for the joint.

Temporomandibular Ligament/ Lateral Ligament

Temporomandibular ligament is triangular in shape. It helps to strengthen the fibrous capsule as it is intimately related to it. Ligament is attached above to the root of Zygoma, below to the lateral surface and posterior to border of the neck of mandible. Its fibers are covered by parotid gland superficially and run obliquely downward and backward. The flexible ligament consists of dense band of fibrous tissue with some elastic fibers in between.

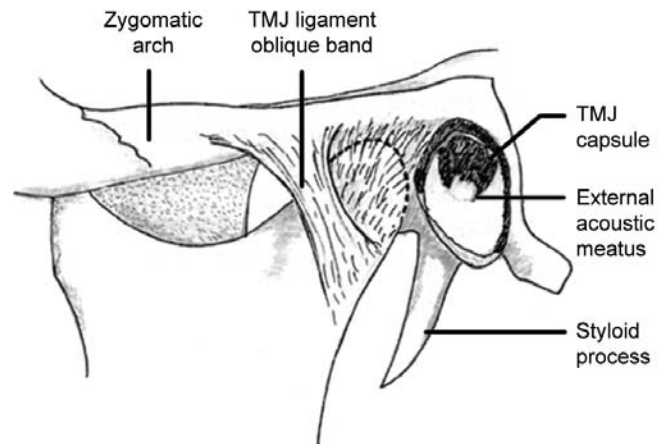


Figure 14.12: Capsule and ligament of temporomandibular joint

Sphenomandibular Ligament

Sphenomandibular ligament is an accessory ligament. It is attached above to the spine of sphenoid and below to the lingula of the mandibular foramen. This ligament is a vestigial part of dorsal end of Meckel's cartilage and its contribution to the joint is doubtful.

Stylomandibular Ligament

Stylomandibular ligament is also another accessory ligament of the joint. It is a thickened band of deep cervical fascia, which stretches from the apex of the styloid process to the angle and posterior border of ramus of mandible. This ligament is an accessory ligament and is functionally not useful to the joint movement.

MOVEMENTS

Muscles Producing Movements

Depression: Contraction of lateral pterygoid muscles is mainly supported by digastric, geniohyoid and mylohyoid muscles. This causes depression. This helps when mouth is opened wide or against resistance.

Elevation: Produced by temporalis, masseter and medial pterygoid. The rest position of jaw is maintained by temporalis muscle. These muscles of both sides helps to elevate the mandible.

Protrusion: Lateral and medial pterygoids.

Retraction: Posterior fibers of temporalis. It is assisted by some fibers of the masseters, digastric and geniohyoids muscles.

Lateral movements: Medial and lateral pterygoid muscles act simultaneously, i.e. muscles of both side acts alternately.

Movements

Inter-articular disk or meniscus divides the joint space into two compartments.

- *Condylodiskal* (between condyle and disk).
- *Temporodiskal* (between disk and temporal bone).

Condylodiskal or lower compartment is associated with rotational hinge movement. The forward translational movement takes place in the upper temporodiskal compartment in which the condyle moves downward, forward, towards and over the articular eminence. During opening of jaw, condyle first rotates which is followed by forward and downward translocation of condyle. Meniscus follows the condyle closely as it moves forward. During sideways movements of jaw, the condyle toward the movement rotates with slight lateral shift known as Bennett shift. The

contralateral or opposite side condyle shows corresponding forward translation and rotation movements (Fig. 14.13).

CLINICAL CONSIDERATIONS

Developmental Disturbances

Aplasia of Condyle

Failure of development of mandibular condyle may occur unilaterally or bilaterally. In unilateral condylar aplasia, there is obvious facial asymmetry toward the affected side.

Hypoplasia of Mandibular Condyle

Underdeveloped or defective formation of condyle, which may be congenital or acquired because of reasons like forcep deliveries in newborns and damage following radiation therapy of jaws. This also causes facial asymmetry producing lateral deviation (excursion) of mandible on the affected side.

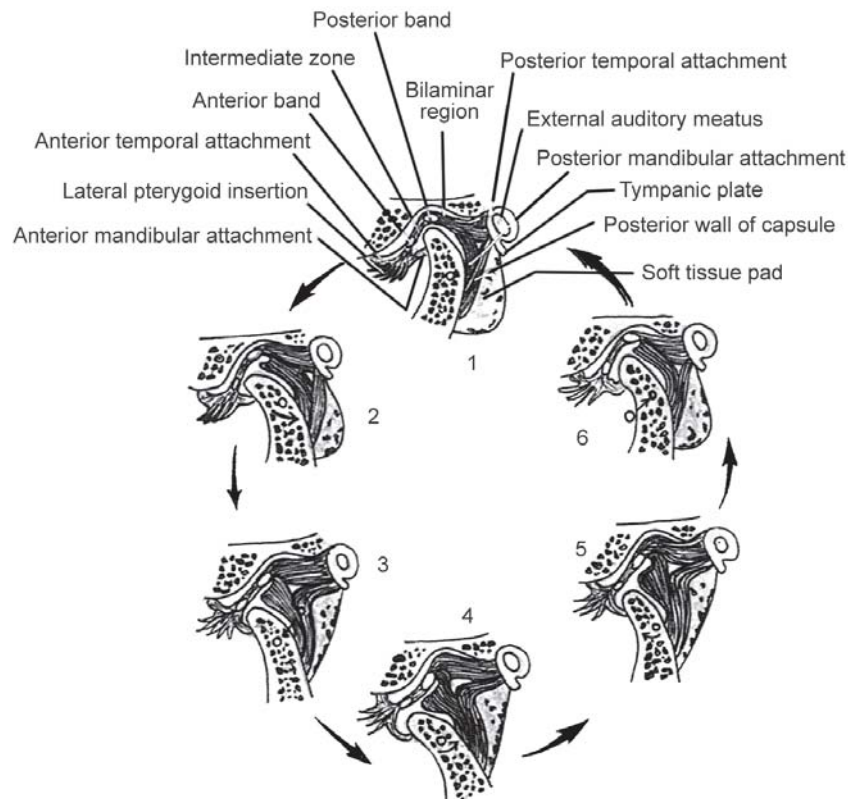


Figure 14.13: Changes in the position of condyle of the mandible, the articular disc and the articular surface of temporal bone during a full cycle of opening (1→2→3→4) and closing (4→5→6→1) of the mouth

Hyperplasia of Mandibular Condyle

Chronic mild inflammation may cause hyperplasia of condyle unilaterally causing unilateral elongation of face and deviation toward the normal side. Approximately in 35 percent of the population, during opening movement, the TMJ produces very mild to moderate sounds. Usually the intensity of these sounds gradually increases as the age advances.

Dislocation

During wide opening of the mouth like in yawning, condyle moves extremely anteriorly and may pass over the articular eminence and get locked in a position so that it cannot be voluntarily returned to its normal position. This condition needs very careful external manipulation of mandible in downward and backward direction for extrusion of the mandible at proper place. This happens mostly in aged persons and mostly in females because of (a) wearing and resorption of articular eminence (b) reducing muscle tone with the advancing age. For treatment, injections of various sclerosing solutions are given in TMJ capsular ligaments to tone them up with variable success rate.

Ankylosis

Ankylosis is immobility and consolidation of joint and its most frequent cause is injury to the joint or infection in or around the joint. It may occur due to; (i) intrauterine fetal abnormality, (ii) forcep delivery, (iii) malunion of condylar fractures, (iv) metastatic malignancies, (v) rheumatoid arthritis, (vi) inflammation of joint from infection in ear like otitis media.

In complete ankylosis, the bones of condyle and mandibular fossa are completely fused and there is complete lack of mobility. Ankylosis after birth causes failure of development of mandible.

Myofacial Pain Dysfunction Syndrome (MPDS)

Myofacial pain dysfunction is a dysfunction of TMJ, it is also called the temporomandibular joint pain dysfunction syndrome. It is characterized by:

- i. Pain.
- ii. Muscle tenderness.

- iii. A clicking or popping noise in the temporomandibular joint.

- iv. Limitation of jaw motion, unilaterally or bilaterally, sometimes with deviation on opening.

The pain is usually unilateral. It is described as a dull ache in the ear or preauricular area which may radiate to the angle of the mandible, temporal or cervical area. The intensity varies frequently between morning and the remainder of the day.

The muscle mainly involved in MPD syndrome is lateral pterygoid muscle.

The treatment for MPD syndrome is conservative since the condition may be related to stress.

Fracture of Articular Fossa

The bone of articular fossa, being thin, is prone to fractures. By heavy blow mandibular head is driven into the fossa.

Structural Changes in TMJ

Several structural changes take place if there is change of force or direction of stress. This frequently occurs after the loss of posterior teeth. The important change is separation between the collagen bundles. This results in the pain at the TMJ.

Trauma at TMJ

Function of the joint is impaired in severe trauma. In this the articular bone is destroyed. Cartilage and new bone develop in the marrow spaces and at the periphery of the condyle. Articular surface gets remodeled according to the functional demands.

Factors like loss of teeth, attrition and orthodontic treatment show the changes in articular surface.

Arthritis

Besides other joints of the body osteoarthritic changes take place in TMJ also. The synovial fluid is reduced in amount and becomes thicker in viscosity. This results in swelling in joint capsule and pain. Sometimes very small cysts also appear in the condylar heads. Diagnostic techniques like, computerized tomography, magnetic resonance imaging and arthrographic technique are used in the diagnosis of TMJ diseases.

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Maxillary Sinus

- Introduction
- Development and developmental anomalies
- Structure and variations
 - Variations in structure of maxillary sinus
- Microscopic features and histology
 - Control of secretion
- Functions
- Clinical considerations

INTRODUCTION

The sinuses are the air spaces within the pneumatic bones situated at the fronto-basal region of the skull. These are four in number, namely frontal, maxillary, sphenoidal and ethmoidal. These form the boundaries of the nasal cavities. All these open into the nasal cavity through their lateral walls.

Maxillary sinus is a large pneumatic space present in the body of the maxilla communicating with the environment through middle nasal meatus and the nasal vestibule. It is the largest of all the sinuses. Maxillary sinus is also known as antrum of Highmore. Highmore was the first person to describe the morphology of the maxillary sinus in detail and also explained the function of pneumatization by the sinuses.

DEVELOPMENT AND DEVELOPMENTAL ANOMALIES

Among all the sinuses, maxillary sinus is the first to develop. It appears as a shallow groove on the medial surface of maxilla during the fourth month of intrauterine life. It grows rapidly during 6 to 7 years of life. The maxillary sinus starts developing at sixteenth week of the intrauterine life (Fig.15.1). In early gestation, when the crown-rump length (CRL) of an embryo is 32 mm, many events occur. These morphogenic events take place to differentiate the nasal cavity, after which the initial development of maxillary sinus follows (Fig. 15.2).

At first, the horizontal shift of the palatal shelves occurs. These shelves then fuse with each other and also with the nasal septum. With this fusion, the secondary oral cavity gets separated from the secondary nasal chambers. All these changes result in the expansion of the lateral nasal wall, which starts folding. As a result of this folding, three nasal conchae and three meatus arise (Figs 15.3A and B). For first half of the intrauterine life, the superior and inferior meatus remain as shallow depressions along the lateral nasal wall. The middle meatus expands into the lateral nasal wall. The middle meatus grows inferiorly because of the formation of the cartilaginous skeleton of the lateral nasal wall. It occupies maximum space of the future sinus. Thus, in an embryo of about 32 mm CRL, the sinus appears and grows vertically to form maxillary sinus and gradually attains a measurable diameter (Table 15.1).

In perinatal period (between 28th week of pregnancy and ending 28 days after birth) the human maxillary sinus further grows in all the three directions, that is anteroposteriorly, superoinferiorly, mediolaterally, and attains a bigger size and volume (Table 15.2). From one year to the adulthood, maxillary sinus grows and attains approximately 15 mm of capacity (Table 15.3).

Table 15.1: Measurement of growth of maxillary sinus during embryonal period

Crown-rump length (CRL) (in millimeter) of embryo	Diameter (in millimeter) of the maxillary sinus
50	1
160	3.5
250	7.5

Table 15.2: Measurement of growth of maxillary sinus during perinatal period (between 28th week of pregnancy and ending 28 days after birth)

Direction of measurement	Amount of growth (in millimeter)	Standard deviation(SD) of growth
Anteroposterior	7 to 16	2.64
Superoinferior	2 to 13	1.52
Mediolateral	1 to 7	1.18

Table 15.3: Measurement of growth of maxillary sinus from one year to adult

Age (in years)	Direction and amount of growth (measurement in millimeter)		
	Anteroposterior	Superoinferior	Mediolateral
1	15.0	6.0	5.5
15	31.5	19.0	9.5
>15	34.0	33.0	23.0

The maxillary sinus expands and modifies in form and it reaches its final height after the eruption of all permanent teeth. It gains an average capacity of about 15 mm.

Developmental Anomalies

Several developmental anomalies like hypoplasia (underdevelopment), agenesis (failure of a part or an organ to develop or grow), aplasia (failure of an organ or tissue to develop normally) may occur alone or with other anomalies. These may be atresia of concha, cleft palate, high palate, septal deformity, absence of a concha, mandibulofacial dysostosis, malformation of the external nose and pathological conditions of the nasal cavity.

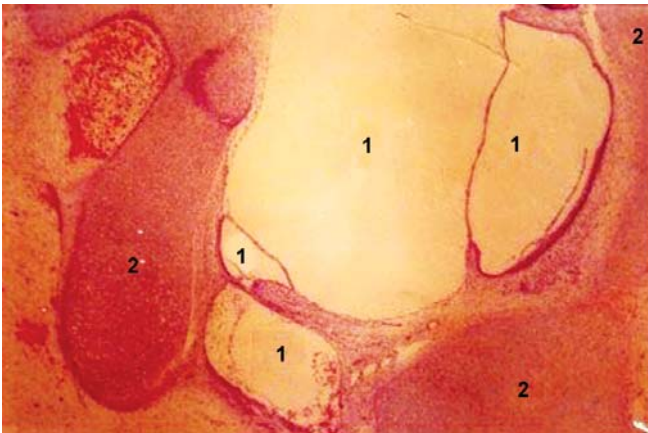


Figure 15.1: Maxillary sinus: Development in 16-week-old fetus: Sagittal section showing large air spaces (1) Surrounded by nasal cartilages. (2) Air spaces are partly lined by columnar epithelium. (H and E x 30)

There may be the presence of the two completely separate sinuses on the same side. This is due to outgrowth of the nasal mucosa into maxillary body from two different points. These may be either in middle nasal meatus or in middle and superior or middle and interior nasal meatus. This results in the formation of two separate ostia of the sinus.

STRUCTURE AND VARIATIONS

The maxillary sinus is a large pyramidal cavity, which is present in the body of the maxilla. It is a four-sided pyramid. The walls of maxillary sinus are thin and these correspond to the orbital, alveolar, anterior and

infratemporal aspects of the body of the bone (Fig. 15.4). Its apex directs laterally and extends into the zygomatic process. Its base faces medially which forms the lateral wall of the nasal cavity. The four sides are related to the surface of maxilla as follows.

- i. Anterior side to the facial surface of body.
- ii. Inferior to the alveolar and zygomatic process.

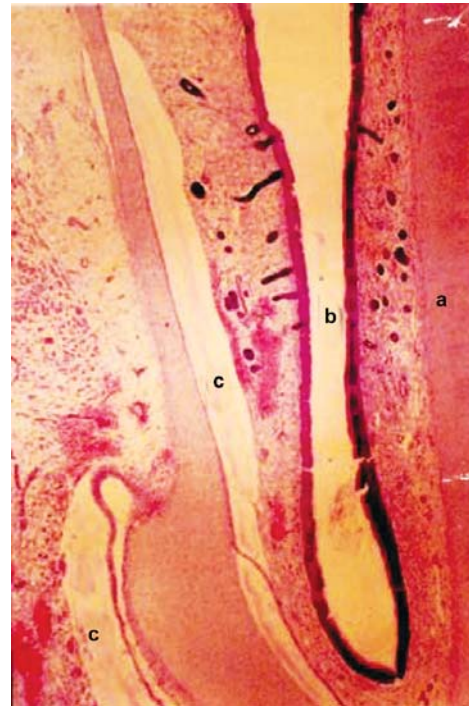
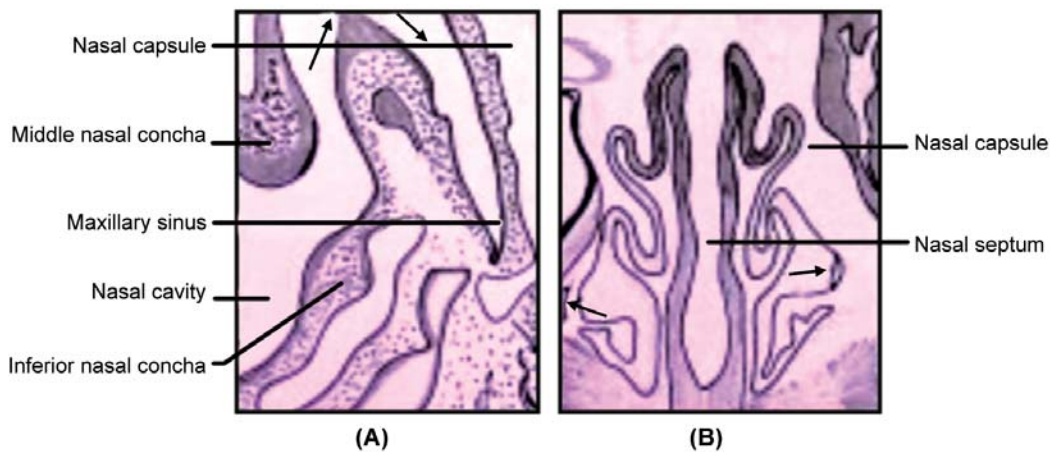


Figure 15.2: Development of maxillary sinus: Fetus 18 weeks old: Part of nasal septum (a) Nasal cavity, (b) Maxillary sinus or air spaces, (c) (x 30)



Figures 15.3A and B: (A) Coronal section of 69 mm fetus showing various developing structures around maxillary sinus. (B) Coronal section of 60 mm fetus showing nasal cavities separated by nasal septum, three conchae with palate

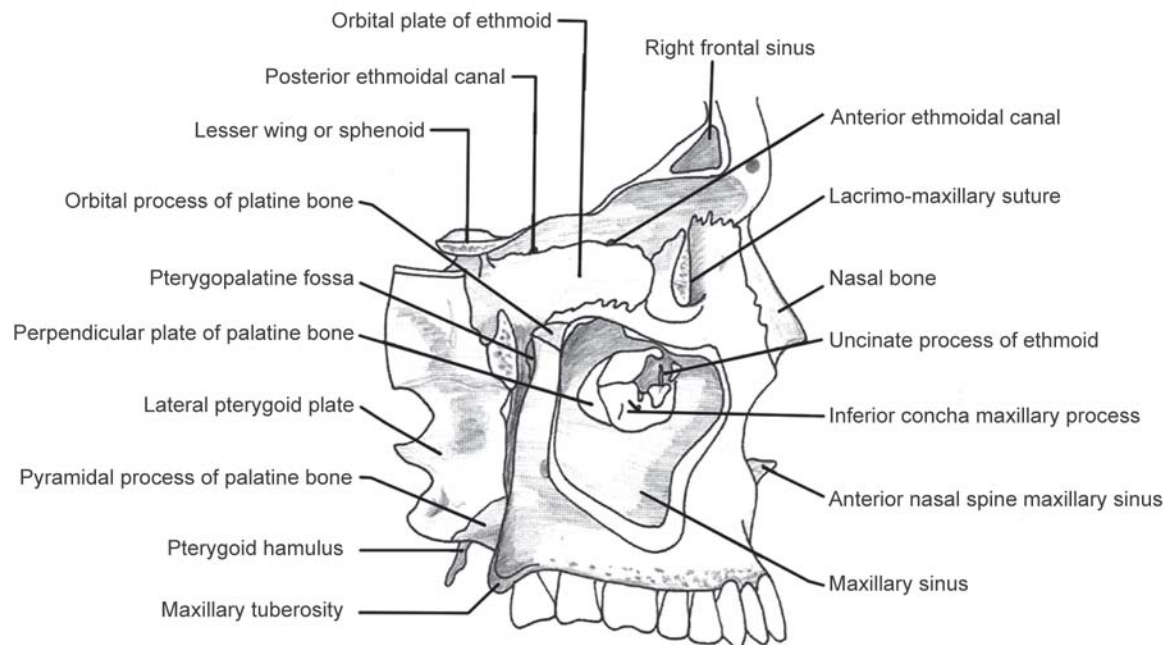


Figure 15.4: An oblique sagittal section through the anterior part of the skull, showing the medial wall of the right orbit and the medial wall of the right maxillary sinus

- iii. Superior to the orbital surface.
- iv. Posterior to the infratemporal surface.

The base of the sinus is thinnest of all walls, and it presents a perforation or natural opening the ostium at the level of middle nasal meatus. The floor of sinus is formed by the alveolar process of the maxilla. The roof of sinus is formed by the orbital floor.

Variations in Structure of Maxillary Sinus

Number of variations in shape, size and mode of the developmental pattern are found in maxillary sinus. The maxillary sinus communicates with the middle meatus of the nose, by an ostium. But in some individuals beside main ostium, two or many more accessory ostia connect the sinus with the middle nasal meatus.

There may be differences in the location of ostium. The main ostium may be located within the anterior third of the hiatus semilunaris in rare cases. In some cases, it is located within the middle third while in majority of the cases, it is found within the posterior third of the hiatus semilunaris. The accessory ostia may be found in middle nasal meatus and are rarely present in inferior nasal meatus.

During the course of development maxillary sinus pneumatizes the maxilla. But this may occur beyond boundaries of the maxillary body. As a result the processes

of the maxilla become invaded by air spaces. These spaces are called as recesses. In majority of cases, this space is found in the alveolar process and is in descending order in zygomatic process, frontal process and palatine process of the maxilla.

Because of the zygomatic recess, the superior alveolar neurovascular bundle comes near to the space of the sinus. The frontal recess invades and surrounds the content of the infraorbital canal. The alveolopalatine recesses reduces the amount of the bone between the dental apices and the sinus space.

In fully developed alveolar recess, there is presence of three depressions. These are separated by two incomplete bony septa. The anterior depression corresponds to the site of premolar buds, the middle to the first and second molar bud, and the posterior to the third molar bud.

When the two maxillae are articulated, a funnel shaped depression, the incisive fossa, is seen in the median plane immediately behind the incisor teeth. Caudal to the incisive fossa, the midline palatal intermaxillary suture is flat. In some cases however, its bony margins are raised, forming palatine torus. Its incidence shows facial variations.

A very thin bone separates floor of the sinus and roots of the maxillary molars. Very rarely, there may be no bone separating the root apices from the sinus, they are separated only by sinus lining.

MICROSCOPIC FEATURES AND HISTOLOGY

The space of the maxillary sinus is surrounded by three layers. These are the epithelial layer, the basal lamina and the subepithelial layer, which also includes the periosteum. Its epithelium is pseudostratified, columnar and ciliated (Fig. 15.5). It is derived from the olfactory epithelium of the middle nasal meatus. The columnar ciliated cells are the most numerous cell type in maxillary sinus epithelium. Other cells present in sinus epithelium may be basal cells, columnar non-ciliated cells and secretory goblet cells, which produce mucous.

Nucleus, cytoplasm, mitochondria and enzyme containing organelles are present inside the ciliated cells. In apical segment of the cell are present the basal bodies. These serve as the attachment of the ciliary microtubules to cell. The cilia are composed of 9 + 1 pairs of microtubules. They provide the motile apparatus to sinus epithelium. As cilia beat, the mucous on epithelial surface moves from sinus interior towards the nasal cavity.

All the features of secretory cells are present in the goblet cell. In basal part of the goblet cell are present the nucleus, cytoplasm containing rough and smooth endoplasmic reticulum, and the Golgi apparatus. These all helps in secretion of mucosubstances. The zymogen granules of the Golgi apparatus transport mucopolysaccharides to the cell apex. These are finally secreted by exocytosis on the surface of the epithelium. Along with the epithelial secretion, there is the presence of mixed secretory product from the subepithelial gland. The subepithelial layer of the sinus contains sub-epithelial glands. These glands secrete mixed secretory product. The glands reach the sinus lumen, after piercing the basal lamina, by excretory duct (Fig. 15.6).

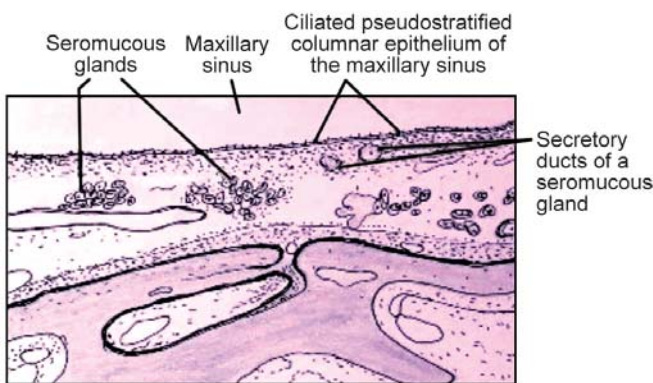


Figure 15.5: Structure of mucous membrane of maxillary sinus



Figure 15.6: Secretory ducts of maxillary gland of maxillary sinus. Ductal cells, cuboidal and columnar in shape, surround the lumen (L) which is upto 12.5 micron in radius

Subepithelial glands consist of acini. These acini contain two types of secretory cells, serous cells and mucous cells. The serous cell produces an electron dense homogenous secretory material. The mucous cell shows positive reaction to the alcian blue, due to mucin production, which is an electron lucent, heterogenous secretory material.

The myoepithelial cells are present around the acini. The myoepithelial cells are like smooth muscles cells in morphology, contain myofilaments and form the contractile units (Fig. 15.7).

Control of Secretion

Both the divisions of the autonomic nervous system control the secretion from these glands. Maxillary sinus is supplied with autonomic axons with general sensory components from maxillary nerve complex.

In the subepithelial layer of sinus are present nonmyelinated and myelinated axons. Nonmyelinated axons are more in number than myelinated axons. These axons are closely related to blood capillaries, fibroblasts, fibrocytes, collagen bundles and other connective tissue elements in the subepithelial layer (Fig. 15.8).

FUNCTIONS

The structure of the maxillary sinus and its relationship to surroundings has resulted in its various functional importance. The functions of the paranasal sinuses are

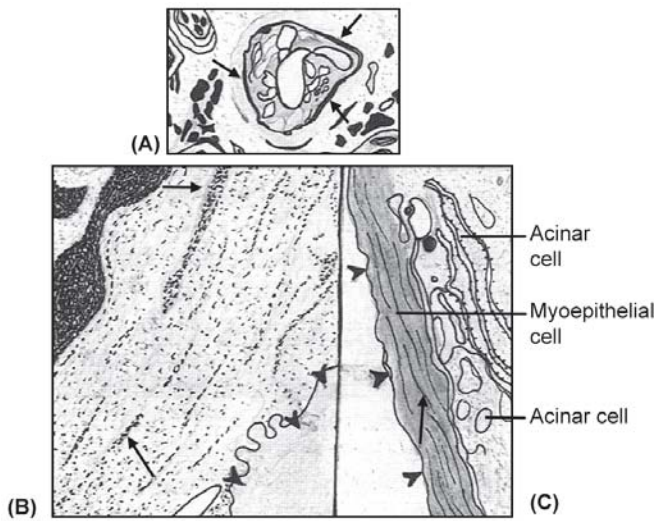


Figure 15.7: Myoepithelial cells (A) Thin section (0.5 micron) of several mucous and ductal cells from maxillary gland. Periphery of this acinus is surrounded by dark-appearing myoepithelial cells (arrows). (B) and (C) show relationship between acinar cells and myoepithelial cells and numerous bundles of filaments (arrows) which occupy most of the cytoplasm of the myoepithelial cells. Basement lamina adjacent to myoepithelial cell is indicated by arrowhead. (as seen under electron microscope)

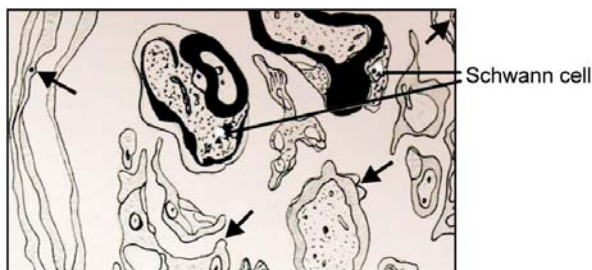


Figure 15.8: Two myelinated and several nonmyelinated axons in mucoperiosteal layer of maxillary sinus. Schwann's cells (labeled and at arrowheads) are intimately related to axons which contain individual mitochondria and microtubules or microfilaments cut in different planes. Arrows show connective tissue elements surrounding either individual axon or entire nerve (as seen under electron microscope L x 11200)

not very well known because of the relative inaccessibility of the sinuses to systemic functional studies and because of the great variation in sizes of the sinus.

In the individuals with large maxillary ostium, which is conveniently situated in the hiatus semilunaris, the air pressure in the sinus fluctuates from + 0.7 to + 4.0 mm of water between the nasal expirations and inspirations.

Following are some of the functions performed by the maxillary sinus:

1. Maxillary sinus is regarded as an accessory space to the nasal cavity. This occurs as a result of an inadequate process of ossification. Thus, it reduces the weight of the skull.
2. Add resonance to the voice.
3. Related to the structure and topography of ostium, it is concluded that the sinus humidifies and warms the inspired air and thus protects the internal structures, especially the brain against cold and dry air.
4. Sinus moistens the nasal cavity and probably contributes to olfaction.
5. It enhances the faciocranial resistance to mechanical shock.
6. Maxillary sinus produces certain bactericidal lysozyme to the nasal cavity.

CLINICAL CONSIDERATIONS

There is some correlation between the extent of pneumatization by sinuses with the general dysfunctions of the endocrine system. In case of pituitary gigantism, the sinuses assume a larger volume. In some congenital infections like spirochetes in congenital syphilis, the pneumatic processes are suppressed. It results in small sinus.

Maxillary sinus, due to its topographic location and its functional and systemic association with the orodental complexes, results in the transfer of pathologic conditions between each other. The transfer of pathogenic condition is either by mechanical connections or by way of blood and lymphatic pathways. The base of the sinus is separated from apices of the maxillary molars (usually I molar) and maxillary premolars (Figs 15.9 and 15.10) by a thin bone, or sometimes there may be no bone in between.

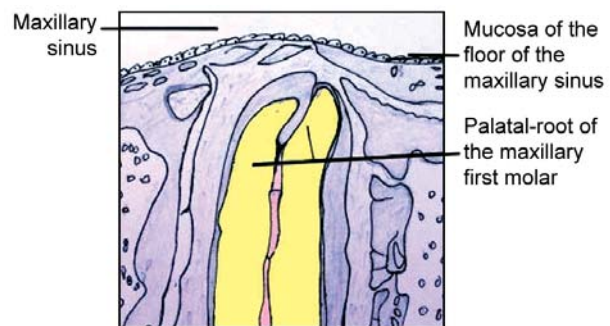


Figure 15.9: Relationship of maxillary sinus and root apex of maxillary molar

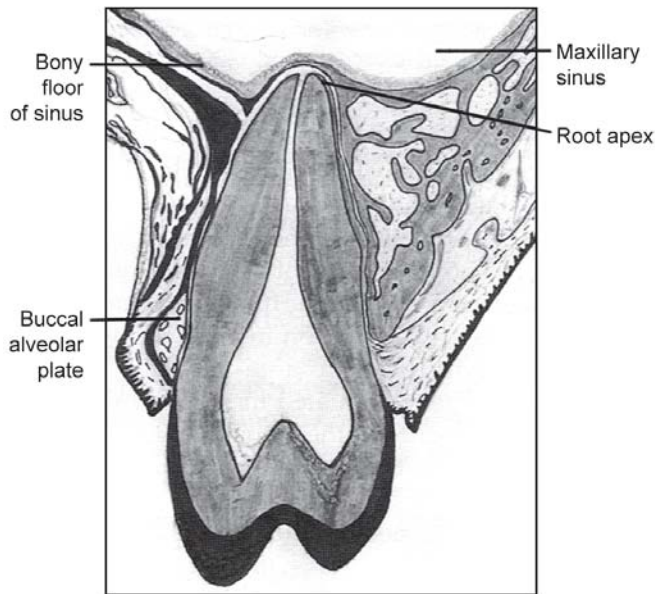


Figure 15.10: Buccolingual section through second maxillary premolar. Apex is separated from sinus by very thin plate of bone



Figure 15.11: Excessive dipping of floor of maxillary sinus causing underdeveloped roots of maxillary premolars

Surgical manipulation of these teeth often results in an oroantral fistula. Such untreated fistula may result in a permanent connection between the maxillary space and the oral cavity.

Due to the approximation of the sinus to the roots of the maxillary molars and premolars certain specific

conditions are observed. In some cases, there is dipping of the floor of maxillary sinus. This results in underdevelopment of the roots of maxillary premolar and molars, so roots of these maxillary teeth remain short (Fig. 15.11).

Presence of molar or premolar radicular cyst, hypercementosis abscess or granuloma may also result in oroantral fistula. Therefore, radiographs should be taken and analysed before any surgical procedure related to maxillary premolar or molar teeth is done. If extraction of the tooth affected with hypercementosis is done, it may lead to the perforation.

Inflammation of sinus is known as sinusitis. It causes headache with thick purulent, persistent discharge from the nose. So, its diagnosis should be assisted by transillumination and radiograph. In chronic infections of the maxillary sinus, the superior alveolar nerve is involved. It may cause pain like that of dental origin, i.e. pathology may be in maxillary sinus but pain may be felt in teeth or vice versa. It is sometimes observed that the pathology like chronic alveolar abscess is present in relation with maxillary premolars and molars, but pain or heaviness is felt in maxillary sinus of that side. So careful examination of the maxillary sinus as well as of the maxillary teeth is required to differentiate their cause.

Trigeminal neuralgia may have its origin in the maxillary nerve. It may also present with pain in the teeth. Various infections of pneumococcal, streptococcal, staphylococcal or viral origin may spread from either teeth or maxillary sinus, and involve the other. Pain, loosening, supraeruption of teeth, or bleeding of their gingival tissue may be a primary manifestation of malignant tumors of the maxillary sinus.

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Age Changes in Oral Tissues

- Introduction
- Age changes in enamel
- Age changes in dentin
- Age changes in pulp
- Age changes in periodontium
 - Changes in gingiva
 - Changes in periodontal ligament
 - Changes in cementum
 - Changes in alveolar bone
- Age changes in oral mucosa
- Age changes in salivary glands
- Age changes in temporomandibular joint and occlusion

INTRODUCTION

Aging is a gradual slowing down of the natural functions. It is a regressive process of physiologic and morphologic disintegration. Dorland Medical Dictionary has described aging as "the gradual structural changes that occur with the passage of time, that are not due to disease or accident, and that eventually lead to increased probability of death as the organism grows older". Taber's Medical Dictionary has defined aging as "the physiological changes occurring with age. Many changes occur with aging which affect the various structures of the oral cavity. Aging is manifested to different limits in different tissues. General features of aging are tissue desiccation, reduced elasticity, diminished reparative capacity and altered cell permeability.

As age advances in human beings, the teeth, jaws and other oral tissues exhibit changes in their structure and functions.

AGE CHANGES IN ENAMEL

Being a nonvital tissue, enamel is incapable of replacement. As age advances, the enamel is progressively worn away on occlusal surfaces and also on proximal surfaces due to masticatory attrition. Wear facets are more often found in older people because large portion of the crown is eroded.

The main characteristics of aging in enamel include changes in color, permeability and in the nature of the surface layer. These all result in reduction in incidence of caries. Teeth darken with age. This darkening can be due to addition of organic material from the environment to the enamel. This can also be due to deepening of the dentin color, which is seen through the translucent enamel, which becomes thin with advancing age.

With age, enamel becomes less permeable. In young age, enamel permits the slow passage of water and substances of small molecular size through pores between the crystals. So it behaves as semipermeable membrane. With age the pores diminish because crystal size increases due to uptake of more ions. As bulk of the water lies in the pores it means that water content of the enamel decreases with age.

With age the composition of the surface layer changes. This is because the ionic exchange takes place between the enamel surface and the oral environment. Incidence of caries progressively decreases with age. This indicates

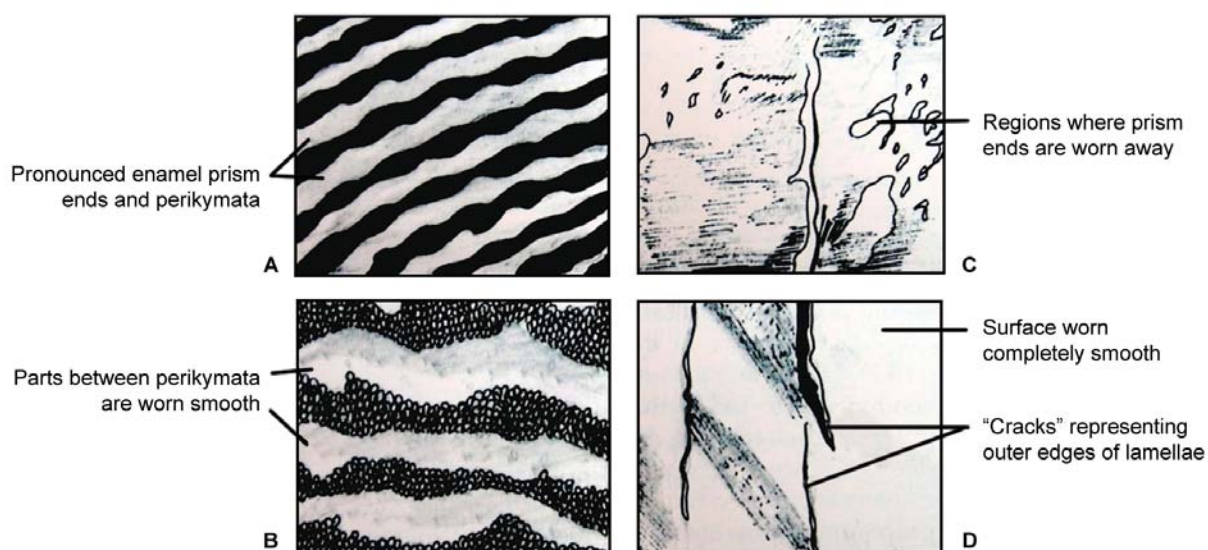
a change within the enamel which gives it greater resistance. Pits and fissures on occlusal surfaces of teeth are worn down in old age. This removes susceptible sites for caries. This also accounts for the decreased incidence of caries with age. The surfaces of the unerupted and recently erupted teeth have pronounced rod ends and perikymata, which disappear completely with age (Figs 16.1A to D). Adults generally take fewer refined carbohydrates and there is increase in fluoride content of enamel. These factors result in decreased incidence of caries with age.

AGE CHANGES IN DENTIN

Dentin is a vital tissue. It is laid throughout the life. However, after the teeth have erupted and have been functioning for some time, the rate of dentinogenesis that is further dentin formation is gradually very much reduced. Even then during aging dentin gradually becomes thicker due to secondary dentin and the reparative dentin or tertiary dentin deposition. With age gradually, due to wearing of enamel, the dentin is exposed to the oral environment. Bacteria, living or dead, or their toxic products, chemical substances from restorative materials, migrate down the tubules to the pulp and stimulate pulpal response. This leads to formation of reparative dentin. The deposition of the reparative dentin is to seal off the irritant, which may be invading in any form and the zone of injury. It occurs as a healing process, which is initiated by the pulp and result in reduction of the inflammatory process and removal of the dead cells.

Reparative dentin has fewer and more twisted tubules than normal dentin. With age, there is a continuous deposition of the intratubular dentin. This leads to complete closure of the tubule and the dentin becomes translucent or sclerotic. With advancing age, attrition of the teeth occurs. As a result sufficient stimuli are generated to cause collagen fibers and apatite crystals to appear in the dentinal tubules. Blocking of the tubules is a defensive reaction of the dentin. The refractive indices of the dentin and the occlusal tubules become equal and such areas become transparent. Sclerotic dentin is mostly found in apical dentin near the root tip in the teeth of the elderly people (Fig. 5.44). The sclerosis increases the brittleness and decreases the permeability of the dentin. Thus, it prolongs pulp vitality.

Another age change observed within dentin is the occurrence of dead tracts (Figs 5.66 and 5.68), often found



Figures 16.1A to D: Tooth surface showing progressive loss of structure with advancing age. A. Surface of recently erupted tooth at 12 years of age. B. Early stage of loss of surface structure at 25 years of age. C. Later stage of loss of surface structure at 52 years of age (Since these are negative replicas, surface details appear inverted. Raised structures represent depressions in actual surface). D. Surface worn completely smooth and showing only "CRACKS" at 50 years of age

in the coronal dentin and frequently bounded by bands of sclerotic dentin. At places where reparative dentin seals dentinal tubules at their pulpal end dentinal tubules fill with fluid or gaseous substances. These areas show decreased sensitivity and appear in older teeth.

AGE CHANGES IN PULP

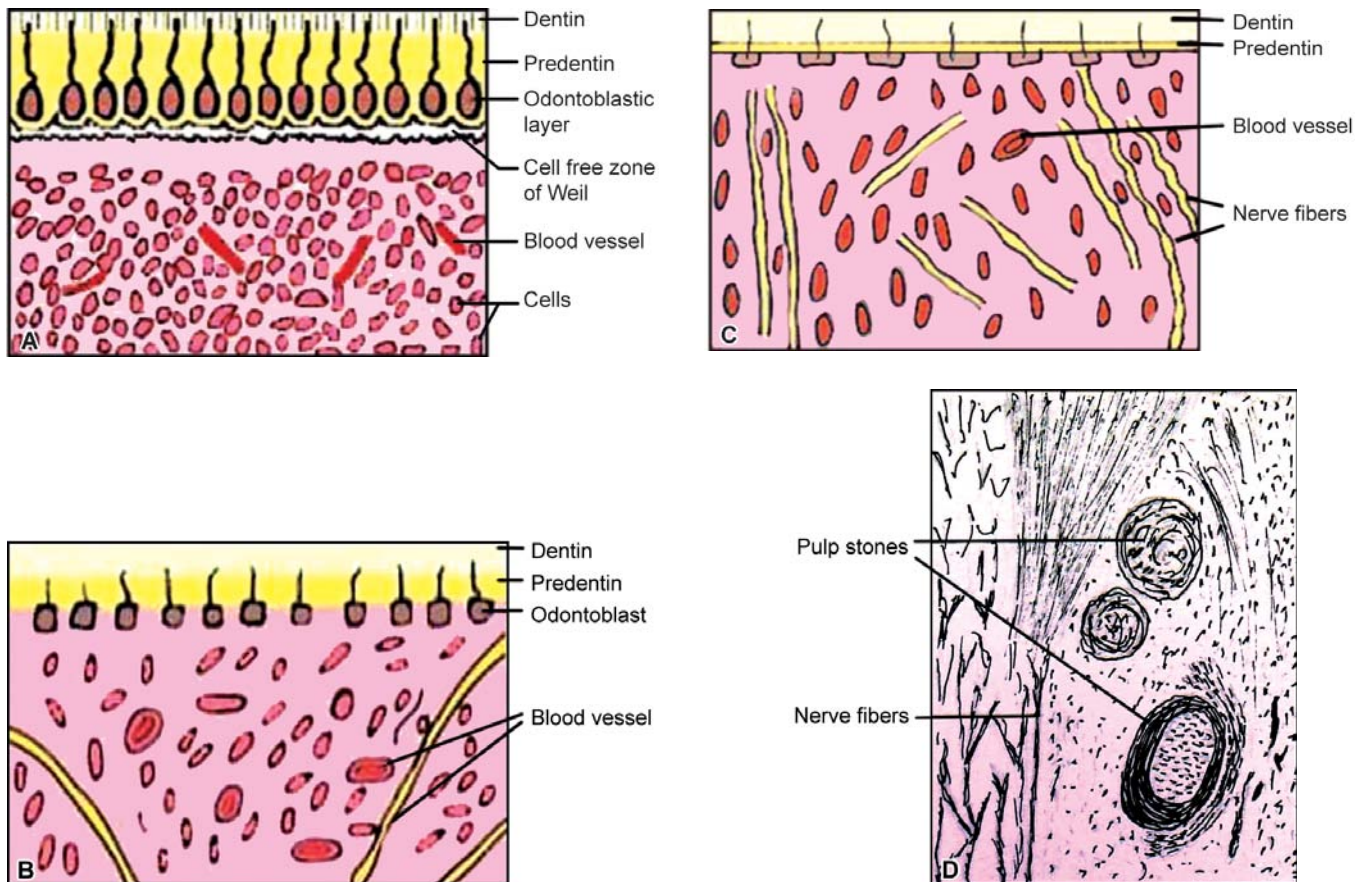
A number of changes are seen in the pulp as age advances. These constitute the regressive changes or aging of the pulp. These changes are the cell changes, fibrosis and the formation of the pulp stones.

The most common change is the decreasing volume of the pulp chamber and root canal due to continued deposition of the dentin. Sometimes the root canal appears to be almost completely obliterated in teeth of aged persons. The pulpal cells gradually decrease in number as the age advances, whereas the collagen content (fibrillar component) of the pulp increases. The increase in collagen fibers is attributed to the decrease in size of the pulp chamber. Along with decrease in number of cells in aging pulp, there is decrease in size and number of cytoplasmic organelles. The fibroblasts in ageing pulp show less perinuclear cytoplasm and possess long thin cytoplasmic processes. The decrease in cells and increase in fibers in pulp organ is gradual and generalized throughout the organ.

Certain vascular changes occur in the aging pulp. There is appearance of the atherosclerotic plaques in pulpal blood vessels. A true change is the occurrence of the irregular areas of dystrophic calcifications. These are found mostly in the central areas of the pulp in the form of concentric layer or linear pattern. These are known as pulp stones. The pulp stones are nodular, calcified masses which may appear in coronal or in root portion of the pulp (Figs 16.2 A to D). These can be seen in functional as well as embedded unerupted teeth.

These may be true, containing dentinal tubules, or false pulp stones, in which there is absence of dentinal tubules and shows concentric layers of calcified tissue. (Figs 16.3, and 16.4). These pulp stones may also be embedded in dentin, attached to dentin or free in the pulp (see Fig. 6.77). The ground substance of pulp becomes less aqueous as age advances. The free pulp stones are entirely surrounded by pulp tissue, attached stones are partly fused with dentin and embedded stones are completely surrounded by dentin.

In aging pulp, there is generally the accumulation of the diffuse fibrillar components. There is also the appearance of bundles of collagen fiber. The increase in fibers in the pulp organ is gradual (Fig. 16.5). Diffuse calcification appears as irregular calcific deposits in the



Figures 16.2A to D: Age changes in dental pulp; A. Pulp of newborn infant (a) More cellular elements (b) Less fibrous element (c) High vascularity (d) More columnar odontoblasts (e) Thick predentin layer; B. Pulp of six years old child; C. Pulp of 30 years old adult. By aging cellular elements decrease and fibrous intercellular substances increase in the pulp; D. Pulp stone seen in aged pulp tissue

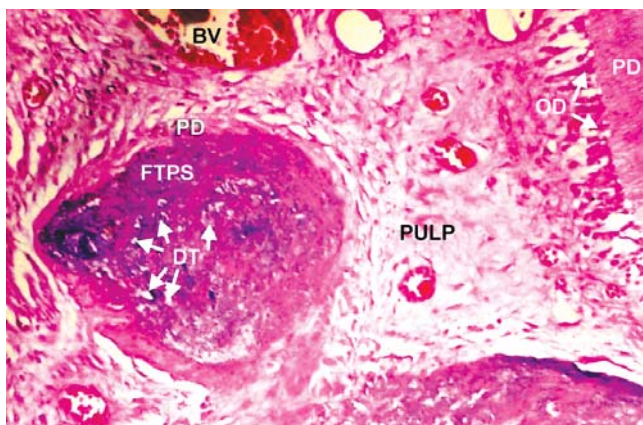


Figure 16.3: Free, true pulp stone (FTPS) in pulp chamber showing peripheral rim of pink predentin (PD) and irregularly dilated dentinal tubules (DT) in the blue amorphous central mass. (BV) blood vessels (OD) odontoblast

pulp tissue. Diffuse calcification, which surrounds the blood vessels is known as dystrophic calcification (Fig. 16.6). Diffuse calcification is found in root canal and less often in coronal area while pulp stones are seen mainly in coronal pulp.

AGE CHANGES IN PERIODONTIUM

Periodontium comprises of four tissues, two mineralised and two non-mineralised. Two mineralised tissues are the cementum and alveolar bone while the non-mineralised tissues are the lamina propria of the gingiva and the periodontal ligament.

With aging, a healthier periodontium can result from general good health and good oral hygiene of an individual. A loss of gingival height related to gingival and periodontal disease promotes regressive and destructive changes.

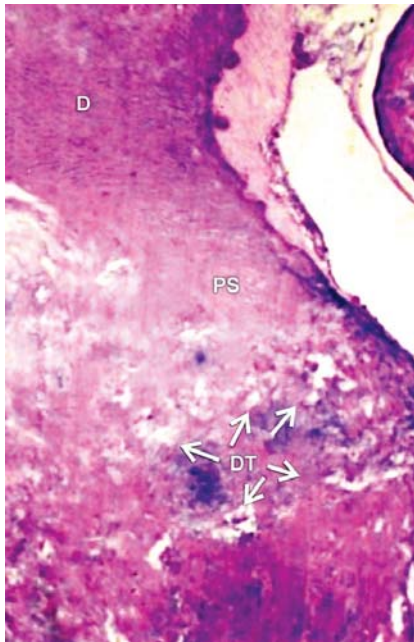


Figure 16.4: True attached pulp stone (denticle): As seen under higher magnification (Fig. 16.3). Peripheral part of pulp stone (PS) where it is arising from dentin shows clear dentinal tubules (DT). The rest of the denticle shows disorganized structure (D) dentin

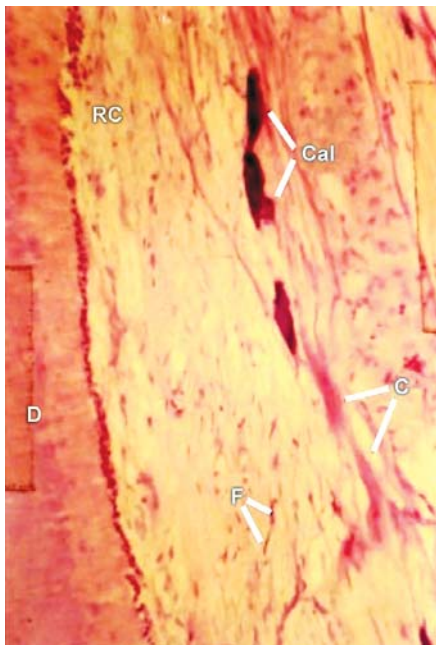


Figure 16.5: Pulp fibrosis and calcification: Decalcified section of tooth showing fibrous tissue replacing the normal loose areolar tissue of root canal. Long pink collagen fibers are traversing the canal longitudinally. Large number of fibroblasts with thin spindle-shaped nuclei can be seen. Areas of focal calcification of fibrous tissues are present. (D) Dentin, (RC) Root Canal, (C) Collagen fibers, (Cal) Calcifications (F) Fibroblasts

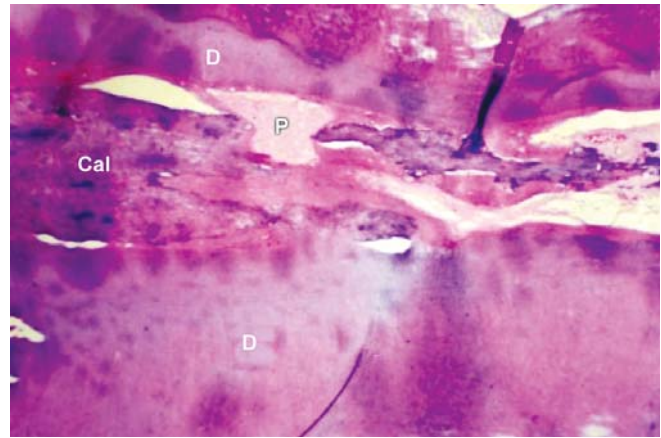


Figure 16.6: Diffuse heavy calcification of the pulp. (D) Dentin, (P) Pulp, (Cal) Calcium deposition. (H & E x 100)

Changes in Gingiva

Some important changes which occur with aging are diminished keratinization, a reduced amount of stippling, increased width of the attached gingiva. There is a decreased connective tissue cellularity, a greater amount of intercellular substances and reduced oxygen consumption.

There is a progressive apical migration of the dentogingival junction due to a gradual loss of the tooth supporting tissue. This results in the exposure of the outer surface of root. This exposure of the cementum to oral environment results in discoloration of root and onset of root caries. Gingival recession also reflects increased tooth mobility due to progressive loss of the tooth supporting tissues. Gradually calcification occurs in the lamina propria of the gingiva.

Changes in Periodontal Ligament

Most of the workers believe that with advancing age there is decreased elasticity in periodontal ligament like all other tissues of the body. But Haim and Baumgartel (1968) observed that aging in periodontal ligament results in a greater number of elastic fibers. There is decrease in vascularity, mitotic activity, fibroplasia and the number of collagen fibers and mucopolysaccharides. Various types of cementicles appear in periodontal ligament. There is increase in arteriosclerotic changes. If there is lowered functional demand; there is reduction in width of periodontal ligament fibers. This may be due to a decrease in strength of masticatory musculature.

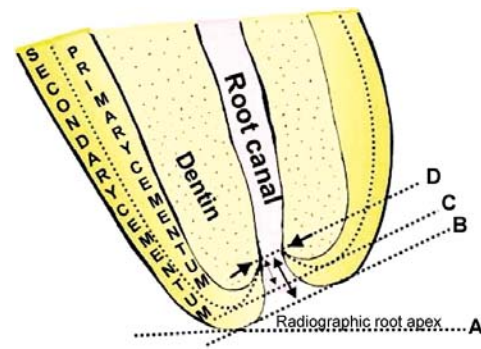
There is reconstruction and reorientation of the periodontal ligament to compensate for the mesial drift, as age advances.

Changes in Cementum

The thickness of the cementum on root surface increases with age. This is due to the cementoid which is laid down in successive lamellae. Deposition of cementoid lamellae is followed by the mineralization of cementum. This indicates that a continuous increase in amount of cementum occurs with advancing age. Irregularity in the surface of cementum facing towards the periodontal ligament also increases with advancing age. As thickness of the cementum increases due to hypercementosis permeability of the cementum decreases. The total width of the cementum at 60 years of age is approximately three times of that, present at ten years of age. With age permeability of cementum diminishes. In a very aged person, the phosphate exchange in the tooth by way of cementum increases to 50% of the total.

With age, as a result of the functional influences on the teeth, the location and shape of the apical foramina undergo changes. Because of mesial migration of the tooth, apex may tilt in the opposite direction. Cementum, by its growth, thickens around the apex and contributes to the length of root (Figs 16.7A to D).

The cementodentinal junction at D is the narrowest point of the root canal. Manipulation at this point meets with greatest resistance, like during condensation filling procedures. With continued cementum deposition apex increases (from C to B in Fig. 16.7), similarly the distance from the radiographic apex, A to D (in Fig. 16.7) increases.



Figures 16.7A to D: Magnified view of root apex of mandibular central incisor at the age of 60 years (A) The level at which in radiograph the root will appear (B) Level of tip of cementum at the age of 60 years (C) Level of tip of cementum at the age of 20 years (D) Narrowest point of root canal at cementodentinal junction

Changes in Alveolar Bone

With advancing age, there are certain changes, which take place on almost all bones of the body. These changes also occur in the alveolar bone. Resorption activity is increased and rate of bone formation is decreased, resulting in bone porosity. With age, socket lining becomes much rougher and it can be seen in histological section as bone with ragged outline (Fig. 16.8). Other changes which occur with aging are osteoporosis and decreased vascularity. There is reduction in metabolic rate and healing capacity. Increased alveolar bone loss in aged may be due to less efficient oral hygiene. Bone loss and pathological migration of teeth may be the result of periodontal disease.

With time and gradual wear, the proximal contact areas of the teeth are flattened and the teeth tend to move

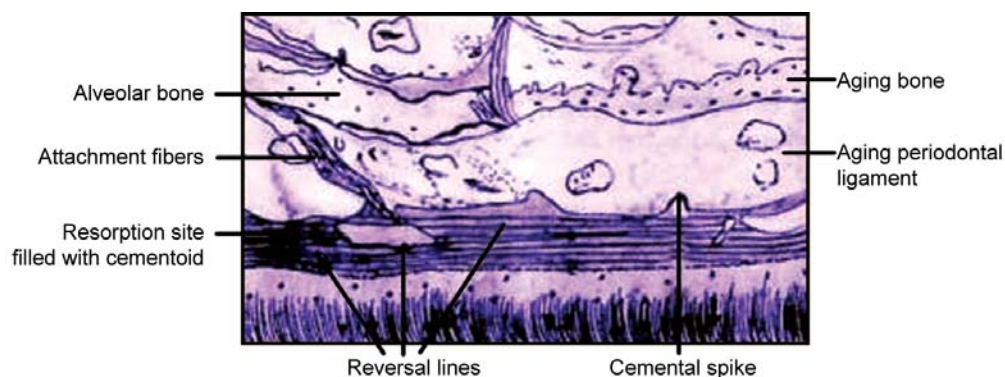


Figure 16.8: Age changes in periodontal ligament, cementum, and alveolar bone. Fiber bundles decrease in number. Excessive deposition around depleted fiber bundles takes place causing cemental and bony spikes

mesially. This is a gradual process. By the age of 40 years, it effects a reduction of 0.5 cm in length of the dental arch from the midline to the third molars.

Alveolar bone is reconstructed in response with the physiologic mesial migration of the teeth.

AGE CHANGES IN ORAL MUCOSA

Oral mucosa found in elderly person often has a smoother and drier surface. It becomes atrophic or friable. The epithelium becomes thinner and more fragile with age. There is variation in size and shape of both cells and their nuclei and flattening of the epithelial connective tissue interface. This results from flattening of the epithelial ridges. The dorsum of the tongue may show a reduction in the number of filiform papillae. So foliate papillae become prominent. Such changes increase with a deficiency of iron and vitamin B complex.

Vascular changes associated with aging may be quite prominent, because of the development of vascular nodules on the lips and cheeks, and the development of nodular varicose veins which are present on the undersurface of the tongue. This is known as cavier tongue. Sebaceous glands (Fordyce's spot) (Figs 9.45 and 9.76) of the lips and cheeks also increase with age and the minor salivary glands show marked atrophy with fibrous replacement.

In the lamina propria of the oral mucosa, there is decreased cellularity and an increase in the amount of highly cross-linked collagen associated with aging. Due to attrition, abrasion and erosion, or due to a combination of any of these regressive changes, hypersensitivity is present in elderly patients.

With aging, a reduced amount of stippling, increased width of attached gingiva, decreased connective tissue cellularity and increased amount of intercellular substances are found in the gingiva. Keratinization of lip and cheek mucosa has also been reported as age advances.

Other age changes in oral mucosa include atrophy of the connective tissue with loss of elasticity and increase in the number of mast cells. There is also a decrease in protein contents.

Clinically, certain features of aging which reflects in oral mucosa of elderly patients especially post menopausal women are, dryness of the mouth, burning sensation and abnormal taste. The ability to repair is reduced and the length of the healing time is increased.

AGE CHANGES IN SALIVARY GLANDS

Salivary glands become hypoactive with age. This often results in painful, burning sensation of the oral mucosa. Histologically, fatty degenerative changes, fibrosis and the progressive accumulation of lymphocytes in salivary glands are associated with aging.

Oncocytes and epithelial cells have marked granularity and acidophilia when seen under light microscope. These also represent the age change. Ultrastructurally, an accumulation of structurally altered mitochondria and oncocytes is found in acini, in the intercalated and the striated ducts of the salivary glands.

With age, reduced salivary flow results in inflammation and ulceration of the oral mucosa. So patient complains of dryness and increase in viscosity of saliva. There is gradual atrophy of the minor salivary glands and less activity of the major salivary glands.

AGE CHANGES IN TEMPORO-MANDIBULAR JOINT AND OCCLUSION

With advancing age, if loss of teeth occurs, it causes stress on the capsular ligament of the temporomandibular joint. As a result capsular ligament gets lengthened. If these missing teeth are not replaced, there is reduction in masticatory efficiency. In aged persons, reduction in masticatory efficiency may also be due to loose teeth, poorly fitting dentures or an unwillingness to wear dentures.

As age advances the articular eminence gradually wears and flattens out and condylar head rests in more backward position in glenoid fossa. The articular disc also degenerates and may cause TMJ pain and headache. There is gradual atrophy of muscles of mastication as age advances.

Due to the age changes, sometimes (more commonly in females) condyle of mandible after wide opening of the mouth fails to go back and the patient is unable to occlude the teeth and masticate food. In such cases, downward and backward force is applied with the thick cotton and gauze padded thumbs at retromolar areas on both the sides by the clinician. The condyles go back with a sudden forceful jerk. To protect the thumbs being bitten in between the teeth they are covered and protected by thick cotton and gauze wrapped around them.

The floor of the maxillary sinus comes close to oral cavity with a thin layer of intervening bone. Similarly

mental foramen comes close to upper border of mandible and may cause pain during mastication specially with artificial dentures as the nerve fibres coming out of mental foramen get pressed.

In old age, incisal edges and cusps are worn off and may cause edge to edge bite, thereby decreasing the chewing efficiency of the food. Chronic traumatic occlusion develops from gradual changes in the occlusion produced by tooth wear, drifting movements and extrusion of teeth.

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Methods Used in Preparation of Specimen for Histological Study

- Introduction
- Types of microscopy
- Preparation of specimen for microscopy
 - Paraffin embedded sections
- Calcified tissues
 - Decalcification
 - Ground section
- Electron microscopy
- Exfoliative cytology

INTRODUCTION

The microscopic examination of tissue sections and their proper preparation is essential in the study of oral tissue morphology. Therefore, a basic knowledge of the various types of microscopes and histologic techniques is important to learn. This helps to study the structure and function of oral tissues.

For various types of microscopy, the fundamental methods of tissue preparation are same as for light microscopy, but there are differences in some specific procedures. Depending upon the nature of the specimen and the type of the microscope to be used for examination, the basic procedures are modified.

TYPES OF MICROSCOPY

The main instrument used in the study of histology is bright field light microscope also called light microscope (Fig. 17.1). Microscope presents an enlarged image of the specimen. Tissues and cells which cannot be seen with naked eye can be seen with the help of the microscope. The main components of a microscope are as follows:

1. *Light Source:* Light is provided through a 6 volt 20 watt tungsten bulb. Bulbs with brighter illumination are used in advanced research microscopes. Reflected light from sun or tube light using reflecting mirror is still used but is not satisfactory.
2. *Condenser:* The parallel light rays emerging from the light source are converged onto the object with the help of a lens called Condenser.
3. *Mechanical Stage:* A rectangular stage with a large hole in the center and clips to grip the object or glass slide is placed above the condenser. Light passes through the hole and illuminates the object under study.
4. *Objective:* Objective is a compound lens and is made of series of convex and concave lenses. It enlarges the image of the object and transmits to the eyepiece. The generally used magnifications of objective are 6x, 10x, 40x and 100x.
5. *Eyepiece:* The eyepiece lens gathers the image transmitted from the objective and relays to the eye of the observer. It also further magnifies the image. Microscope is monocular if only one eyepiece is present and binocular if two eyepieces are attached for use of both the eyes simultaneously (Fig. 17.1).

Microscopes used for specialized study of the cells include dark field microscope, phase contrast microscope and fluorescence microscope. Very minute subcellular structures can be studied with the help of electron microscope (Fig. 17.2).



Figure 17.1: Binocular bright field light microscope used for examination of histological sections



Figure 17.2: Transmission electron microscope used to study the subcellular structures at very high magnifications upto one lac times

PREPARATION OF SPECIMEN FOR MICROSCOPY

The basic techniques in the preparation of tissue sections for various types of microscopy are almost the same, with certain differences in some procedures. Tissue preparations for the light microscopic study are made thin enough to transmit light, and contrasts is given to the components using dyes or stains for easy differentiation. The tissue for histologic study is fixed in a protoplasmic coagulating solution, dehydrated and cleared in organic solvents, embedded in paraffin, cut into thin sections on a microtome, stained, mounted and examined under a specific microscope.

Four techniques for oral tissue preparation are usually used for microscopic examination. These are as follows:

1. *Paraffin embedded section of soft tissues:* This is the most common technique used for soft tissue study under light microscope. The tissue is cut into 4 to 5 microns thick sections with a microtome, which are then taken on microscopic glass slides, stained and then covered with cover slip.
2. *Decalcified sections for hard tissue:* The hard tissue specimens are first decalcified and embedded in paraffin wax. They are then sectioned on a microtome.
3. *Ground sections for calcified tissues:* The calcified tissues like bone and teeth are sliced and ground to a thin section of about 30 to 50 microns thickness, first by a revolving carborundum disk and then by grinding on carborundum lathe wheel or flat stones of gradually increasing fineness.
4. *Frozen sections for soft tissues:* This technique is used when the tissue has to be examined immediately or when the constituent under examination is sensitive to the agents used for embedding. The fresh, unfixed or fixed soft tissue is then frozen and sectioned with freezing microtome (cryostat) without being embedded in paraffin wax.

Paraffin Embedded Sections

Usage: In preparing sections of the soft tissues such as gingiva, cheek, tongue, lip, salivary glands, etc. that is, the tissues, which are not calcified.

Obtaining the Specimen: The specimen should be removed carefully, without crushing it.

Fixation of the specimen: After obtaining the specimen, it should be immediately placed in a fixing solution. Ten

percent neutral formalin is the most commonly used fixative for oral tissues.

Fixation is important to coagulate the proteins, which preserves the tissue structure and prevents alteration during subsequent treatment. It also makes the tissue more readily permeable to the reagents used. Depending on the size, thickness and density of the tissue and on the type of fixative used, the fixation period varies from several hours to several days. The specimen is then washed overnight in tap water to remove excess of formalin.

Processing of the tissue: Tissue is passed through different solutions to prepare it for embedding in the paraffin wax. This treatment is called processing of the tissue. Three important steps in processing are:

1. Dehydration
2. Clearing
3. Infiltration.

Processing of the tissue can be done in the electrically operated automatic tissue processor (Fig. 17.3) or manually using lid covered glass jar bottles (Fig. 17.4).

Dehydration

During this process water is removed from the specimen, since water is not miscible with paraffin wax in which the tissue is embedded in the final step of processing. Two widely used dehydrating agents are; (i) Alcohol (ii) Acetone. The dehydrating schedules can be either passing the tissues through ascending grades of alcohol concentration from 60 percent, 70 percent, 80 percent, 95 percent, and absolute alcohol or a combination of alcohol and acetone is used.



Figure 17.3: Automatic Tissue Processor: The different chemicals are filled in the glass jars. Two jars are filled with wax and the temperature of wax jars is fixed at 65°C. The tissue samples are automatically transferred from one chemical to another after desired prefixed time set on the dial

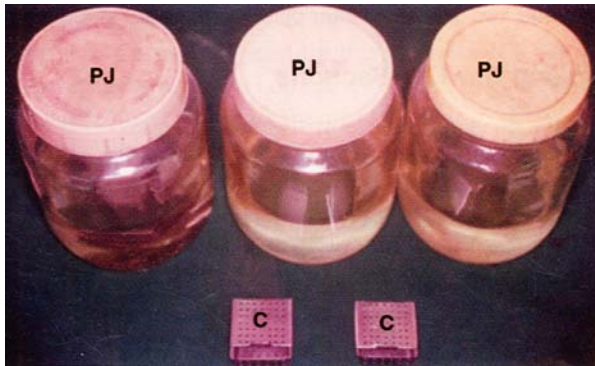


Figure 17.4: Manual processing of tissues: Wide mouthed covered glass processing jars (PJ) filled with different chemical reagents are used for processing. Tissue specimens are kept in the metal perforated capsules or cassettes (C) shown in the foreground of the photographs

Clearing

In this process, tissue is treated with a chemical that increases the refractive index of the tissue to bring it to the near the refractive index of glass. Tissue becomes translucent or clear. Another property of clearing agent is that these chemicals are miscible with alcohol and acetone on one hand and paraffin wax on the other, and thus help in infiltration of tissue by wax. Commonly used clearing agents are; (i) Xylene and (ii) Chloroform, but xylene is preferred and widely used. Chloroform is highly volatile and has anesthetic properties. Extreme care is required in its handling and use.

Paraffin Infiltration of the Specimen

The specimen is infiltrated with paraffin wax after cleaning of tissue with xylene. It is placed in a dish of molten paraffin wax, which has been in an oven at 65°C. To remove all the xylene from the tissue and replace it with paraffin, the specimen is changed to two or three successive dishes of paraffin. The size and density of the specimen determine the oven-time, ranging from a couple of hours to twenty-four hours. Automatic wax baths are convenient and they regulate the temperature correctly (Fig. 17.5). Some laboratories still use wax in glass jars kept in an incubator set at 65°C temperature (Fig. 17.6). In automatic tissue processor, wax baths are integral part of the machine and contain molten wax (Fig. 17.3).

Specimen Embedding

The specimen is embedded in the paraffin wax after it has been completely infiltrated with paraffin. The specimen is removed from the molten paraffin with warm forceps and placed in the center of a paper box or a small metallic

tray (Fig. 17.7) or rectangle formed by joining two L-molds (Fig. 17.8). The surface of the specimen to be cut is placed facing the bottom of the box, which is then filled with molten wax and wax is allowed to solidify. It is then immersed either in cool water or kept in the freezing chamber of the refrigerator to harden the wax. The hardened paraffin block is then separated from the paper box or metallic tray or L shaped molds and is mounted either on a paraffin coated wooden cube or on a metallic block holder (Fig. 17.9). The tissue is ready for preparation of thin sections.



Figure 17.5: Automatic wax baths used for infiltration of wax into the tissues. The temperature of the wax baths is set at 65°C so that wax remains melted

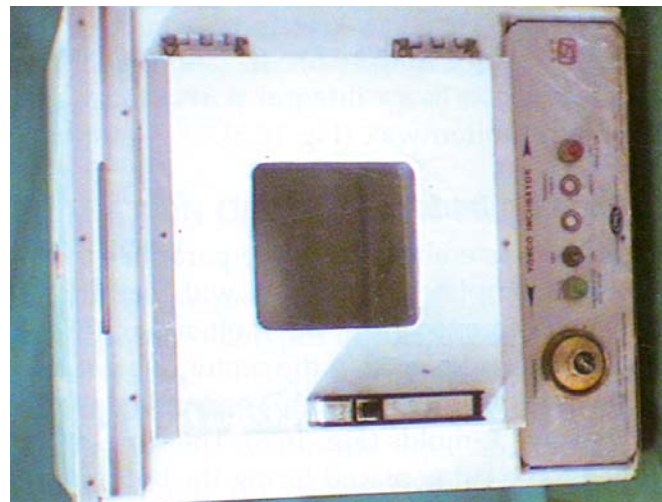


Figure 17.6: Wax can be kept in glass jars and placed in the incubator set at 65°C so that wax remains molten and can impregnate the tissue

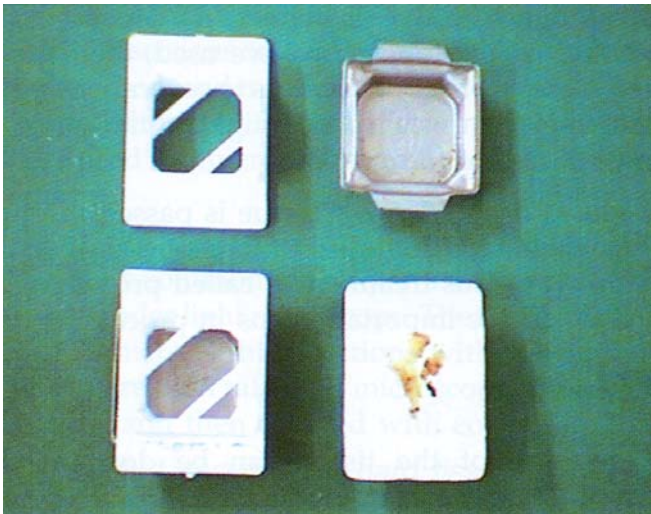


Figure 17.7: Preparation of Paraffin wax block: Molten wax is poured into metallic tray after the tissue has been kept at the bottom of the tray and plastic mold has been kept at the top

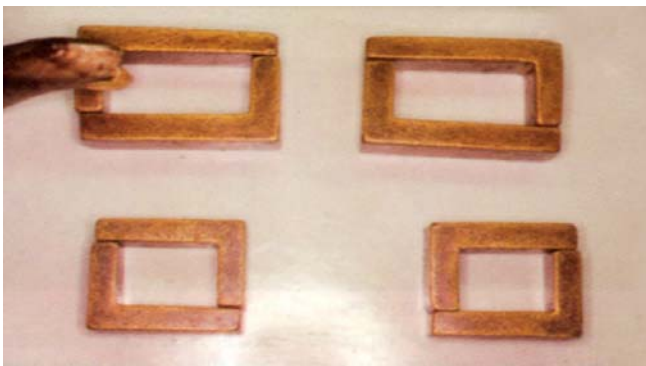


Figure 17.8: Preparation of Paraffin wax block: Two L- shaped brass moulds are kept together to form a rectangle and molten wax is poured into the mould after placing the tissue in the center

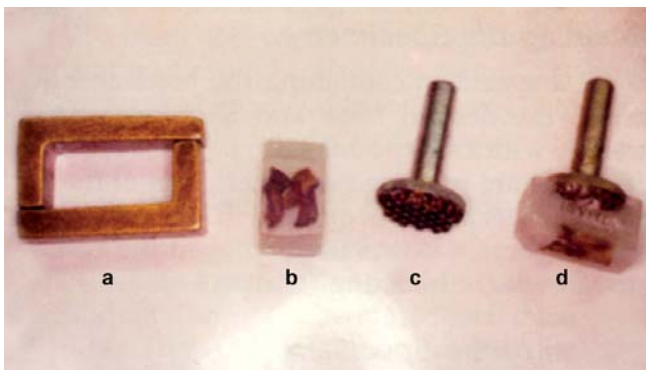


Figure 17.9: Paraffin wax block (b) after detaching from the L-mould (a). The wax block is then mounted on the metal block holder (c). Paraffin wax block is shown mounted on the holder (d)

Sectioning of the Specimen

A precision rotary microtome is used to cut the specimen (Fig. 17.10). The paraffin block attached to the wooden block or metallic block holder is cut into 4 to 5 micron thick sections with a sharp microtome knife.

Picking the Cut Sections

To pick the sections, clean slides are coated with a thin film of albumin adhesive. In a pan of warm water or floatation bath, the paraffin section is floated and the coated slide is slipped under it (Fig. 17.11). This is then lifted from the water with the section. To stick the sections to the slides and to make them dry, the slides are placed on a drying table or slide warming table (Fig. 17.12) regulated at about 42°C.

Staining the Sections

The most commonly used tissue stain for microscopic study is a combination of hematoxylin and eosin, also known as H and E. To stain the sections with H and E, the dried slides are kept vertically in glass staining trays or plastic jars and then passed through various staining dishes containing different reagents (Table 17.1) and (Fig. 17.13). The slides are removed one at a time from the last jar and covered with a mounting medium and a cover glass. Commonly used mounting media are DPX and Canada Balsam. Once the mounting medium has set and hardened, the slides are examined under the microscope. Automatic staining machines are used in some laboratories.

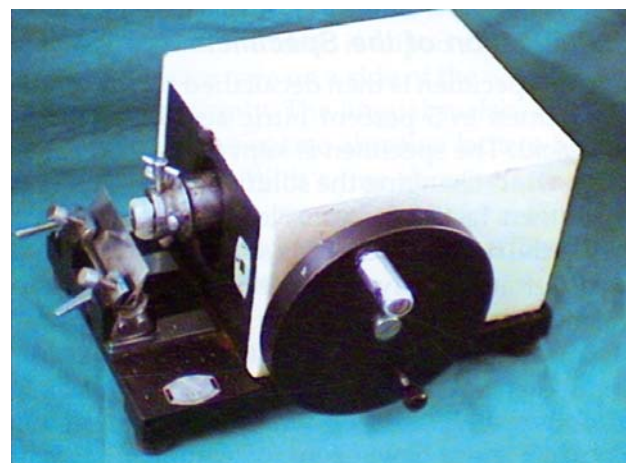


Figure 17.10: Rotary microtome used for cutting 4 to 5 micron thick sections of the tissue specimens



Figure 17.11: The thin sections prepared on the microtome are floated on water in the floatation bath shown above. The sections are gently picked up on clean glass slides



Figure 17.12: Slide warming table is used to dry the sections so that they get fixed to the slides

CALCIFIED TISSUES

Decalcification

If the specimen contains calcified tissues like bone and teeth, these are first made soft by decalcification. Bones and teeth can not be sectioned on the usual rotary microtome due to hardness and high calcium content. The calcium mineral is removed from the tissue before processing. This process is called 'decalcification'.

Better sections of decalcified bone and teeth are cut when embedded in parlodion (celloidin) than paraffin wax. But parlodion processing is time consuming and expensive, therefore decalcified tissues are embedded in paraffin wax in most of the laboratories. Hence, parlodion embedding details have not been described.

Table 17.1: Reagents used for staining of sections the time period required and their actions

Reagent	Time period required	Action
1. Xylene	2 minutes	Removes paraffin
2. Absolute alcohol	2 minutes	Removes xylene
3. 95% alcohol	1 minute	Hydration
4. 80% alcohol	1 minute	Hydration
5. 60% alcohol	1 minute	Hydration
6. Distilled water	1 minute	Precedes stains dissolved in water
7. Hematoxylin	3-10 minutes	Stains nuclei
8. Water	Rinse	Rinses off excess stain
9. Acid alcohol	2-10 seconds	Differentiates, nuclei retains stain
10. Running tap water	2-5 minutes	Makes nuclear stain blue, removes acid.
11. 80% alcohol	1 minute	Partially dehydrates
12. 95% alcohol	1-2 minutes	Precedes stains dissolved in alcohol
13. Eosin	1-2 minutes	Stains cytoplasm and intercellular substance
14. 95% alcohol	Rinse	Removes excess stain
15. 95% alcohol	Rinse	Removes excess eosin
16. Absolute alcohol	1 minute	Dehydrates
17. Absolute alcohol	2 minutes	Dehydrates
18. Xylene	2 minutes	Removes alcohol and clears
19. Xylene	2 minutes	Clears the section



Figure 17.13: Staining of histology sections: Lid covered plastic jars are used for staining the histologic sections fixed on glass slides. The glass bottles in the back row contain hematoxylin (H) (blue eye) and eosin (pinkish orange dye). Plastic jars are used for staining. Front row shows DPX mountant, coverglass (CG), forceps (F) and stained cover glass mounted sections (S)

Obtaining the Specimen

To get a specimen containing the bone and dental tissues, the desired bone and teeth are sectioned carefully with a sharp bone saw and a scalpel. Smaller specimens are

preferred as penetration of the fixing solution to its center is quicker. To include the pulp in the section, a bur is used to open the root apex through which the fixing solution will enter.

Fixation of the Specimen

The obtained specimen is quickly rinsed in running water and placed immediately in 10 percent neutral formalin for fixation. It is kept for about a week in the formalin but smaller pieces need lesser time for fixation.

Decalcification of the Specimen

The fixed specimen is then decalcified by suspending the specimen in 5 percent nitric acid or 10 percent formic acid. The specimen is kept in the acid for 8 to 10 days while changing the solution daily. The specimen is then tested for complete decalcification by piercing the hard tissue with a sharp and fine needle. If it penetrates easily, the specimen is completely decalcified.

Complete decalcification is also tested by keeping the specimen in a test tube containing 5 to 6 ml of nitric acid and adding 1 ml of concentrated ammonium hydroxide and a few drops of saturated aqueous solution of ammonium oxalate. Precipitation of calcium, means traces of calcium are still present. After complete decalcification the tissue is washed with water and processed. Since overdecalcified tissues does not stain well, over decalcification should be avoided.

Washing the Specimen

To remove all the acid, the specimen is washed in running water for at least 24 hours after complete decalcification. After this, specimen is taken through the routine processing and embedded in paraffin, and sections are cut and stained in routine, manner as is done for soft tissues.

Ground Section

Teeth to be used for ground sections should be preserved in 10 percent formalin to prevent them from becoming brittle.

Usage

Ground sections of teeth are essential for the study of enamel and to some extent dentin and cementum. Enamel

has very high content of calcium which makes 96 percent of its weight. Organic matter constitutes only 4 percent of enamel. Hence, enamel gets completely destroyed during decalcification.

Obtaining the Section of the Specimen

To obtain a thin ground section of a tooth, it is cut longitudinally in a mesiodistal plane. A coarse abrasive carborundum disc is attached to a dental micromotor (Fig. 17.14). The tooth is held securely in the fingers while water is sprayed on the tooth. The buccal surface is applied firmly to the rapidly rotating carborundum disc. A fine-abrasive lathe wheel is then used to ground the tooth.

So as to direct the other side outward, a 13-mm adhesive tape is wrapped round a wooden block. The ground surface of the tooth is wiped dry and pressed onto the adhesive tape on a side of the wooden block so that it sticks firmly. The lingual surface of tooth is then applied to the coarse abrasive lathe wheel and ground to 0.5 mm. With a fine abrasive lathe wheel the section is further ground to a thickness of 50 to 70 microns. Ether is used to remove the adhesive tape from the finished ground section.

Then the section is further ground on a fine and smooth flat carborundum stone slab to make it thinner upto the desired thickness of 30 to 40 micron. Ground sections can be prepared on grinding stone slabs of various grades using pumice and water paste (Fig. 17.15). The section is then dried for several minutes.

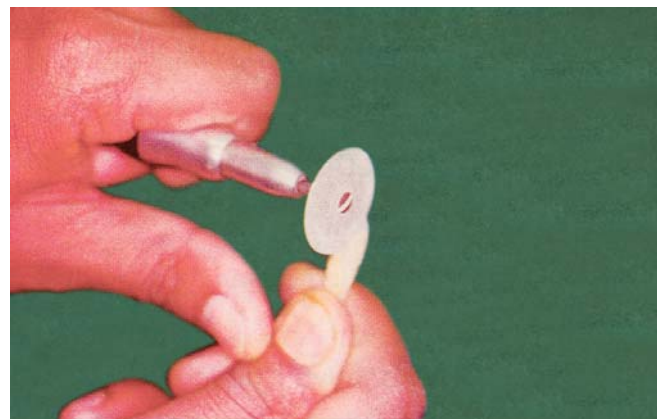


Figure 17.14: Making thin slices of tooth using carborundum disc before grinding on lathewheel or stone



Figure 17.15: Grinding of cut section tooth using stone slab for preparation of thin ground sections

Mounting the Ground Sections

The section is lifted with a camel's hairbrush and placed on a drop of mounting medium placed on a glass slide. Another drop of mounting medium is put on the section and covered with a cover glass. The slide is now ready for examination under the microscope.

Frozen Sections

A cryostat or freezing microtome is used to cut fixed or unfixed soft tissues, which have been frozen by either liquid nitrogen carbon dioxide gas into 10 to 15 microns thick sections (Figs 17.16A and B). Frozen sections are useful for immediate examination of a specimen for rapid diagnosis in cancer cases as they can be quickly prepared. Tissues whose constituents are sensitive to the reagents used in paraffin embedding, e.g. enzymes and fats are studied as frozen sections. Frozen sections are also required for fluorescence microscopy of the tissues since paraffin sections can not be used for fluorescence. Fluorescent staining is carried out with the help of FITC conjugated antibodies (Fig. 17.17) and sections are examined under fluorescence microscope (Figs 17.18).

ELECTRON MICROSCOPY

Very minute details of the cells cannot be studied by light microscopy due to the limitation of magnification and resolution. Maximum magnification of only 1600 times can be achieved with bright field light microscope.

To study subcellular structure, electron microscope is used which gives magnification upto 1,00,000 times. Structures as minute as ribosomes, mitochondria,



Figures 17.16A and B: Cryostat Microtomes: A. Outside view of cryostat used for cutting the frozen sections. B. Inside view of Freezing chamber of cryostat maintained at temperature of -25°C in which microtome is fitted for cutting frozen sections

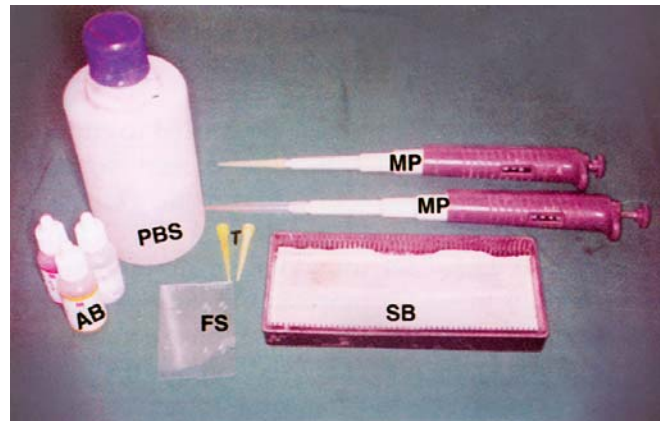


Figure 17.17: Staining of frozen sections for fluorescence. The frozen sections are treated with very small quantity (50 microliter) of antibodies with the help of micropipettes and incubated in the wet chamber. The antibodies are washed with phosphate buffered saline (PBS). (MP) Micropipettes, (T) Tips, (AB) Antibodies, (FS) Frozen sections mounted on slides, (SB) Staining box.

endoplasmic reticulum, virus particles can be examined with the help of an electron microscope.

Electron microscope can be of following two types; (1) Transmission electron microscope (Fig. 17.2), which provides cellular details in the cell. (2) Scanning electron microscope which provides surface structure of the cells (Fig. 17.19).

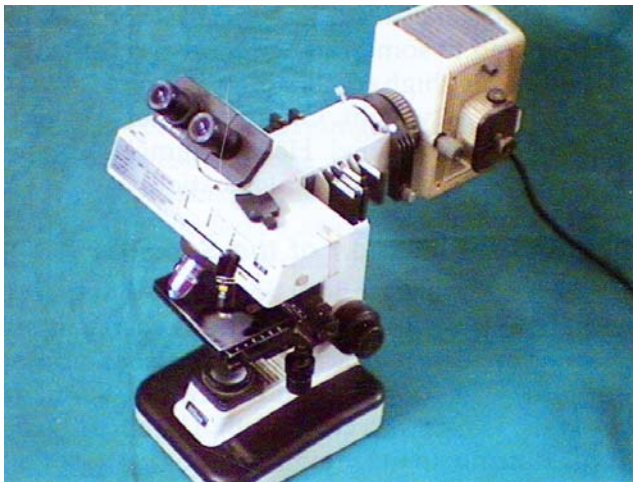


Figure 17.18: Fluorescence Microscope: Fluorescence microscope is equipped with Ultraviolet blue filter and high voltage mercury lamp



Figure 17.19: Scanning electron microscope used to study surface characteristics of the cells at very high magnifications upto 100,000 times

EXFOLIATIVE CYTOLOGY

Since exfoliative cytology (cytology of shed or scraped cells) is used frequently in premalignant and malignant conditions, it is better to understand the procedure of exfoliative cytology.

Preparation of Exfoliative Cytology Smear

A drop of saline is placed on a clean glass slide. With the help of a wooden or plastic or metal spatula, mucosal surface of cheek and tongue or palate, etc. is scraped gently and the whitish fluid on the surface of the spatula is mixed with saline and spread on the slide with a smooth circular movement. The smear is then dried and stained.

Staining of cytology Smear

The most commonly used stain for cytological preparations is May-Grünwald Giemsa stain (Fig. 17.20).

Procedure

Dried smear is fixed for 1 minute with absolute alcohol. 1:10 freshly diluted solution of Giemsa stain is flooded over the smear and left for 10 to 15 minutes. Smear is then washed with tap water, dried and covered with mountant and cover glass and examined under the light microscope (Fig. 17.20). Cells in saliva and scrape from oral mucosa can be clearly seen with exfoliation cytology (Fig. 17.21).

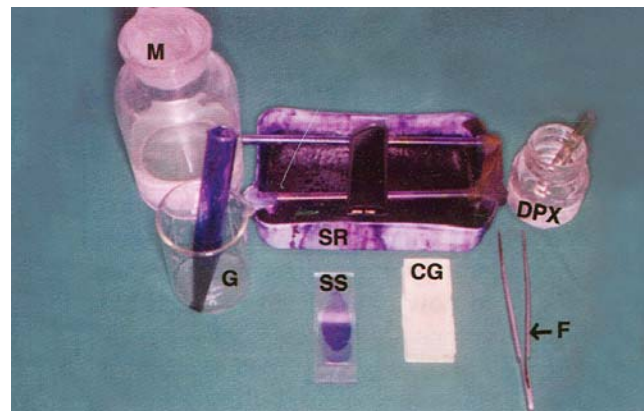


Figure 17.20: Staining of cytological specimens: Fresh Giemsa stain is diluted in the test tube and poured over the methanol fixed smear. Then it is covered with thin cover glass shown in the foreground with the help of DPX mountant. (M) Methanol, (SR) Staining rack, (G) Giemsa Stain, (SS) Stained slide, (CG) Cover glass, (F) Forceps

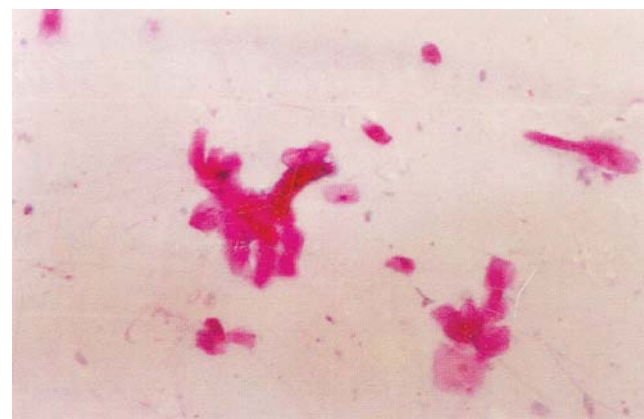


Figure 17.21: Exfoliative Cytology: Cells in saliva and scrape from oral mucosa are polyhedral in shape. Their cytoplasm is abundant, pink and nuclei are small, round, pyknotic. Giemsa Stain (x 200)

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Advanced Techniques in the Study of Oral Tissues

- Introduction
- Histochemical and advanced techniques
 - Immunohistochemistry
 - In situ hybridization
- Chemical components of oral tissues
 - Connective tissue

- Cells and fibers
 - Preparation of oral tissues for histochemistry
 - Histochemistry of oral soft tissues
 - Histochemistry of oral hard tissues
 - Clinical considerations



INTRODUCTION

Different chemicals have the ability to stain different biochemical substances of the tissues. Various specialised histochemical techniques are based upon the ability to identify these substances. These techniques provide the information that usually is not obtained by biochemical means. Information about normal and pathologic conditions of the tissue can be obtained by histochemical and other advanced techniques.

HISTOCHEMICAL AND ADVANCED TECHNIQUES

Usual structures of the oral tissues can be studied in detail using histological methods described in Chapter 17. However, many other advanced techniques are employed for understanding the biological processes involved in the functioning of the cells. The biomolecules which play important role in normal tissue development, functioning and pathologic conditions can be detected by these techniques.

The most commonly used methods are biochemical and histochemical techniques. Biochemical investigations are useful in determining normal and abnormal concentration of biomolecules, enzymes, etc. However, these do not provide *in-situ* information in relation to structural changes. Histochemical techniques are of immense importance in providing insight into the biochemical changes taking place inside the cell and their relationship with the structural components. However, the preparation of the tissue like fixation and processing is much more demanding as chemical nature of the reagents, pH of the solution, temperature of the reaction, etc. have bearing on the elucidation of the biochemical processes undergoing in the cells.

In recent years newer techniques have been developed which help in the precise localization of biomolecules. Molecular biology is playing increasingly important role and techniques of immunohistochemistry, *in situ* hybridization, immunofluorescence are being used now not only in research but also in clinical applications. Development of monoclonal antibodies has ushered in a new era of diagnostic techniques and of localizing biomolecules of interest.

Immunohistochemistry

Immunofluorescence is used to detect the biomolecules, antigens, cytoplasmic fibers, nuclei or any structure of

interest. Monoclonal antibodies developed against the targetted antigens are used and their reaction is elucidated with the help of fluorescence probes like fluorescein isothiocyanate (FITC) (Figs 18.1 and 18.2). While technique of immunofluorescence is simple and extremely sensitive, special fluorescence microscope is required for visualization (Fig. 17.18). This problem has been overcome by the use of immunoperoxidase technique where marker for elucidating the reaction is enzyme horse-radish peroxidase. Brown insoluble precipitate is formed at the site of reaction and can be visualized with the help of light microscope only (Fig. 18.3). No special microscope is required. Another advantage with this technique lies in its versatility. While immunofluorescence can be used only on frozen tissues, immunoperoxidase can be used on paraffin embedded tissues also and hence this technique

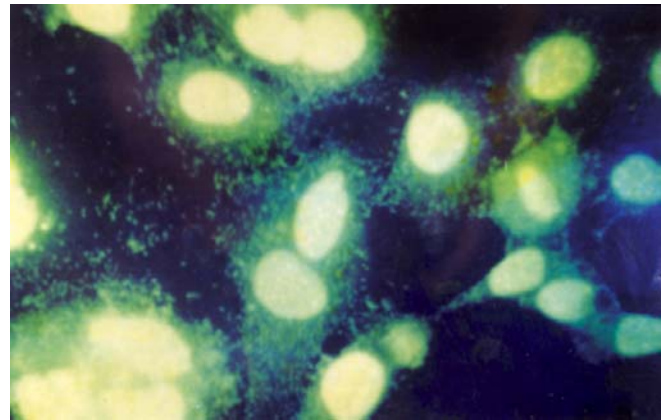


Figure 18.1: Immunofluorescence showing strong greenish yellow fluorescence of the nuclei of epithelial cell monolayer prepared from cultured cells. Cytoplasm shows greenish fluorescence in dark background

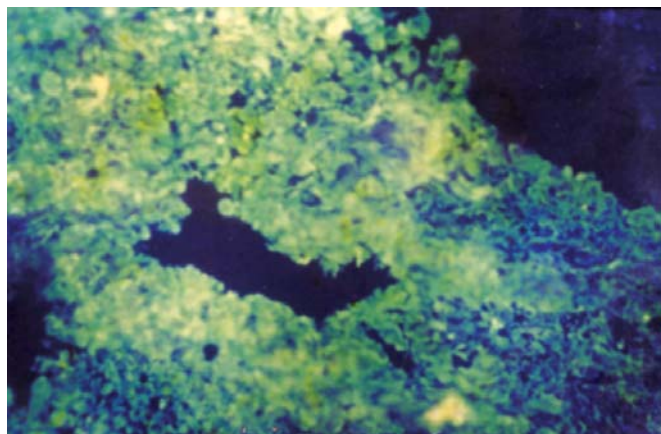


Figure 18.2: Another case of epithelial cell monolayer showing positive fluorescence staining of the cytoplasm. Nuclei are not stained

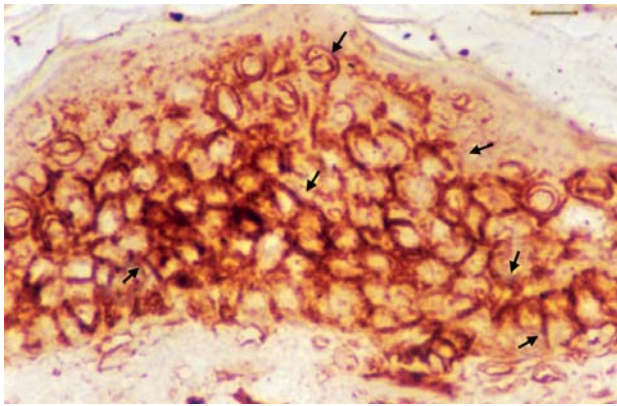


Figure 18.3: Immunoperoxidase staining of oral epithelium showing positive staining of cytoplasmic membrane indicated by arrows in a case of oral pemphigus

has wider clinical application. Sections for immunofluorescence have to be examined immediately and cannot be preserved but immunoperoxidase section can be mounted and preserved for long time. Section or tissues with immunoperoxidase reaction can be studied with electron microscope also.

In Situ Hybridization

In situ hybridization is used to investigate nucleic acids of DNA as well as RNA. This technique utilizes hybridization of synthetic nucleotide sequences with complementary molecular sites in the tissue sections. The hybridization can be detected with the help of fluorescent dyes in FISH or with enzymatic probes like in immunoperoxidase or with radio-isotopic molecules like P^{32} , H^3 or S^{35} . Polymerase chain reaction (PCR) is the last technique which used the principle of amplification of the nucleic acid sequences of the cell. Microphotocell counter, double beam recording, microdensitometry, scanning and integrating micro-densitometry are the techniques used for the quantitative analysis of histochemical reactions.

The qualitative and quantitative analysis of the inorganic components of hard tissues like enamel, dentin and bone can be done with the help of interference microscopy, X-ray diffraction, X-ray spectrophotometry, laser spectroscopy and elemental analysis. Techniques of polarized light and phosphorescence using tetracycline binding have been used in the study of calcium metabolism in bone, dentin and enamel. Scanning electron microscopy (SEM) is used in the analysis of changes in the bone

architecture. Confocal laser scanning microscope can produce three-dimensional image with submicron spatial resolution. Radioautographic techniques elucidate the chemical substance uptake by metabolic pathways of different tissues in different regions of cytoplasm.

CHEMICAL COMPONENTS OF ORAL TISSUES

The oral tissues are composed of connective tissue, epithelial lining and associated muscle fibers.

Connective Tissue

Connective tissue is of mesenchymal origin and consists of various types of cells and fibres embedded in amorphous semigel and colloidal ground substance. The ground substance is made of proteoglycans and glycoproteins secreted by connective tissue cells like fibroblasts and mast cells (Figs 18.4 and 18.5). Proteoglycans can be sulfated like chondroitin sulfates, keratan sulfates and heparin sulfates or can be nonsulfated like hyaluronic acid.

Glycoproteins are protein molecules with lesser number of carbohydrate moieties than in proteoglycans. Some of the glycoproteins well-studied include fibronectin, laminin, chondronectin and osteonectin. These glycoproteins help in cell attachment to the extracellular matrix and thus are responsible for the maintenance of normal cell morphology and also control cell function. Proteoglycans are important for growth and motility. Proteoglycans are of two types. Aggregating proteoglycans

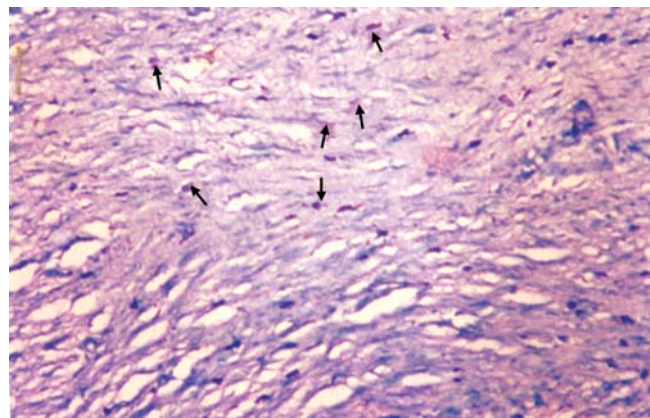


Figure 18.4: Mast cells indicated by arrows can be picked up very easily in the connective tissue using toluidine blue Stain x 40

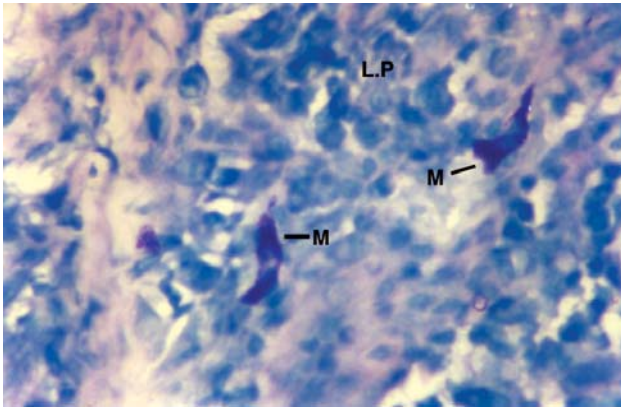


Figure 18.5: Mast cells (M) show metachromasia and cytoplasmic granules stain purple while rest of the cells of the lamina propria (LP) of oral mucosa stain blue. Toluidine blue stain x 400

and non-aggregating proteoglycans. Decorin, fibromodulin, perlecan, agrin and syndecans are non-aggregating proteoglycans. Changes in proteoglycan and glycoprotein components can be seen in pathological conditions like inflammation, early stages of wound healing and cancer.

Cells and Fibers

Fibroblasts

Fibroblasts are the common cells in the connective tissues and elaborate the glycoproteins, proteoglycans and fibrous components of the ground substance, Fibroblasts play important role in wound healing, inflammatory process and developmental process.

Collagen and reticular fibers are elaborated by the fibroblasts and these fibers stain positively for glycoproteins with silver stains and periodic acid -Schiff (PAS) method. Elastic fibers are also elaborated by fibroblasts and stained by aldehyde fuchsin, resorcin fuchsin and orcein dye.

Epithelial cells show presence of glycogen in superficial layers in addition to the proteins and carbohydrates in cytoplasm (Fig. 18.6). The basement membrane or basal lamina is composed of type IV collagen fibrils, laminin, fibronectin and proteoglycans (Fig. 18.6). Salivary glands produce mucins or mucoids which are extremely rich in proteoglycans and glycoproteins. The glycosaminoglycan content of these mucins determines the staining reaction (Figs 18.7 and 18.8).

Histochemistry is the main tool for investigating enzymatic reactions in the cells. The most commonly

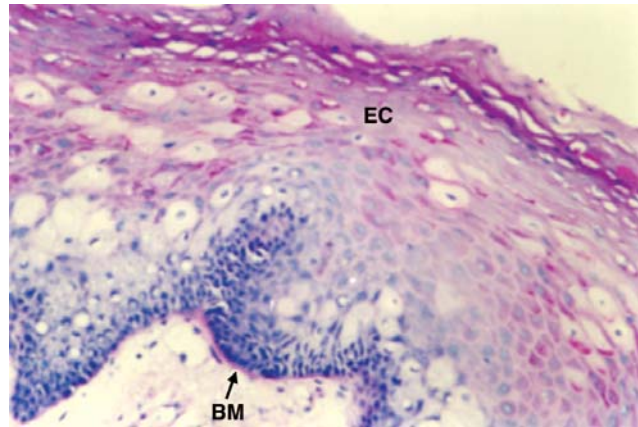


Figure 18.6: Periodic Acid Schiff (PAS) Staining of oral mucosa: Periodic acid schiff stain gives positive magenta color reaction with glycogen present in squamous epithelial cells and polysaccharides present in the basement membrane. (EC) Epithelial cells, (BM) Basement membrane (x 500)

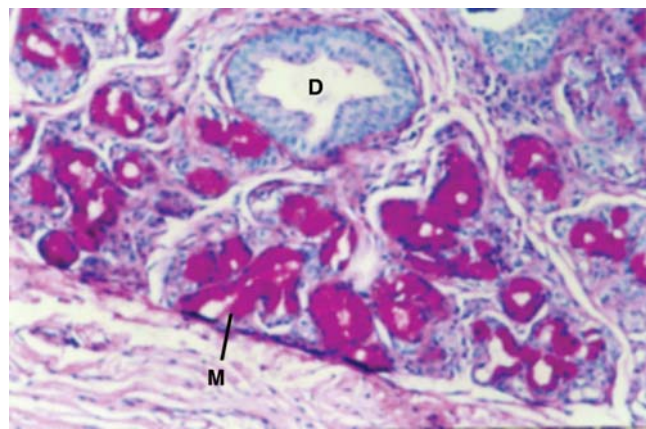


Figure 18.7: PAS stain of mucus salivary gland: Periodic acid schiff stain gives strong positive magenta color reaction with mucin present in the cells. Note the epithelium of the duct is negative. Collagen fibers and ground substance also show positive reaction due to polysaccharide component. (M) Mucus cells, (D) Duct (x 200)

studied enzymes in oral tissue include acid and alkaline phosphatases, oxidases, dehydrogenases, esterases and other enzymes involved in the metabolic activities of the cells.

PREPARATION OF ORAL TISSUES FOR HISTOCHEMISTRY

While the standard good fixative is 10 percent buffered formalin for most of the reaction, some of the molecules get denaturated at high temperature. Thus, fixation is carried out at cold temperature of 4°C. Some biomolecules

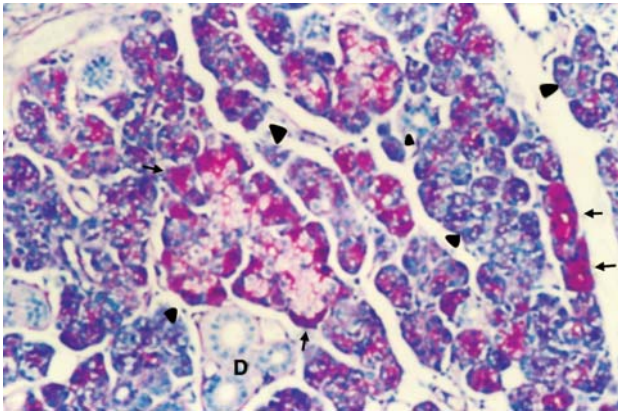


Figure 18.8: PAS staining of submandibular salivary gland shows strong reactivity and magenta stain of mucin-secreting cells indicated by arrows. The serous cells indicated by triangles are either negative or show sparse reactivity. Duct epithelial cells (D) do not stain (x 200)

are preserved better in special fluids like Rossman's fluid for demonstration of glycogen and mucopolysaccharides and Cornoy's fluid for visualization of nucleic acids.

Most of the enzymes get inactivated by fixation, hence frozen section of fresh unfixed tissues are used for their demonstration (Fig. 17.17). Similarly, frozen sections are used for the demonstration of lipids as they get dissolved in chemicals used for processing of tissues.

Histochemical study of hard tissues like teeth and bone requires proper fixation and controlled decalcification as described in Chapter 17. Decalcified ground sections also have been used. Some workers have tried the technique of deorganification (deproteinization) of hard tissues for study under scanning electron microscope.

HISTOCHEMISTRY OF ORAL SOFT TISSUES

Various chemical components of cells can be visualized by staining reactions or formation of insoluble dye or precipitate at the reaction site.

Carbohydrates

The most common staining method used for the study of glycogen, proteoglycans and glycoproteins is Periodic acid-Schiff (PAS) reaction. When periodic acid is treated with leucofuchsin (Schiff's reagent), it oxidises the glycol groups to aldehyde and these in turn produce reddish purple dye product. PAS stain is used extensively to study epithelial mucopolysaccharides, basement membrane and mucins of salivary glands (Figs 18.6 and 18.7). Basement

membrane can also be demonstrated by silver methenamine or other methods involving oxidation of aldehyde groups. Mucicarmine is another histochemical stain employed for demonstrating salivary mucins.

Metachromatic stain toluidine blue is used for the demonstration of proteoglycans and metachromatic reaction (different from original color of stain solution) from purple to red is produced depending upon the degree of polymerization of the dye molecules tagged to the anionic residue of the glycosaminoglycan. Metachromatic reaction is routinely carried out for the demonstration of mast cells (Figs 18.4 and 18.5) which are rich in heparin and intercellular ground substance of young bone and cartilage.

Mucins

Salivary mucins are composed of high molecular weight carbohydrate protein complexes. Two types of mucins are fucomucin, which is rich in α -fucose and sialomucin, which is rich in sialic acid.

The dyes mucicarmine and mucihematin are used for non-specific staining of mucin. Neutral mucins are identified by PAS technique (Figs 18.8 and 18.9). Acid mucins are localized by alcian blue, toluidine blue, colloidal iron and aldehyde fuchsin method. Alcian blue stain is used for the demonstration of types of mucins. Weakly acid sulfated mucins can be demonstrated at pH 2 to 2.8 while highly acidic mucins stain at pH 1 to 1.2.

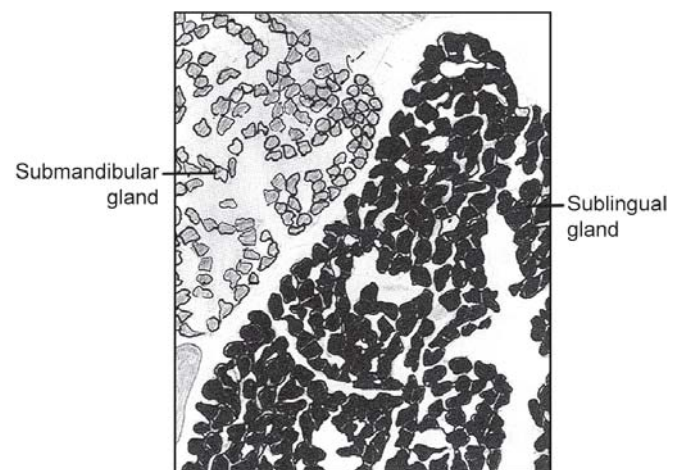


Figure 18.9: PAS reactivity of mucins seen in submandibular and sublingual glands

Proteins

Reactions of amino, iminocarboxyldisulfide and sulfhydryl groups form the basis of histochemistry of proteins. These groups are demonstrated histochemically by ferric-ferricyanide method.

Red colored reaction product is formed on reaction with dinitrofluorobenzene (DNFB) and ninhydrin-Schiff reagent.

Lipids

Frozen sections are used for study of lipids and staining with Sudan black or oil red is employed.

Enzymes

Histochemical demonstration of enzymes is carried out on frozen sections. The reaction is carried out at 37°C, the temperature at which enzymes are active *in vivo*. The substrate on which particular enzyme acts is used. The reaction is made visible with the help of cobalt or lead compounds and end product is insoluble black.

Alkaline Phosphatase

Alkaline phosphatase is present in capillary endothelium of lamina propria (Fig. 18.10). The basement membranes associated with salivary gland acini show high alkaline phosphatase activity.

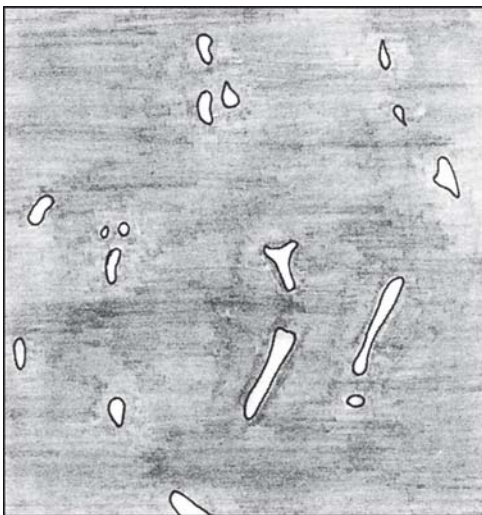


Figure 18.10: Alkaline phosphatase activity demonstrated in capillary endothelium of lamina propria in gingiva revealed by ultraviolet fluorescence

Acid Phosphatase

Acid phosphatase activity is related to the degree of keratinization. It is very high in the zone of keratinization and is low in non-keratinized areas.

Esterase

Some esterase activity is present in superficial layers. High esterase activity is present in salivary gland ducts and also in serous demilunes of the sublingual gland. This is also present in taste buds (Figs 18.11 and 18.12) and mast cells of oral tissues (Fig. 18.13).

Cytochrome Oxidase

Histochemical techniques show low levels of cytochrome oxidase activity in human gingiva. This is localized in

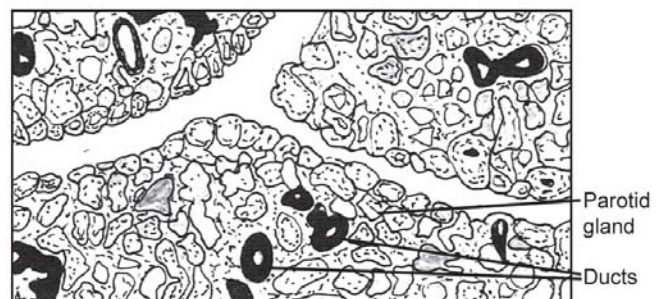


Figure 18.11: Ducts of freeze dried parotid gland showing esterase activity



Figure 18.12: Esterase activity of demilune cells of freeze-dried sublingual gland

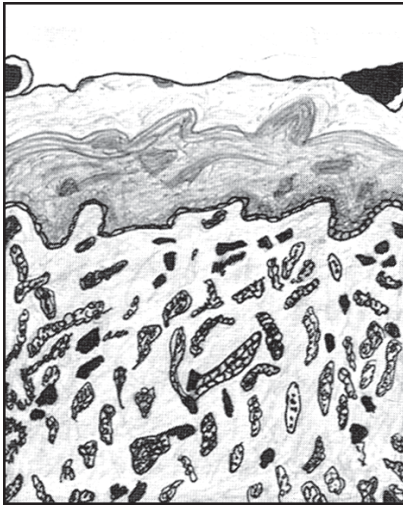


Figure 18.13: Mast cells of tongue incubated with substrate solution containing naphthol AS- D chloroacetate. (As seen under light microscope)

basal layers of the free and attached gingiva, crevicular epithelium and epithelial attachment (Fig. 18.14). An increase in cytochrome activity is observed in chronic gingivitis. In the salivary glands, especially in the duct system, an increase in cytochrome oxidase activity is seen (Fig. 18.15).

β -Glucuronidase

β -Glucuronidase is present in the basal cell layers of oral epithelium. This enzyme is important for cell proliferation, conjugation of steroid hormone and in hydrolysis of conjugated glucuronides.

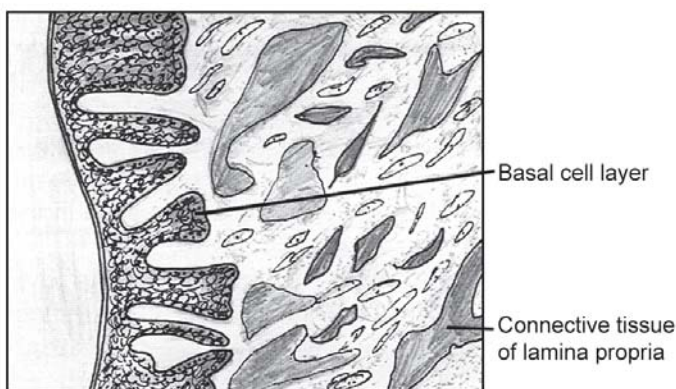


Figure 18.14: Cytochrome oxidase activity seen in basal cell layer and in connective tissue of lamina propria of attached gingiva

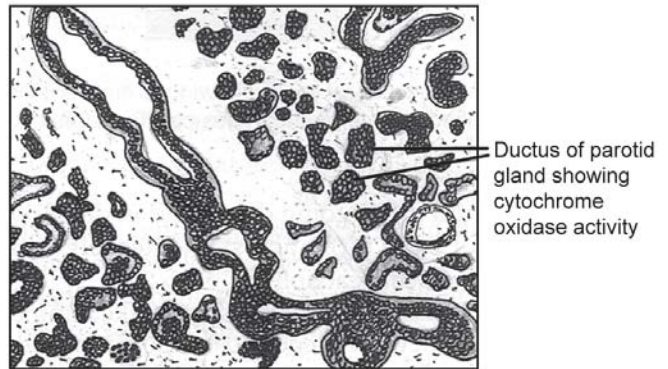


Figure 18.15: Cytochrome oxidase activity demonstrated in parotid gland

Succinate Dehydrogenase and Glucose 6-Phosphate Dehydrogenase

Succinate dehydrogenase is present in the basal cell layers of the gingival epithelium and in ducts of salivary gland. Glucose 6-phosphate dehydrogenase is present in the epithelium of oral mucosa and the level of this enzyme is highly elevated in malignant dysplastic lesions of oral mucosa.

Angiogenic Factor

Angiogenic factor, 67 KD protein was found in the macrophages of inflamed gingival tissue and inflamed tissue of rheumatoid origin.

Differentiation of connective tissue components like collagen and muscle is carried out on the basis of pore size and permeability of fixed tissues and molecular size of the anionic dyes. Collagen has bigger pore size as compared to muscle fibres therefore reacts with dyes of bigger molecular size in the trichrome stains like von Gieson and Masson's trichrome. In von Gieson stain, collagen stains red while muscle takes yellow color (Fig. 18.16). In Masson's trichrome stain muscle stains red and collagen picks up green or blue color depending upon the dye used (Fig. 18.17). Reticular and elastic fibres have strong affinity for silver and are demonstrated by silver impregnation techniques (Figs 18.18 and 18.19).

HISTOCHEMISTRY OF ORAL HARD TISSUES

Carbohydrates

The most commonly used technique in the study of ground substance of bone and teeth is PAS stain to demonstrate

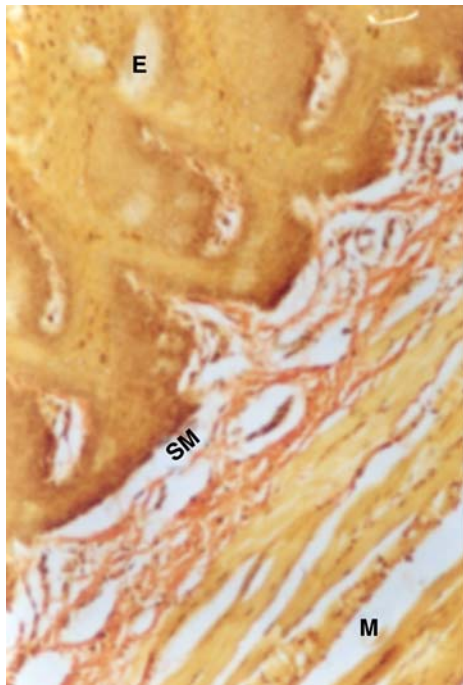


Figure 18.16: von Gieson stain imparts red color to the collagen fibers while muscle stains yellow. This stain is used to differentiate these two tissues. Section of adult tongue show red color of submucosa (SM) and yellow color of muscle (M) and epithelium (E) x 200



Figure 18.17: Masson's trichrome stain of adult tongue showing different colors of fibrous tissue and muscle making the distinction between the two easier. Fibrous tissue of submucosa (SM) takes green color while muscle (M) and epithelium (E) pick up red stain x 100

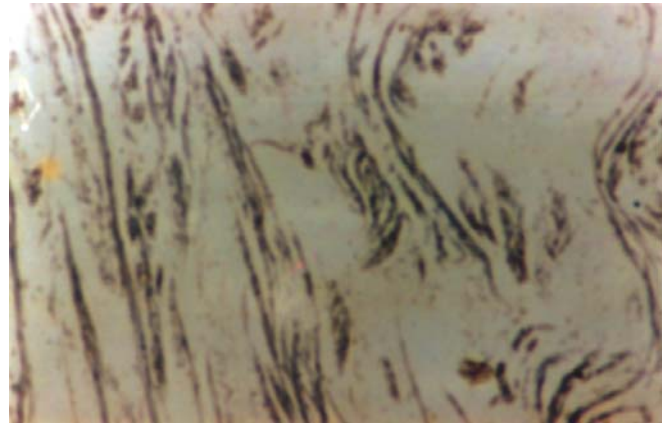


Figure 18.18: Reticulin stain of fetal tongue showing young collagen fibers which have histochemical properties similar to reticular fibers. (Reticulin stain x 200)

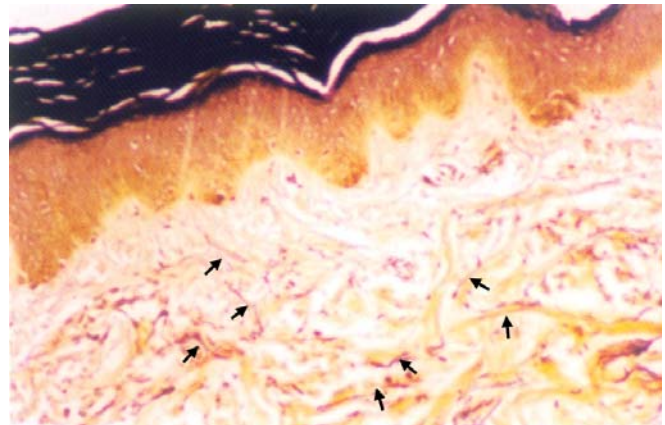


Figure 18.19: Elastic fibers indicated by arrows seen in the cheek skin by Verhoeff's elastic stain x 200

carbohydrates. By this stain, developing and resorbing bone and dentin show stronger PAS reactivity than mature tissue. Poorly calcified dentin matrix in interglobular dentin, dentinogenesis imperfecta and in odontomas show distinct PAS reaction (Fig. 18.20).

Enamel matrix is nonreactive with PAS but enamel lamellae stain intensely (Fig. 18.21).

Proteins

Proteins of dentin undergoing decay and in developmental stages of tooth formation stain intensely with protein histochemical reactions.

Many amino acids or their groups like amino, carboxyl or sulfhydryl are identified by specific protein methods. Some of these methods are used to study teeth and bone.



Figure 18.20: Ground section of tooth showing a distinct PAS reaction exhibited by interglobular less-calcified dentin

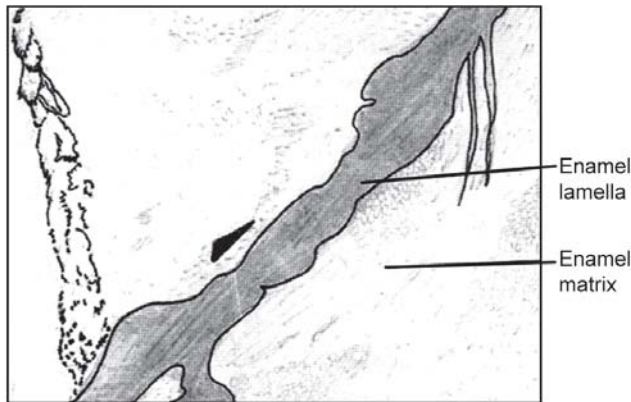


Figure 18.21: Ground section of enamel showing PAS reactivity. Enamel matrix shows no reaction, however, lamella is stained with PAS method

Two of the important methods are dinitrofluorobenzene (DNFB) and ninhydrin Schiff methods.

DNFB reagent reacts and combines with α -amino groups of proteins in tissue sections and forms a pale yellow complex. An azo dye is formed by reduction and diazotization technique to give deep-red color.

The ninhydrin-Schiff method depends upon the formation of imino group. It gets decomposed to ketoacid and forms aldehyde group. These in turn react with Schiff reagent (leucofuchsin) and form a red colored product.

Lipids

Two of the important methods are dinitrofluorobenzene (DNFB) and ninhydrin Schiff methods. Mature dentin shows low lipid content but enamel rod sheaths and odontoblastic processes are strongly sudanophilic due to high phospholipid content. Sudanophilia depends upon the solubility of Sudan dyes with lipids. In a developing

tooth, this dye is seen in zone of mineralization and predentin and in the basal zone of the ameloblasts. This indicates the role of phospholipids in the process of mineralization of dentin and enamel matrix.

Enzymes

Alkaline phosphatase

Alkaline phosphatase activity is seen in endosteum, periosteum, osteocytes, stratum intermedium, odontoblasts, adjoining Korff's fibres and the ground substance of developing molars and incisors (Fig. 18.22). Alkaline phosphatase is thought to be associated with the process of mineralization. It is also associated with osteogenesis and dentinogenesis (Fig. 18.23). The osteoblasts and odontoblasts give an intense staining reaction with alkaline phosphatase.

Acid phosphatase

Acid phosphatase is localized in osteoclasts and odontoclasts lying apposed to the resorbing surface of bone and dentin in hard tissues. It is localized mainly in membrane bound organelles, the lysosomes. Osteoclasts in bone and odontoclasts in resorbing dentin show an intense acid phosphatase activity (Figs 18.24 and 18.25).

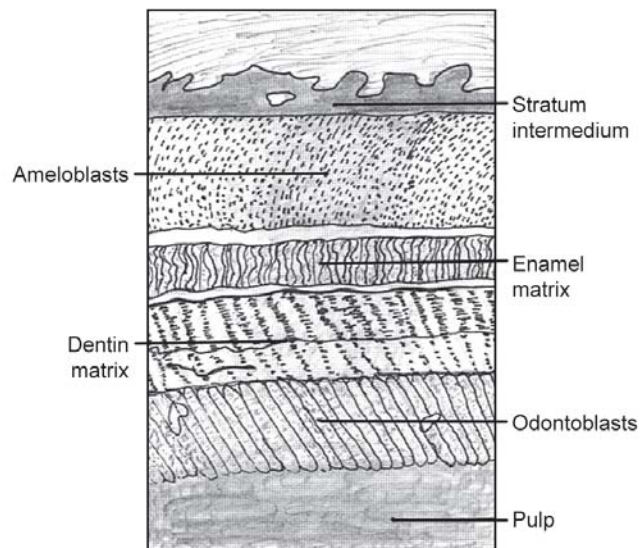


Figure 18.22: Freeze-dried undecalcified incisor showing alkaline phosphatase reactivity of stratum intermedium, odontoblasts and subjacent Korff's fibers

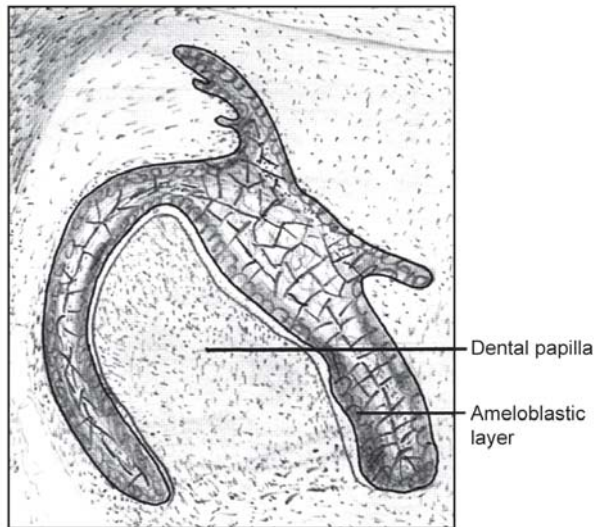


Figure 18.23: Reaction of tooth of the embryo to alkaline phosphatase. No reaction is observed in ameloblastic layer

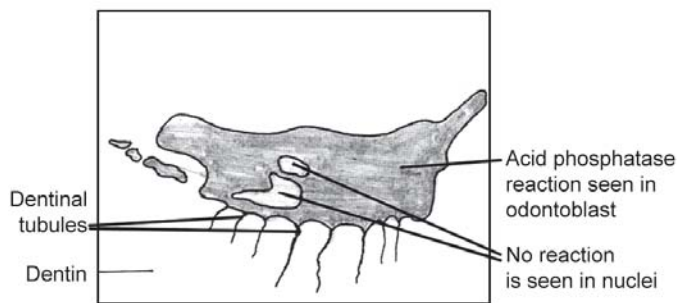


Figure 18.24: Acid phosphatase reaction seen in odontoclast and dentinal tubules of a deciduous tooth undergoing resorption. Cytoplasm shows localized discrete granules of reaction product

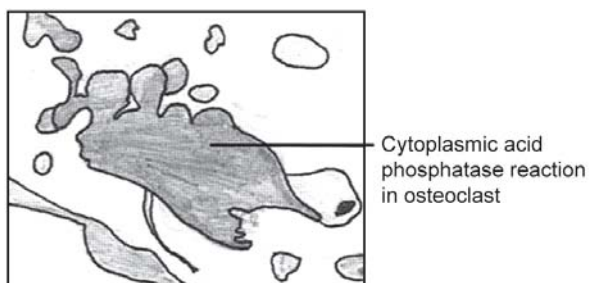


Figure 18.25: Cytoplasmic acid phosphatase reaction shown by osteoclast resorbing bone matrix. It also shows some enzyme activity

Esterase

By the use of specific naphthol esters as naphthol acetate, an intense staining reaction is seen in calcifying matrix of bone and dentin. Esterase activity has also been found in

the cells and microorganisms associated with the formation of calculus deposits on teeth.

Cytochrome Oxidase

Cytochrome oxidase enables cells to utilize molecular oxygen. Its presence reflects the oxygen requirement of the cells and tissues. It also shows their metabolic and physiologic activity. The osteoblasts and osteoclasts show oxidase activity. This is also present in stratum intermedium of molars and incisors.

Other enzymes that are identified by histochemical methods in the hard tissues of the tooth are succinate dehydrogenase and α -ketoglutaric dehydrogenase.

Adenosine Triphosphatase

In early transitional or smooth-ended ameloblasts, AT Pase located at the basolateral membrane and in late transitional or ruffle-ended ameloblast, AT Pase located at distolateral membranes. This distribution of AT Pase in ameloblasts is important for enamel mineralization.

Aminoamidase

Aminoamidases are proteolytic enzymes and demonstrated in the stratum intermedium and odontoblasts during dentinogenesis. Aminoamidases are localized in the macrophages. This enzyme is demonstrated histochemically by azo dye techniques using L-leucyl- β -naphthylamide or DL-alanyl- β -naphthylamide.

Succinate Dehydrogenase

The enzyme is closely associated with cytochrome oxidase and distribution of enzyme is similar to that of cytochrome oxidase. The activity of this enzyme is higher in osteoclasts than in osteoblasts.

CALCIUM-binding sites in Enamel organ.

Calcium was localized in rapidly frozen, freeze-substituted enamel organ tissues and stained with glyoxal bis 2- hydroxyanil (GBHA). Large number of granular calcium-GBHA is located along the lateral plasma membrane of ameloblasts in enamel organ. During enamel matrix formation staining is more intense.

Histochemical Techniques

Fixation

Fixation is important for histochemical study. Fixatives minimize the changes in the reactivity of the cytoplasmic and extracellular macromolecules. Ideal fixative for enzyme and other proteins is formaldehyde. Formaldehyde is used as a 10% solution buffered to pH 7 at temperature of 0° to 4°C. Formaldehyde react with major reactive groups of proteins and form polymeric or macromolecular networks, without affecting their native reactivity to histochemical procedures. Other fixatives are as follows.

Rossman's fluid—This fluid contains formaldehyde, alcohol, picric acid and acetic acid. Fluid is used for visualization of glycogen, glycoproteins and proteoglycans.

Carnoy's mixture—This mixture contains ethyl alcohol, acetic acid and chloroform and is used for visualization of nuclei acids. Acrolein and glutaraldehyde are also used for fixation of tissues.

Post-fixation—Post fixation is a secondary fixation, done on lipid rich tissues and freeze dried tissues.

Imidazole—buffered osmium tetrachloride is post fixative and used for localization of lipids rich in unsaturated fatty acids.

Uranyl acetate is a post fixative, preserve the phospholipid membrane and dehydration with acetone minimizes the extraction of phospholipids.

For cerium, lead—based techniques of enzyme demonstration, Nakan's periodate-lysine – paraformaldehyde and periodate-lysine-glutaraldehyde are superior than classical glutaraldehyde / paraformaldehyde double fixation procedure.

Cytochrome oxidases are highly labile, therefore visualize on fresh frozen sections. Freeze- drying procedure prevent the diffusion and preserve the in- vivo status of tissue macromolecules. Freeze dried tissues were embedded in glycol methacrylate resin without fixation at lower temperature range from 4°C to 20°C.

Freeze-fracture and freeze-etching techniques provide excellent three-dimensional images of the surface of various cell membranes.

In the study of teeth and bone, formaldehyde or glutaraldehyde and ethylenediaminetetraacetic acid are used.

Specific Histochemical Methods

Carbohydrates

Periodic acid-Schiff (PAS) technique is used for detection of carbohydrate. Substitute of Schiff reagent is anthracene-9-carboxyaldehyde carbohydrazone fluorescent reagent.

Thiazine dyes like toluidine blue, azure A and Alcian blue are used for the demonstration of proteoglycans. Cationic dyes like ruthenium red, silver tetraphenylporphine sulfonate, bismuth nitrate and cuproinic blue are also used in localization of proteoglycans. High-iron diamine-thio-carbohydrazide silver proteinate (HID-TCH-SP) method of Spicer is used for localization of sulfated glycoconjugates. Ruthenium hexamine trichloride is used for the detection of anionic groups of proteoglycans and glycolipids.

In the study of predentin and dentin, cuproinic acid with magnesium chloride at critical electrolyte concentration, allows a larger surface representation of proteoglycans. Biotinylated hyaluronic acid- binding complex stain the hyaluronic acid. Lectin binding sites on carbohydrate are visualized by fluorescein dyes or horseradish peroxidase techniques.

Proteins

Dinitrofluorbenzene, ninhydrin and ferric ferricyanide reagents are used for histochemical study of proteins.

Lipids

Frozen or freeze-dried sections are used for the histochemical study of lipids. Fat colorant dyes like Sudan dyes are used for the study of total lipids. Phospholipids are studied by iodoplatinate and malachite green aldehyde.

Enzymes

Phosphatases

Methods used for the demonstration of phosphatase are Gomori method and simultaneously coupling azo dye technique.

Immunohistochemistry

In immunohistochemical techniques, antiserum or solution containing antibody is applied to a tissue section. Antibody react with antigen present in tissue section and

form an antigen antibody complex. This complex is conjugated to fluorescent dye like rhodamine and fluorescein isothiocyanate (FITC) or to enzyme like peroxidase antiperoxidase (PAP) and examined in a fluorescence microscope. The enzyme-bound antigen-antibody complexes are exposed to enzyme substrate and antigenic sites are examined by light or electron microscopy.

CLINICAL CONSIDERATIONS

Histochemical techniques are very important in the diagnosis of any oral lesions. A tumor of salivary gland origin may be differentiated from that arising from the nonglandular epithelium with the help of a histochemical stain. Presence of certain mucopolysaccharides demonstrated by histochemical stain of the fungi found in the infected human tissues can lead to its correct diagnosis. Candidiasis, histoplasmosis, actinomycosis, blastomycosis and coccidioidomycosis can be diagnosed only after special histochemical stains.

Tumors that arise from the fat cells (lipoma and liposarcoma) may be correctly diagnosed by histochemical stains that reveal lipids.

Advanced techniques are used basically for the structural and histogenetic research of oral tissues. Since tumors arising from epithelial derivatives, lymphoid tissue and mesenchymal components not only have different biological behavior but also show difference in the therapy, it is of immense value to know the exact nature of the tumor. Histochemical stains and immunoperoxidase staining of tumor markers are of extreme help in the correct diagnosis of the tumors of oral cavity and identifying their tissue of origin. 'In-situ hybridization' and 'FISH' are being extensively used nowadays in identifying the chromosomal aberrations or gene products characteristic of a particular tumor. These help in diagnosis as well as in predicting the behavior of tumor and prognosis. Immunofluorescence studies are useful in the diagnosis of vesiculobullous lesions of oral cavity and in diagnosis of collagen disorders. Electron microscopy is useful in elucidating the nature of tumor as well as in the understanding of etiopathogenesis of disease. Immunohistochemistry, *in situ* hybridization, FISH, PCR and electron microscopy are useful in diagnosis of

infective agents like rickettsia, mycoplasma viruses and bacteria. PCR is proving extremely helpful.

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Repair and Regeneration of Dental Tissues

- Introduction
 - Repair
 - Regeneration
- Repair of skin
 - Epithelial response
 - Connective tissue response
- Repair of enamel
- Repair of dentin
- Dental caries
 - Cavity preparation
- Repair of pulp tissue
- Repair following tooth extraction
- Repair of periodontium
- Repair of the oral mucosa
- Recent advances in periodontal regeneration

INTRODUCTION

The ability of damaged tissue to repair itself is a sign and response of life. An unhealed wound may result in death of the organism. Hence, healing is considered one of the primary survival mechanisms from birth onwards. Healing includes very complex series of biologic events.

The inflammatory reaction is an important phase of repair of tissues. Oral wounds are common, some being sustained accidentally and others are inflicted by a surgeon for a specific purpose. If damage or insult is sufficient to elicit an inflammatory response, the healing process is evoked. There are certain tissue responses like hyperkeratinization of epithelium or tubular occlusion in dentin in response to attrition; they do not involve inflammation. Tissue injury evokes different responses. If tissue injury is sufficient to cause inflammation, a healing process results. Healing involves repair and regeneration.

Repair

Repair is the restoration of tissue continuity with scar tissue and distortion of normal architecture.

Regeneration

Regeneration is the complete restoration of architecture of the tissue without distortion of normal architecture. Several factors affect the rate of healing of oral wounds. These are the location of wound, physical factors, circulatory factors, nutritional factors, age of the patient and infection. Diffuse calcifications are found in root canal and less often in coronal area while pulp stones are seen mainly in coronal pulp.

REPAIR OF SKIN

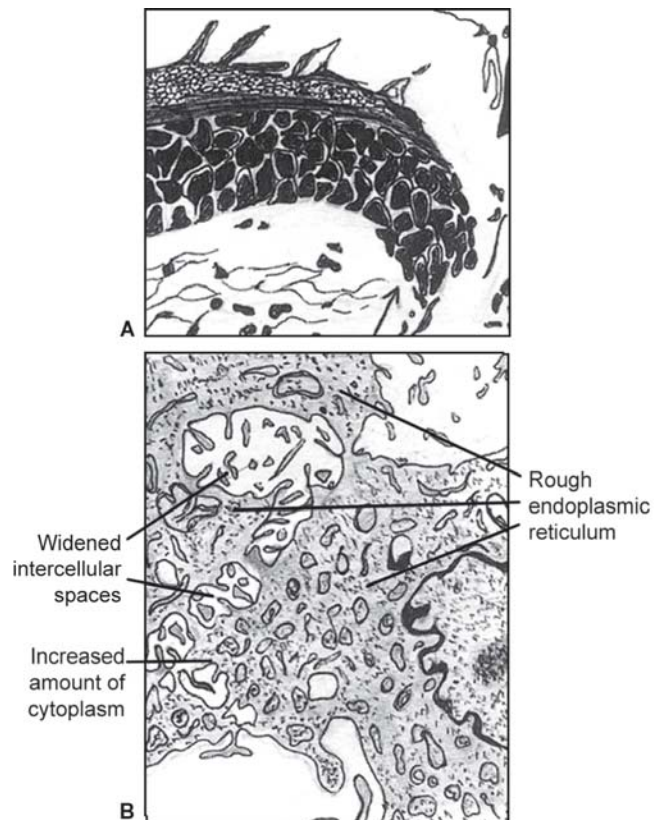
The repair process is irrespective of the kind of injury. An account of the repair of skin after being damaged by a simple incision and where healing is uncomplicated by infection has been described. This is done to explain the repair process in the dental tissues. Repair of skin involves mainly following two processes.

- i. Epithelial response
- ii. Connective tissue response.

Epithelial Response

The outcome is a re-establishment of epithelial continuity and the covering of the exposed connective tissue. There

is mobilization and migration of the epithelial cells at the wound margin. The epithelial cells lose their strong attachment to each other and to the underlying connective tissue. This histologically appears as a widening of the intercellular spaces between cells. Basal epithelial cells adjacent to the immediate margin of wound shows increased mitotic activity. There is increase in the amount of cytoplasm of the epithelial cell and rough endoplasmic reticulum. These cells are then mobilized and they migrate across the exposed wound surface and finally cover it. All these features are characteristics of epithelium, which is supported by a disturbed connective tissue. Epithelization occurs in about 24 hours in a clean incised uninfected wound (Figs 19.1A and B).



Figures 19.1 A and B: Epithelial response in a wound of lip epithelium seen with light microscope; A. Separation of epithelial cells from each other at wound margin before migration takes place; B. Migrating epithelial cells as seen under electron microscope

Connective Tissue Response

This response involves the following.

- a. Hemostatic changes (Hemostasis).
- b. Inflammatory changes (Inflammation)

- c. Proliferative changes (Proliferation and synthesis)
- d. Secretory changes. (Proliferation and synthesis)

Hemostatic Changes (Hemostasis)

Following any injury, there is hemorrhage into the tissue defect with aggregation of platelets, which coagulate to form a clot. This clot serves the following three important functions.

1. It acts as a hemostatic barrier
2. Unites the wound margins
3. Also provides a scaffold for the eventual migration of reparative cells.

Inflammatory Changes (Inflammation)

The first inflammatory cells to invade the wound and to appear within a few hours of an injury are the neutrophils. These reach in maximum number within 24 hours. They have a short life-span at the wound site before degenerating. While degenerating rapidly at the wound site, they release various enzymes that help to destroy the damaged tissue. Their main function however is to control bacterial invasion and subsequent infection. The predominant inflammatory cells present between second and fifth days are the macrophage, which enter the wound after 24 hours (Fig. 19.2). Macrophages help in the following ways:

- i. Secrete many biologically active proteins, which include a mitogen specific for fibroblasts. Therefore, in their absence, fewer fibroblasts are formed. So the rate of repair is slowed.
- ii. The classic function of macrophages, however is to destroy foreign and damaged materials.

Proliferative Changes and Secretory Changes (Proliferation and Synthesis)

The stage of proliferative and secretory changes involves the multiplication of fibroblasts and synthesis of collagen to form scar tissue.

The fibroblasts for wound repair are obtained from two sources:

- a. Division of undamaged fibroblasts at the wound margins, that is at the periphery.
- b. By the differentiation and proliferation of undifferentiated perivascular cells (Figs 19.3 and 19.4).

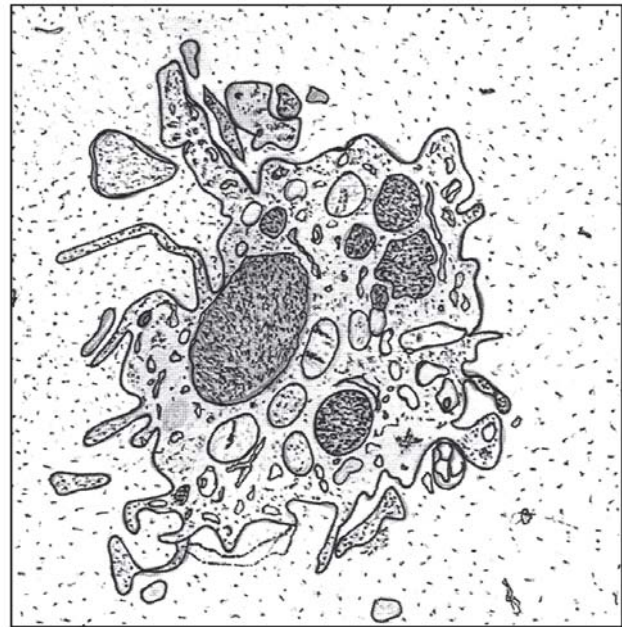


Figure 19.2: Macrophage in the wound defect 24 hours after wounding as seen under electron microscope

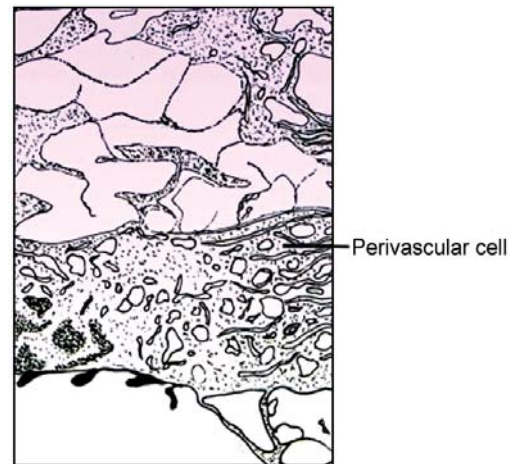


Figure 19.3: Cell division taking place in an undifferentiated perivascular cell

REPAIR OF ENAMEL

The capability of repair or regeneration of enamel is very limited as the cells that formed it no longer exist. Enamel can, however, undergo limited repair by physicochemical means. It is possible for remineralization to occur in the subsurface layer of enamel if the carious process can be arrested and becomes confined to the enamel such that

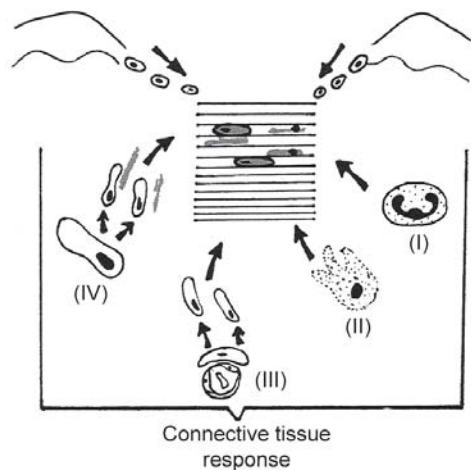


Figure 19.4: Summary of repair process. Epithelial response occurs by proliferation and migration to cover the defect. (I) The connective tissue response involves successively a polymorph response (12 to 24 hours). (II) Macrophage response (2 to 5 days). (III) Fibroblast response (2 days and onward) from undifferentiation perivascular cells and undamaged fibroblasts. (IV) The new fibroblasts form the collagen of scar tissue

there is no breakdown in the surface layer of enamel. In arrested caries surface layer of enamel after remineralization appears shiny, glazed brownish smooth.

The remineralization of enamel depends on calcium and phosphate ions available from saliva. In the presence of fluoride, the remineralized enamel may be more resistant than normal enamel to any further demineralization.

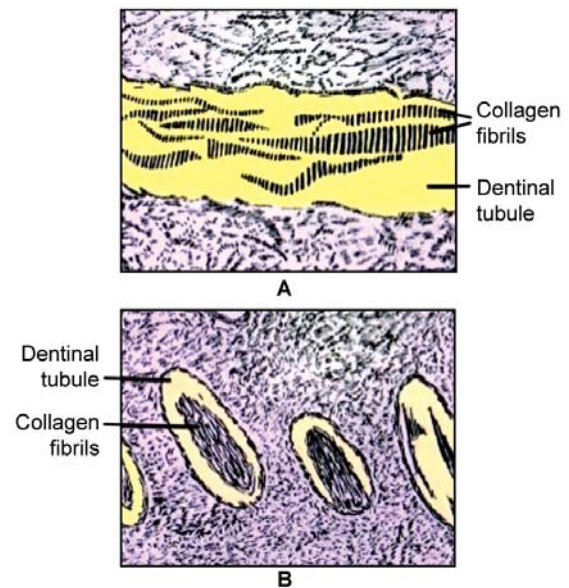
REPAIR OF DENTIN

Dentin is a vital tissue and its response to damage can be divided into two main categories.

- i. Changes within dentin which are manifested by occlusion of the dentinal tubules following retraction of the odontoblastic process. This happens in two ways:
 - a. Extension of peritubular dentin at the expense of process.
 - b. Deposition of collagen and its eventual mineralization to occlude the tubule (Figs 19.5 A and B).
- ii. Deposition of reparative dentin (Fig. 19.6).

Some of the responses like dead tracts and sclerotic dentin have already been discussed in chapter 5.

It is necessary to analyze the repair responses of enamel and dentin in terms of the carious process and to dental procedures undertaken to restore the structural damage caused by that process.



Figures 19.5 A and B: Deposition of collagen fibrils to occlude the dentinal tubule as seen in demineralized section; A. Patent tubule occluded by collagen fibrils; B. Cross-section showing a number of tubules containing collagen fibrils

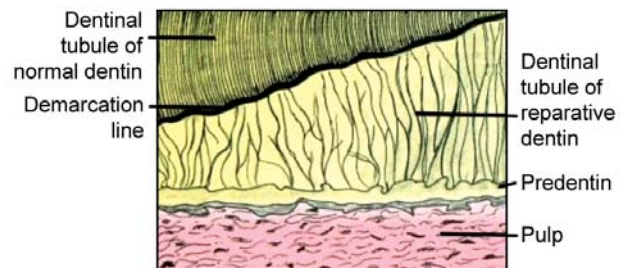


Figure 19.6: Reparative dentin showing fewer and more twisted dentinal tubules than normal dentin

DENTAL CARIES

The carious process is a dynamic one with alternating phases of demineralization and remineralization. Three distinct zones can be recognized in the early carious lesion.

Zones of Caries

Translucent Zone

Translucent Zone depicts the first zone of change in enamel, which is observable with the light microscope. This zone is present at the inner advancing edge of lesion. Removal of mineral occurs in this zone.

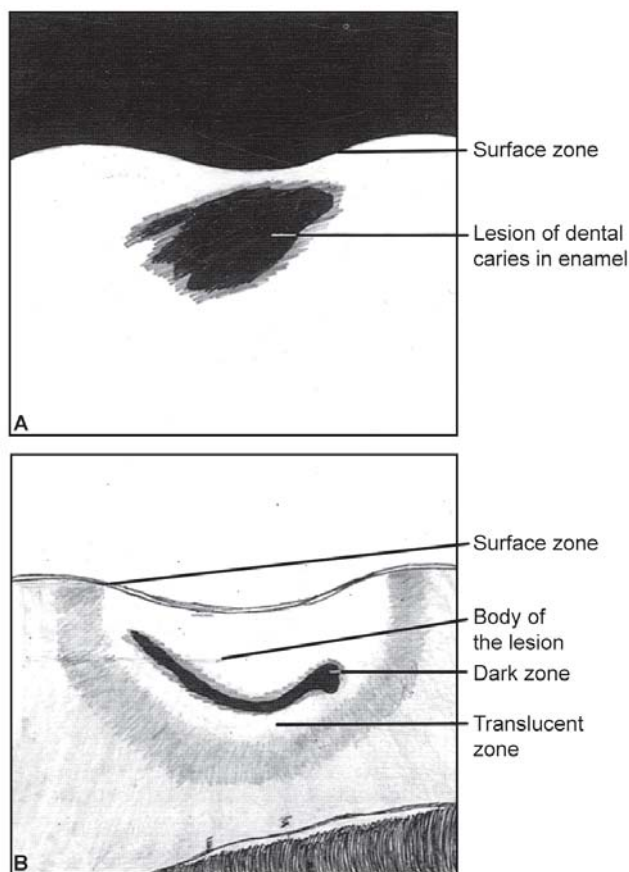
Dark Zone

Dark zone depicts an area, which was initially demineralized but is now undergoing remineralization.

Body of the Lesion

Body of the lesion is third zone that occupies a space between the dark zone and apparently intact enamel surface (Figs 19.7 A and B). This is an area where the bulk of mineral has been lost and where the most destructive morphologic changes have occurred.

A very important characteristic of the early carious lesion is that the surface layer remains intact. This happens because here most of the calcium and phosphate ions either from subsurface dissolution or from the saliva are reprecipitated. Most of the demineralization occurs at subsurface level. The electron microscope has shown that the crystals along either side of the rod sheath in the carious lesion are larger and more electron dense than elsewhere. These crystals are reprecipitated, and their localization suggests that the rod sheath have some role in the development of the lesion. Briefly, the carious



Figures 19.7 A and B: Enamel lesion in dental caries; A. Showing subsurface location of the lesion as seen in a microradiograph; B. The same lesion when seen with polarized light.

process in enamel consists of a diffuse demineralization within the bulk of the lesion which affects crystals in all regions of enamel.

Once the carious process reaches dentin the response is sequential. Sometimes, at this stage, the enamel is still intact and bacterial invasion has not occurred. But the permeability of enamel to acid and various other chemical stimuli is increased by the lesion. This elicits a response from the dentin-pulp complex. The nature of this initial response is such that there is the deposition of reparative dentin, a zone of normal dentin and then a zone of dentin where the tubules have been occluded by one of the methods already described to produce a translucent zone (Fig. 19.8A). This deposition occurs from pulp towards dentinoenamel junction.

Once the enamel cavitates, destruction of dentin matrix starts. Superimposed on the initially described process is a mild inflammatory reaction in the pulp along with bacterial invasion of the dentin enclosed by a translucent zone (Fig. 19.8B)

Acid diffuses ahead of the bacteria, as the bacteria infecting dentin are acidogenic. Further demineralization of dentin occurs. Along with this a mechanism to increase the zone of sclerotic dentin comes into play.

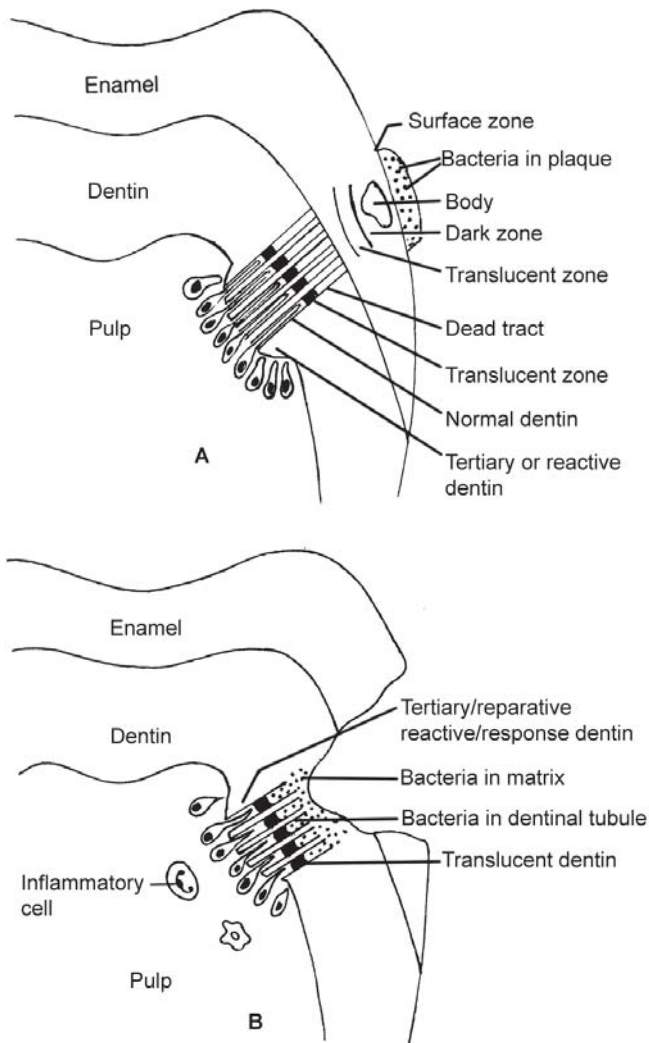
In addition to the extension of intratubular dentin and deposition of collagen, reprecipitation of mineral occurs, most of it in enamel, but some also occur in the dentinal tubules (Fig 19.9). Bacteria are initially limited to the tubules (Fig 19.10), but later they escape this confinement and destroy the dentin matrix (Fig. 19.11). Carious process within dentin can be arrested either naturally or by surgical intervention to remove infected dentin. Surgical intervention is achieved by cavity preparation and restoration of lost tooth tissue by substitute materials.

Cavity Preparation

Cavity preparation involves removal of both enamel and dentin. Enamel is like an epithelium and the dentin-pulp complex is the connective tissue it covers. So, the repair response of these dental hard tissues is similar to that of skin.

Epithelial Response

The ameloblasts (the cells that form enamel) are lost at the time of tooth eruption. So, there is a lack of any kind of



Figures 19.8 A and B: Carious process; A. Subsurface lesion-Enamel is still intact bacteria are present in the plaque on the enamel surface; B. Bacterial invasion resulting due to broken down enamel

epithelial response. In dental practice, substitute restoration materials are used to overcome this deficiency. These materials perform two essential functions, which are as follows

- i. Mimic the hardness of enamel, and
- ii. Serve as an effective seal to the external environment to protect the underlying connective tissue, which is the pulp dentin complex.

Connective Tissue Response

If involvement of dentin occurs to an extent that the odontoblasts and their processes are damaged, a response

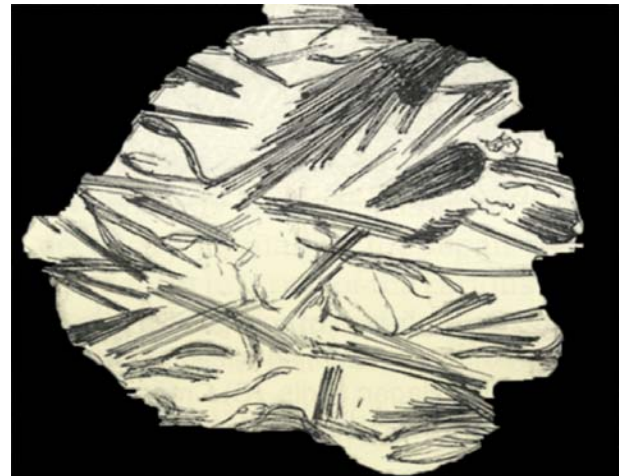


Figure 19.9: Reprecipitation of mineral (reminereralization) in dentinal tubule

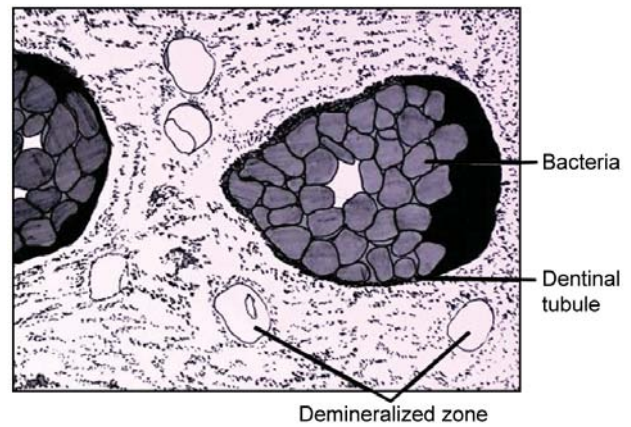


Figure 19.10: Bacteria confined to the dentinal tubules in dental caries. Even than demineralization of intertubular dentin is also seen because acid produced by bacteria travels faster towards the pulp than the bacteria themselves

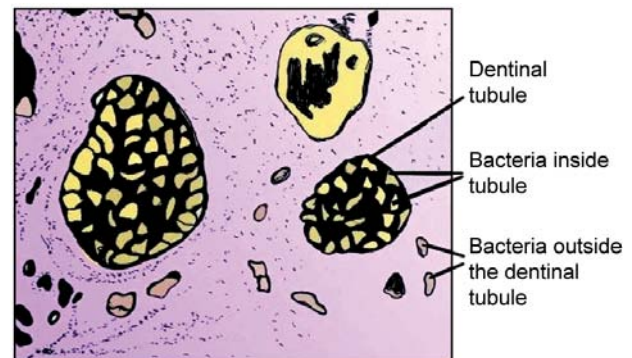


Figure 19.11: Few bacteria have escaped the boundaries of dentinal tubules in dental caries

similar to connective tissue response in skin is seen. There is only one major difference and that is a lack of hemostatic response. This is because the dentin is avascular. But, if the damage is extensive enough to involve the pulpal component of the complex, a hemostatic response also occurs.

There is an inflammatory response involving neutrophils and macrophages. This is followed by the proliferative and secretory response. New fibroblasts are formed which are basically odontoblasts. These odontoblasts eventually lay down collagen (scar tissue), which becomes mineralized to form tertiary dentin.

Certain local factors, however, have always overshadowed this basic reparative response of the dentin-pulp complex. These are as follows.

- i. A low-grade constant infection tends to change the pulpal response. This happens because the restorative materials because acid produced by bacteria travels faster towards the pulp than the bacteria themselves do not completely mimic the ability of the enamel and dentin to cover and seal. Hence, microleakage can occur around some restorative materials.
- ii. With age, the blood vessels of the pulp become compressed and atherosclerotic. Blood flow is decreased and this affects the repair capabilities of the dentin pulp complex. But the repair mechanism is not affected.

This occurs as dentin is formed throughout the life so that the pulp chamber becomes progressively smaller. Along with this there is an increase in the collagen content, decrease in cellularity and loss of water from the ground substance.

- iii. Third is the controversial issue regarding the extent of the odontoblastic process within the dentinal tubules.

An odontoblast has the ability to repair itself, as long as the cavity preparation was not too deep and the cell body was undamaged. This assumption was made on the fact that the odontoblast cell processes extend to the dentinoenamel junction and that once dentin is cut, so is this process.

But if the cavity preparation is very deep, the odontoblast may be irreversibly damaged. The probability, which must now be understood, is that shallow cavity preparation might not involve the odontoblast at all if its process extends only partially into dentin.

iv. The epithelium is needed to start the differentiation of odontoblasts from undifferentiated mesenchymal cells. But in the mature pulp, no epithelium exists to initiate the process of differentiation for tertiary dentin formation. This led to the following assumptions.

- a. Odontoblasts have the ability to repair themselves after damage.
- b. Undamaged odontoblasts undergo mitosis and thereby provide odontoblasts for tertiary dentin formation.

But mitotic divisions are never seen in odontoblasts. Odontoblasts are highly specialized cells and specialized cells generally lose their capacity to divide. However, like other hard tissues (for example alveolar bone), epithelium is needed only for the initial differentiation of odontoblasts. Once differentiated, they have the ability to differentiate further, forming cells as and when needed, in the absence of epithelium.

Briefly, the dentin pulp complex has the same repair mechanism as any other connective tissue. But, the repair potential can be, and is, affected by the above local factors, the important one being the constant presence of pulpal inflammation due to microleakage around restorative materials.

REPAIR OF PULP TISSUE

The pulp responds to irritation, which may be mechanical, thermal, chemical or bacterial by producing reparative dentin and mineralizing any affected dentinal tubules. The pulpal tissue has remarkable reparative abilities. The reparative dentin formed in the pulp and the calcification of the tubules results to wall off the pulp from the source of irritation.

The pulp may get inflamed because of bacterial infection or by cutting action and placement of an irritating restorative material. The pulpal tissue has macrophages, lymphocytes, neutrophils monocytes, plasma cells and mast cells. These aid in the process of repair of the pulp. The rigid dentinal wall protects the pulp.

When inflammation occurs, hyperemia and edema leads to accumulation of excess fluid outside the capillaries. Because of unyielding closure of the apical foramen, pressure on apical vessels increases resulting in ischemia and necrosis of the pulp. If inflammation is not very severe pulp will heal as it has excellent regenerative properties.

REPAIR FOLLOWING TOOTH EXTRACTION

The wound created by extracting a tooth differs from an incisional skin wound in that there is substantially more loss of soft tissue and bone is also involved. But in the repair process the same basic mechanism is followed.

Immediate Reaction Following Extraction

Following removal of a tooth, the blood which fills the socket clots, entrapping the red cells in the fibrin meshwork, and ends of the torn blood vessels in the periodontal ligament become sealed off. The time period, immediately following tooth extraction is very critical, because if the clot is dislodged, healing may be greatly delayed and very painful. If infection intervenes, it leads to a condition known as dry socket. It is so called as the socket has a dry appearance (after loss of clot) because of the exposed bone.

During the first 48 hours after extraction, there is vasodilatation and engorgement of the blood vessels in the periodontal ligament. Along with this there is mobilization of leucocytes to the area around the clot. The surface of the clot is covered with fibrin. The clot itself shows an area of contraction. The collapse of the unsupported gingival tissue into the opening of the fresh extraction wound is of great aid in maintaining the clot in position.

First Week Wound

During the first week following extraction, proliferation of fibroblasts present in the remnants of the periodontal ligament occur. These cells begin to grow into the clot around the entire periphery. This clot which is only a temporary structure serves as a scaffold upon which cells associated with the healing process migrate. The clot is gradually replaced by granulation tissue. The epithelium at the periphery shows proliferation by mitotic activity. The crest of the alveolar bone, which forms the neck of the socket, shows beginning of osteoclastic activity.

During this period, the blood clot begins to undergo organization by the ingrowth around the periphery by fibroblasts and occasional small capillaries. Remnants of periodontal ligament are still visible. There is no evidence of significant new osteoid formation. An extremely thick layer of leukocytes has gathered over the surface of the clot. The edge of the wound continues to show epithelial proliferation.

Second Week Wound

Along with continued organization of the clot by fibroblast there is penetration of new delicate capillaries into the center of clot. The remnants of the periodontal ligament are gradually degenerating and are no longer recognizable. Wall of the bony socket appears slightly frayed. In smaller sockets, epithelization may be complete. But in large posterior teeth, even though there is an extensive epithelial proliferation over the wound surface, the wound is usually not completely covered. Fragments of necrotic bone which may have fractured from the rim of the socket during extraction are seen in the process of sequestration.

Third Week Wound

By third week, the original clot appears almost completely organized by maturing granulation tissue (Figs 19.12 A to D). The entire periphery of the wound from the socket wall is seen surrounded by very young trabeculae of osteoid or uncalcified bone.

The original cortical bone of the alveolar socket undergoes remodeling so that it no longer consists of such a dense layer. Osteoclastic resorption has rounded off the crest of alveolar bone. Surface of the wound may have been completely epithelized.

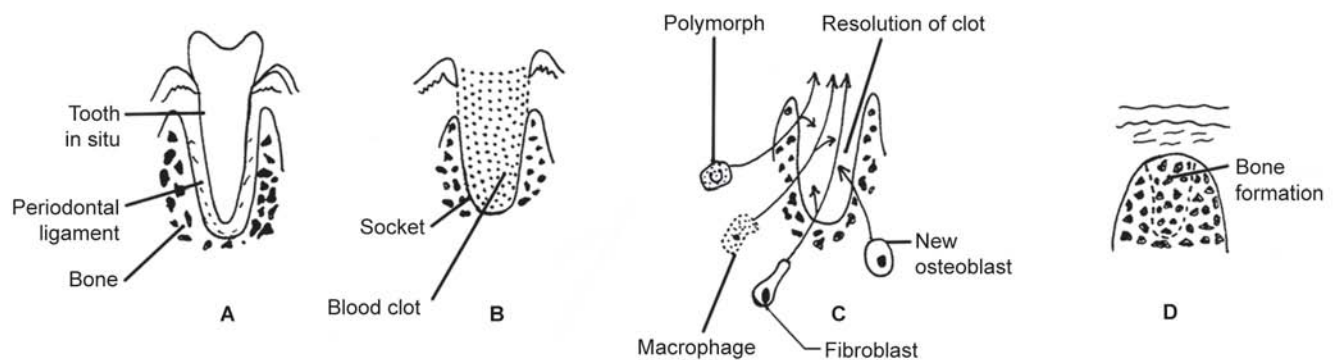
Fourth Week Wound

During fourth week, the wound begins final stage of healing. There is continued deposition, remodeling resorption of the bone filling the alveolar socket. This maturative remodeling continues for many weeks.

Roentgenographic evidence of bone formation is not evident until the sixth to eight week after tooth extraction. There is still roentgenographic evidence of differences in the new bone of the alveolar socket and the adjacent bone for as long as 4 to 6 months after extraction in some cases. By 10 to 12 weeks the extraction site can no longer be clinically distinguished (Figs 19.12 A to D).

REPAIR OF PERIODONTIUM

Repair of periodontium depends greatly on the severity and nature of the damage that occurs. When there is a complete loss of tooth support following avulsion, the clinical practice is to save the soft tissue attached to the root of avulsed tooth as much as possible. This is done before the avulsed tooth is repositioned in the socket.



Figures 19.12A to D: Repair response following tooth extraction. A. The tooth is in situ. B. After extraction of tooth, the socket is filled with blood clot. C. Resolution of clot by responses of macrophages, polymorphs and fibroblasts new osteoblasts colonize the bony defect. D. Remodeling of collagen scar and formation of bone

The proliferative response is derived from the cells of the bone marrow adjacent to the periodontal ligament when the trauma is less severe and only part of the periodontal ligament is damaged. In this case, the synthetic response is osteogenic and bone forms causing fusion of the tooth to the jaw bone. This condition is called ankylosis.

The proliferative response is derived from the cells of periodontal ligament when the damage is slight. This results in regeneration of tooth support as cells of the periodontal ligament have the potential to form all the tissue involved in tooth support. In short, the repair process in the periodontium is determined by the kind of cells which, repopulate after injury.

Repair of the periodontal ligament may also use an important biologic phenomenon, the ability of the fibroblast to remodel collagen. The repair of skin involves the formation of scar tissue. Repair of periodontal ligament after tooth movement shows the same mechanism of repair as found in skin but without scar formation. Scar formation does not take place because even though collagen (scar) is the reparative tissue, this scar tissue is almost immediately remodeled by the fibroblasts of the ligament. Thereby its normal architecture is restored.

The general problem faced following periodontal disease is the reattachment of tooth support. The main aim of periodontal therapy is to eliminate periodontal disease first and then restore lost tooth supportive structures. There are different clinical modalities which are taken up for achieving this:

1. To utilize the osteogenic potential of adjacent bone tissue or of undifferentiated cells to try and replace

bone. Various materials with osteogenic potential such as frozen dried bone are implanted around teeth. Although these materials produce bone, but this bone in no way helps in tooth attachment.

2. The second approach is to influence connective tissue other than the ligament to help in tooth support. In this method, the root surface is treated with a mild acid such as citric acid. This partially demineralizes the cementum and dentin of the root surface. Thereby the collagen fibers of the hard tissue matrix are exposed. This is done with the hope that the exposed collagen fibers will remodel and will unite with the collagen of the soft connective tissue of the gingiva to re-establish an attachment. But such splicing rarely occurs.
3. A new concept, currently being explored is that of guided tissue regeneration. In this, after periodontal surgery, an effort is made (using an inert barrier) to exclude the gingival connective tissue from the repair process. This is done so that repair is achieved totally by periodontal ligament cells.
4. Lastly, the clinical field of implantology has been greatly influenced by the difficulty in regenerating tooth support. Initially it was thought desirable to obtain a fibrous connection between the implant and bone, because this imitates the attachment of tooth to bone. But, when fibrous encapsulation of the implant is achieved, gingiva around it is formed by the cells of the gingiva. These cells do not have the capacity to perform in the same way as periodontal ligament fibroblasts. The result is destruction of the connective tissue and loss of implant. Hence, the process of osseointegration (direct contact of implant with bone) avoids the need to duplicate the periodontal ligament.

REPAIR OF THE ORAL MUCOSA

The principle of repair followed by the oral mucosa is similar to that followed by skin. The only difference being, it does not form scar tissue that readily. Therefore, it is possible to carry out surgery within the mouth without any danger of producing scar tissue. This is because even though scar tissue is formed, it is rapidly remodeled to restore normal architecture (Fig. 19.13).

There are a number of surgical procedures that involve the detachment of the gingiva from the tooth surface. Later, there is reformation of this junction. After surgery, a new junction with the same histologic characteristics as before develops from the phenotypically different oral epithelium. Junctional epithelium has features of immature epithelium. When the basal gingival cells re-establish contact with the tooth surface, they become associated with a deep connective tissue which lacks the instructions for epithelial maturation.

In short, this chapter has clearly described the basic mechanism of repair followed by dental tissues. At the same time, it has brought forth possible factors, which both help and hinder this process. In spite of the absence of ameloblasts, the physicochemical mechanisms allow a limited form of repair in enamel. The substitutes for the epithelial response in the form of dental restorative materials are used in restorative dentistry. Dentin has the capacity to undergo repair using basic mechanisms, but the scar tissue, which forms, becomes mineralized. This basic response has earlier been overshadowed by the occurrence of microleakage and by age changes, which are very evident in the pulp. The repair of tooth support

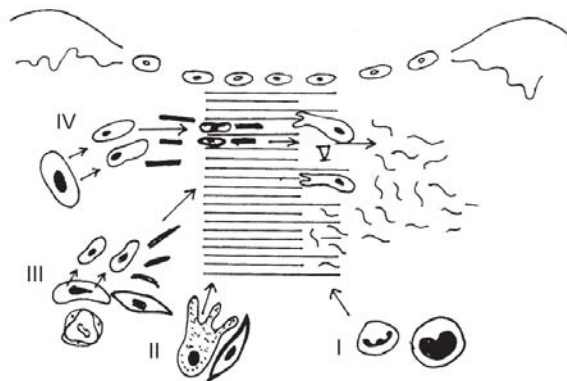


Figure 19.13: Repair process in gingival epithelium showing remodeling of scar tissue to restore normal architecture (that is why the scars are rare in oral cavity); I. The macrophage response; II. Fibroblast differentiation; III. Collagen synthesis; IV. The scar remodels; V. Collagen phagocytosis

depends on the degree of damage. If the damage is minimal and programmed follicular cells are available, repair occurs involving scar tissue formation, but this scar is rapidly remodeled to restore normal architecture. If the damage is substantial, the defect is repopulated by bone-forming cells, as in socket repair, leading to fusion of the tooth to the jaw. Finally, wounds of the oral mucosa, and especially the gingiva, often heal without the formation of scar tissue. Scar tissue in fact forms but is quickly remodeled to restore normal architecture.

RECENT ADVANCES IN PERIODONTAL REGENERATION

Adult stem cells: Tissue engineering is new technique for the fabrication of new tissues to replace damaged and destroyed tissues. They provide new concepts of periodontal regeneration. For periodontal regeneration tissue engineering consists of new concepts in bioengineering and nanotechnology. Adult stem cells are helpful for periodontal regeneration (Fig. 19.14).

For periodontal regeneration tissue engineering along with bioengineering and nanotechnology are very important developments.

The science of bioengineering at molecular level produce such materials which upto now are unknown and have unknown properties. Due to this biodegradable scaffolds will be developed which will incorporate the required instructional molecular messengers to select the adult stem cells. In future this will be the basis of periodontal regeneration.

Targeted gene transfer: By gene therapy site directed delivery of proteins at therapeutic levels for longer periods of time can be provided. In this seeding of cells containing transgenes coding for specific regenerative instructional messages is used (Fig. 19.15). The ability of adenoviruses encoding transgenes for platelet derived growth factor is used to transduce and altered behavior of periodontal cell. There is potential for altered delivery of growth factors within periodontal tissues.

All oral tissues have a primary mechanism of repair. Local factor may hinder and benefit the repair process.

Enamel—The absence of ameloblasts prevents regeneration of enamel. But restricted form of repair takes place by, physicochemical mechanisms.

Dentin—By primary mechanisms dentin has the capacity of repair and the repair tissue become mineralized.

Pulp—If irritation is mild reversible hyperemia and various types of defense cells take care of pulpal irritation. As the pulp is surrounded by hard dentin the repair is restricted.

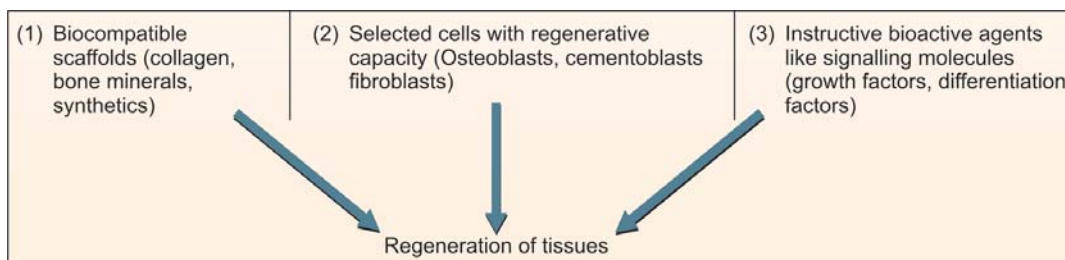


Figure 19.14: Three essential factors responsible for tissue engineering

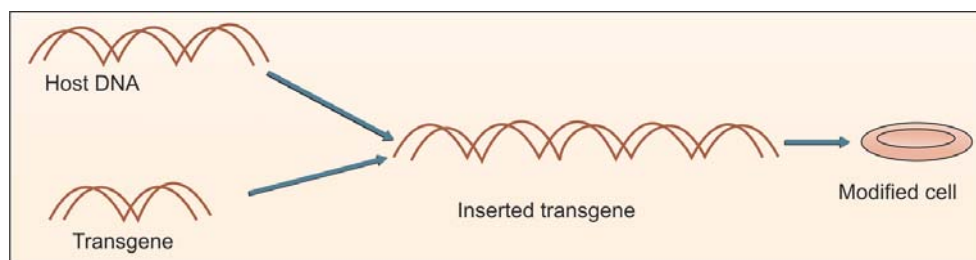


Figure 19.15: In vitro gene therapy by insertion of transgene in host DNA of cells and following synthesis of a synthetic matrix

If the irritation is severe irreversible hyperemia may be followed by nonvitality of pulp

Periodontal ligament—If the damage is minimal with the availability of follicular cells repair is followed with temporary scar which disappears. Gingiva is repaired usually without the formation of scar tissue.

Oral mucosa—Oral mucosa heal without formation of scar tissue hence surgery in the oral cavity can be carried out without fear of scarring.

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Fibroblast and Its Products

- Introduction
- Structure of the fibroblast
- Properties of fibroblast
- Functions of the fibroblast

Aging



INTRODUCTION

The most predominant cell found in the connective tissue is the fibroblast. The tissues and supporting structures of the tooth constitute the connective apparatus. The fibroblasts are important in the development, structure and function of tooth. Main function of fibroblast is the formation of the extracellular fibers of the connective tissues. These fibers are collagen, elastic, and oxytalan. Besides these, fibroblasts perform numerous other functions.

The ground substance in which the fibroblasts and their fibrous products are present is produced by fibroblast. Being motile and contractile, fibroblasts determine the structural organization of connective tissue, mainly during embryogenesis. Thus, the fibroblast acts as an architect and builder of the connective tissue.

STRUCTURE OF THE FIBROBLAST

Fibroblasts are ordinarily found associated with collagen fiber bundles. When seen under light microscope. They are recognized as follows.

Resting Fibroblast

Resting fibroblast have a flattened, darkly stained closed nucleus and a little cytoplasm. These resting fibroblasts are found in the tendon (Fig. 20.1).

Active Fibroblast

Active fibroblasts have a pale-staining open-faced nucleus and abundant cytoplasm (Fig. 20.2). Under electron microscope, the active fibroblasts have cytoplasmic organelles in an exaggerated amount. There are many Golgi complexes and profiles of rough endoplasmic reticulum, mitochondria and secretory vesicles (Fig. 20.3). This indicates synthetic and secretory function of the fibroblasts.



Figure 20.1: Structure of inactive fibroblast as seen under light microscope



Figure 20.2: Structure of active fibroblast seen with light microscope

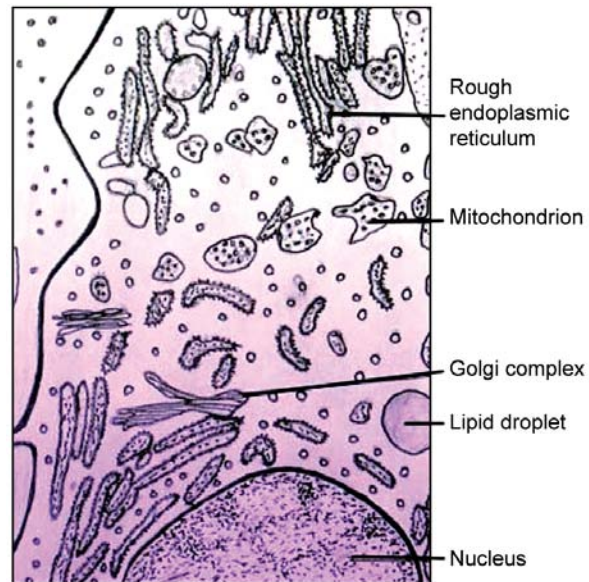


Figure 20.3: Structure of an active fibroblast as seen under Electron Microscope

Microtubules

Microtubules are long cylindrical slightly curved structures. These are proteinaceous in nature. Their average diameter is 240 nanometers. The main protein is tubulin. It is similar to actin, which is the muscle protein. Microtubules function in maintaining the shape of the cell and the position of the intracellular structures.

Filaments

On the basis of the diameter, there are two types of filaments. These are as follows.

- i. Microfilaments
- ii. Intermediate filaments.

Microfilaments

These are less than 80 nanometers in diameter. These are composed of actin and myosin, which are contractile protein. Thus they serve as intracellular muscles and are also concerned with the maintenance of cell shapes, cytoplasmic movement and cell movement.

Intermediate Filaments

The diameter of the intermediate filaments lies between 80 and 120 nanometer. These are composed of the protein, vimentin. These form the cytoskeleton of the fibroblasts, and maintain its shape. These fibers are slender and slightly curved. They are not contractile.

The filamentous system of fibroblasts has a role in tooth movement.

Junctions

Junctional complexes develop, where fibroblasts come into contact with each other. The complexes develop from the plasma membrane of the fibroblasts. The fibroblasts adhere to their external environment through their cell membrane known as fibronexus or adhesion plaque. Fibronexus contain a protein called integrin. Integrin has receptor binding sites at both the ends. On one side that is intracellularly, it binds to the cytoskeleton. On the other side that is extracellularly it binds to an adhesive glycoprotein called fibronectin. Fibronectin has the ability to bind strongly to extracellular collagen and hyaluronic acid.

The ability of the fibroblast to form attachments to its external environment is very important and is a must for cell movements.

PROPERTIES OF FIBROBLAST

Heterogeneity

In a tissue all the cells may look alike. These similar looking cells in a tissue may have different chemical and physiologic properties. These cells have different growth rates, life-spans and also their rates of protein synthesis differ. In the gingiva, there are approximately six different

phenotypic populations of fibroblasts, each one of which has different growth rates, life-span and rate of protein synthesis.

Contraction and Motility

Fibroblasts contract because of the presence of contractile protein. This protein is present in the cytoskeleton of the fibroblast. This capability of the fibroblast to contract enables the fibroblast to move.

A fibroblast moves by extending its one edge and gets attached to the substratum by developing an attachment plaque. After this, it contracts and pulls itself forward. Sometimes, fibroblast remains stationary, contracts and changes its point of attachment.

These functional abilities of the fibroblast help in the repair process. Repair involves the migration of new fibroblasts into a connective tissue defect. Here they deposit collagen to form scar tissue. Scar tissue contracts and results in disfigurement. This contraction is because of stationary fibroblasts, which pull on collagen of the scar tissue. They are used by the fibroblasts of the periodontal ligament in tooth movement and eruption.

FUNCTIONS OF THE FIBROBLAST

The fibroblast helps in collagen fiber formation and is also responsible for the formation and maintenance of the ground substance. In this ground substance the fibrous products are embedded.

Collagen Formation

Collagen is a structural protein. It has a distinctive amino-acid composition and molecular arrangement. There are approximately ten types (Type I to X) of collagen. These are genetically, chemically and immunologically different. In all these types of fibers, the common feature is the presence of an amino acid sequence. This amino acids sequence has a high proportion of glycine and also contains hydroxyproline and hydroxylysine. The amino acids are assembled as three basic polypeptide chains. These polypeptide chains are linked together in the form of a superhelix. Differences are brought about by differences in the assembly of the basic polypeptide chain.

Microscopic Appearance

With the light microscope, collagen fibers are seen as fibers that differ in thickness and orientation. These fibers occupy the extracellular compartment that is present in between

connective tissue cells. With electron microscope, the smallest unit seen is the collagen fibril. It has a periodic banding pattern. These bands repeat at every 64 nanometers along the length of the fibril.

The diameters of the collagen fibers varies. It lies between 0.3 to 0.25 nanometer. These fibrils join to form bundles. These bundles increase in diameter to form collagen fiber and become visible with the light microscope.

Collagen Synthesis

Fibroblasts synthesize the collagen. Inside the cells the amino acids are assembled at the polysomal site. These form long polypeptide chains. These mainly include the

amino acids, proline and lysine. These two amino acids are hydroxylated before the linking of individual polypeptide chains. Vitamin C (ascorbic acid) is required for hydroxylation to take place. The three polypeptide chains form the superhelix and it forms the macromolecule called procollagen (Fig. 20.4).

Glycosylation

Procollagen macromolecules pass by way of cisternae of the rough endoplasmic reticulum to the Golgi region of the cell. Here carbohydrate components are added to the macromolecule. This process is known as glycosylation. In this linkage formation with glycosyl groups takes place.

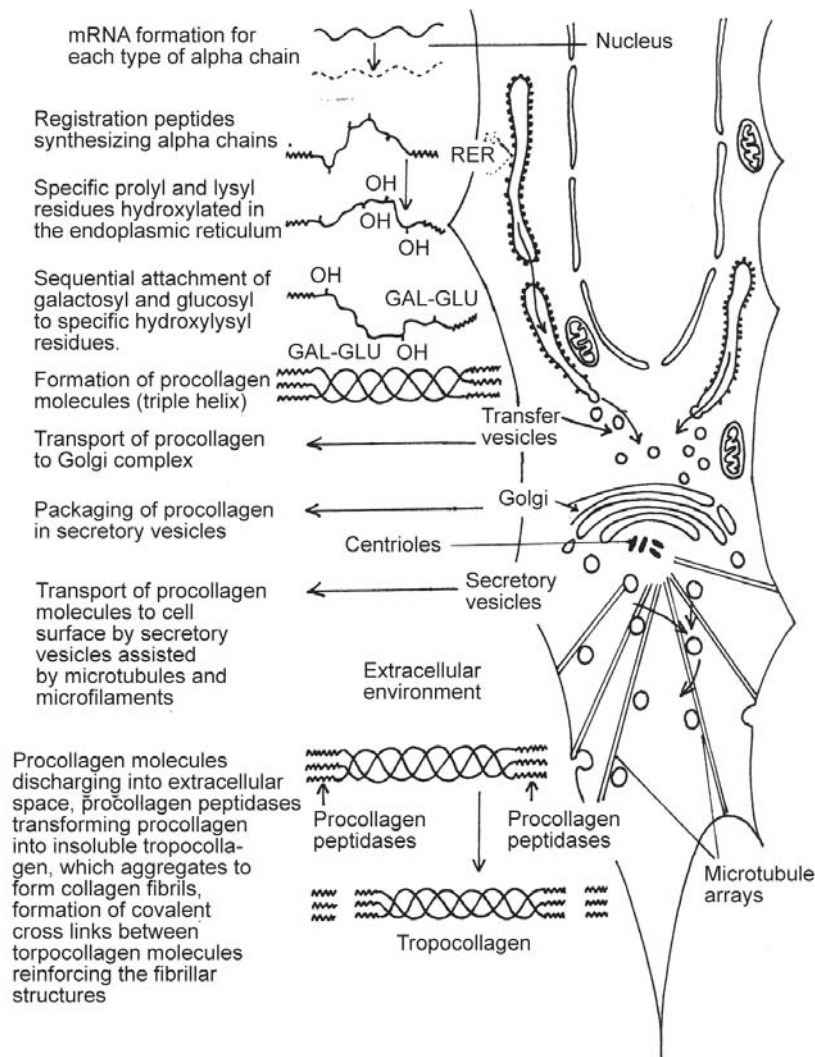


Figure 20.4: Synthesis of collagen showing chemical and structural features (mRNA= messenger ribonucleic acid)

The glycosylated procollagen is transported to the cell surface in vesicles and secreted. It loses its terminal extension. This terminal extension is lost by the hydrolytic enzyme, procollagen peptidase. Now collagen molecule is formed. This collagen molecule is a triple helix, 300 nanometers in length. The chain terminates in a short non-helical peptide, the telopeptide. Thus, telopeptides are the regions where cross-linking is established.

Collagen molecule now forms collagen fibrils. Each collagen molecule overlaps its neighbour by approximately one-quarter of its length, creating holes in between the molecules. Collagen is usually considered to be very stable and inert protein.

In a quarter stagger arrangement of collagen, each collagen molecule overlaps the next molecule by one quarter of its length. This results in the creation of holes in between the molecules. As additional chemical cross linkages develop between and within the collagen molecule, the collagen fibrils become more and more inert and stable collagen once formed persists in the tissues for an extended period of time.

Collagen Degradation

Collagen is degraded by a specific enzyme called as collagenase. Collagenase is secreted by cells, present in latent form. These are then activated by proteases. As a result smaller segments of the collagen molecule are formed.

Some connective tissue cells like pulpal cells and fibroblasts of the dental follicle produce an inhibitor of collagenase activity. These inhibitors help in collagen regulation by determining collagenase activity.

In remodeling of the connective tissue the fibroblasts synthesize collagen and degrade it by intracellular degradation. Collagen fibril segments are ingested into the fibroblast by phagocytosis. This results in the formation of phagosome into the fibroblast. Phagosomes are membrane bound vacuoles in which fibrils are present. The lysosomes of fibroblasts fuse with phagosomes and release their enzymes into it. Now phagolysosome is formed. Inside it, final degradation of the collagen fibers occurs.

Collagen Turnover

For a tissue to remain in a steady and functional state, the rate of collagen synthesis must be equal to the rate of collagen break down. If there is decrease in the formation or degradation by the fibroblast there is disruption in its architecture and function.

During tooth movement, remodeling of periodontal ligament occurs. This is done by fibroblasts. The synthesis and degradation of collagen by fibroblast has an important effect on tooth movement and periodontal disease.

Scurvy is a condition which results from deficiency of vitamin C. Vitamin C is required for the hydroxylation of the amino acids, proline and lysine. This is an important step in the formation of collagen. In scurvy, because of the absence of vitamin C, hydroxylation cannot occur and collagen synthesis in the fibroblast does not take place. But the lack of vitamin C does not affect the degrading function of the fibroblast, that is phagocytosis of collagen continues. This all results in loss of tooth support as disruption of fibroblast function occurs.

Collagen Adaptability

Collagen fibers vary in different connective tissues and adapt themselves to specific functions. There is a variation in the types of collagen found in the connective tissue. Type I collagen is found in hard tissues and type III in embryonic tissues. The number, position and stability of the cross-linkages vary in collagen taken from different tissues. Older collagen is less soluble, because collagen molecules mature and become more stable with age.

The ways by which the collagen fibrils are aggregated into fibril bundles and then fiber bundles vary. The rate of collagen turnover in different connective tissues varies. This concludes, that, there is a considerable potential for variation in collagen, which enables it to adapt to functional needs of the connective tissue in which it is found.

Elastic, Oxytalan and Reticular Fibers

Fibroblast also produces elastic fibers and oxytalan fibers. Elastic fibers run a straight course and show frequent branching. This can be seen under the light microscope, after staining. The oxytalan fibers can be demonstrated by specific staining techniques.

Ultrastructure of Elastic Fibers

These fibers have a central amorphous core of rubbery protein, elastin. It is surrounded by a sheath of tubular glycoprotein microfibrils about 10 nanometer in diameter. The formation of elastic fibers occurs. In this microfibrils are formed first. These can be seen in young elastic tissues. With maturity elastin is added by displacing the microfibrils from the periphery. Oxytalan fibers are added later. Ultrastructural features of oxytalan fibers are like immature elastin, that is microfilaments without the elastin.

The reticular fibers can be seen by light microscope. Under electron microscope they are seen as small type III collagen fibers which are surrounded by ground substance.

Ground Substance Formation

The fibroblasts form and maintain the ground substance. Its fibrous products are embedded in it. Under light and electron microscope, ground substance is seen structureless. Ground substances can be seen clearly with

special staining methods. Fibers and cells are surrounded by ground substance. Glycosaminoglycans constitute the main non-fibrous component of the extracellular compartment. Most of these are bound to proteins and are called as proteoglycans (Fig. 20.5). So glycosaminoglycans do not exist in free State.

Basically ground substance consists of the central component, glycosaminoglycan hyaluronic acid (Fig. 20.6). Proteoglycans are linked to this central component. The number of proteoglycans varies. This structural make up permits variations in types and amounts of glycosaminoglycan chains. Glycosaminoglycans are hydrophilic and thus help in regulation of tissue water.

Aging

Fibroblasts from embryonic tissue are able to undergo many divisions before, they die. This number is reduced when fibroblasts from adult tissue are cultured. Fibroblasts from long-lived species can replicate more than similar cells from short-lived species.

All this suggests that there is some correlation between the age and the life- span of fibroblasts.

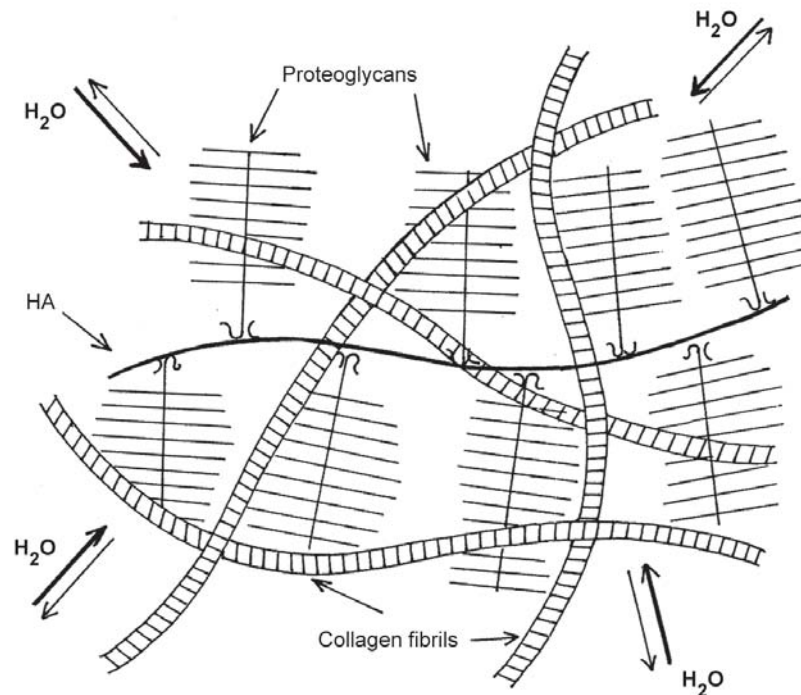


Figure 20.5: Extracellular matrix showing interaction of collagen and proteoglycan aggregates

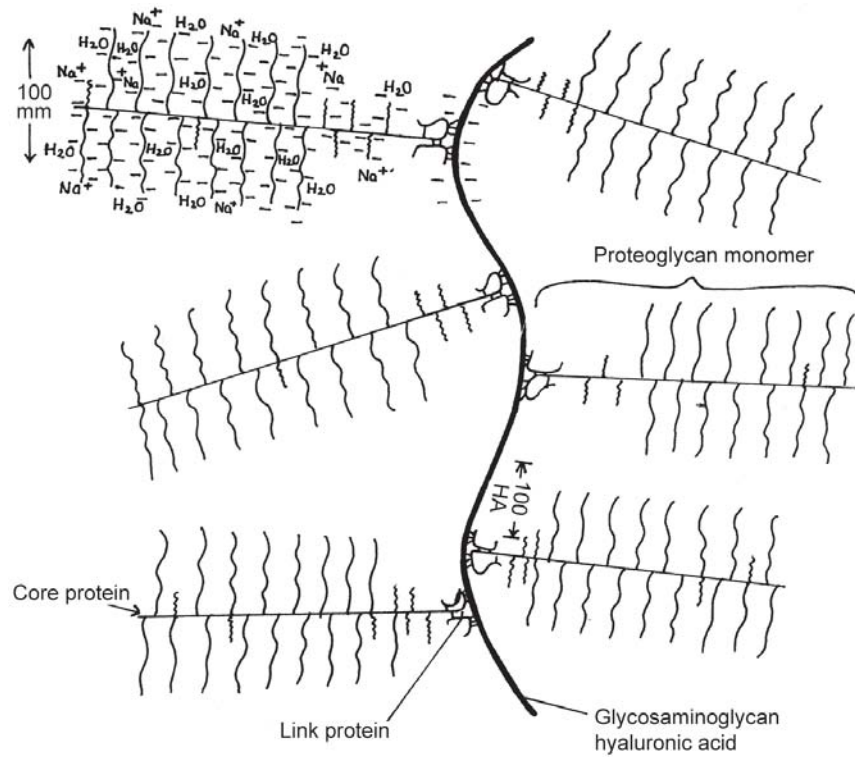


Figure 20.6: Basic model of the structural make up of ground substance

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A close-up, grayscale photograph of human teeth, showing the upper and lower arches. The teeth are white and appear to be in good health. The background is a dark, solid color.

Multiple Choice Questions

CHAPTER-2
**GENERAL EMBRYOLOGY AND
GROWTH & DEVELOPMENT OF
OROMAXILLOFACIAL STRUCTURES**

1. After fertilization of ovum, series of cell divisions give rise to an egg cell mass called:
 - A. Placenta
 - B. Morula
 - C. Embryo
 - D. Embryonic disc
2. In humans, the major portion of the egg cell mass forms the:
 - A. Extra embryonic membranes
 - B. Embryo
 - C. Embryonic disc
 - D. Morula
3. 1/4th of the cells of the egg cell mass form a single layer which forms:
 - A. Embryo
 - B. Placenta
 - C. Extra embryonic membrane
 - D. Morula
4. A unique population of cells develop from the ectoderm along the lateral margin and are called:
 - A. Neural crest cells
 - B. Enamel
 - C. Notochord
 - D. Neural fold
5. All of the following are formed by neural crest cells except:
 - A. Cartilage
 - B. Enamel
 - C. Dentin
 - D. Bone
6. Most of the hard palate and all of the soft palate form from the:
 - A. Primary palate
 - B. Secondary palate
 - C. Accessory palate
 - D. None of the above
7. In humans, how many visceral arches are there?
 - A. 2
 - B. 4
 - C. 5
 - D. 6
8. In humans, which visceral arch is rudimentary?
 - A. 2nd
 - B. 4th
 - C. 5th
 - D. 6th
9. The name of the first visceral arch is:
 - A. Mandibular
 - B. Hyoid
 - C. Maxillary
 - D. None of the above
10. Name the second visceral arch:
 - A. Maxillary
 - B. Mandibular
 - C. Hyoid
 - D. None of the above
11. Which of the following statements is/are true?
 - A. Anterior 2/3rd of the tongue is covered by ectoderm and posterior 1/3rd is covered by endoderm
 - B. Thyroid gland forms by invagination of the most anterior endoderm
 - C. Foramen caecum is present at junction of anterior 2/3rd and posterior 1/3rd of the tongue
 - D. All of the above
12. Which of the following statements is/are true?
 - A. About 2/3rd of patients with cleft of the primary palate also have cleft of the secondary palate
 - B. After cleft involving the primary palate, the second most common facial malformation in humans is cleft involving only secondary palate
 - C. Hemifacial microsomia is the third most common facial malformation.
 - D. All of the above
13. Treacher Collins' syndrome is:
 - A. Mandibulofacial dysostosis
 - B. Maxillofacial dysostosis
 - C. Deformity of hair
 - D. None of the above
14. Median rhomboid glossitis is:
 - A. Caused by persistence of Tuberculum impar
 - B. Red and rhomboidal smooth zone of the tongue
 - C. Found in mid line in front of the foramen caecum
 - D. All of the above
15. Which drug causes malformations similar to hemifacial microsomia when taken during early pregnancy?
 - A. Thalidomide
 - B. Nicotinamide
 - C. Procainamide
 - D. Vitamin D

16. **Globulomaxillary cyst is found between:**
 A. Central incisors
 B. Central incisor and lateral incisor
 C. Lateral incisor and canine
 D. Canine and premolar
17. **Every branchial arch contains which of the following structures?**
 A. Branchial artery
 B. Branchiomic nerve
 C. Branchial arch cartilage rod
 D. All of the above
18. **The philtrum of the upper lip is formed largely by:**
 A. Maxillary processes
 B. Globular process
 C. Lateral nasal processes
 D. None of the above
19. **The nerve supply of the muscles of mastication shows that these muscles are derived from the:**
 A. Third branchial arch
 B. Fourth branchial arch
 C. First branchial arch
 D. Second branchial arch
20. **In the embryo, the upper lip is formed by the fusion of the:**
 A. Premaxilla with maxillary processes
 B. Median nasal process with lateral nasal processes
 C. First branchial arch with second branchial arch
 D. Median nasal processes with maxillary processes
21. **The primitive endoderm is formed from the:**
 A. Embryonic disc
 B. Trophoblast
 C. Amniotic cavity
 D. Primitive yolk sac
22. **The neural tube gives rise to the:**
 A. Nervous system
 B. Primitive endoderm
 C. Embryonic disc
 D. Neural groove
23. **The buccopharyngeal membrane is formed:**
 A. When the stomodeum ectoderm contacts the endoderm of the foregut
 B. From fifth branchial arch
 C. From Tuberculum impar
 D. From the primitive gut

24. **A shelf-like extension of the frontonasal process separates the anterior part of the common nasal and oral cavity. The extension is termed the:**
 A. Primitive mouth
 B. Primitive palate
 C. Primitive nasal septum
 D. Stomodeum

ANSWERS

- | | | | | | |
|-------|-------|-------|-------|-------|-------|
| 1. B | 2. A | 3. A | 4. A | 5. B | 6. B |
| 7. D | 8. C | 9. A | 10. C | 11. D | 12. D |
| 13. A | 14. D | 15. A | 16. C | 17. D | 18. D |
| 19. C | 20. D | 21. D | 22. A | 23. A | 24. B |

CHAPTER-3

DEVELOPMENT OF TEETH

1. **Dental lamina is formed when:**
 A. The embryo is 3 weeks old
 B. The embryo is 4 weeks old
 C. The embryo is 5 weeks old
 D. The embryo is 6 weeks old
2. **Which statements is/are true?**
 A. The development of the first permanent molar is initiated at the 4th month of intrauterine life
 B. The second permanent molar is initiated at about the first year after birth
 C. The third molar is initiated at the 4th or 5th years after birth
 D. All of the above
3. **The successor of the deciduous teeth develops from:**
 A. Lingual extension of the dental lamina
 B. Labial extension of the dental lamina
 C. Buccal extension of the dental lamina
 D. Occlusal extension of the dental lamina
4. **The lingual extension of dental lamina is known as:**
 A. Mental lamina
 B. Mandibular lamina
 C. Successional lamina
 D. Maxillary lamina

5. **Life of successional lamina is from the:**
 - A. 5th month in utero to the 10th month of age
 - B. 3rd month in utero to the 10th month of age
 - C. 5th month in utero to the 8th month in utero
 - D. 6 weeks of embryo to the 8th month in utero
6. **Total activity of the dental lamina extends over a period of:**
 - A. At least 3 years
 - B. At least 4 years
 - C. At least 5 years
 - D. At least 6 years
7. **Epithelial pearls are:**
 - A. Remnants of dental lamina
 - B. Remnants of Hertwig's epithelial root sheath
 - C. Remnants of diaphragm
 - D. Remnants of enamel
8. **Epithelial pearls are found:**
 - A. Within the jaw
 - B. In the gingiva
 - C. None of the above
 - D. Both (a) and (b)
9. **The cells of the dental papilla will form:**
 - A. Tooth pulp
 - B. Dentin
 - C. Both of the above
 - D. None of the above
10. **The cells in the dental sac will form:**
 - A. Cementum
 - B. Periodontal ligament
 - C. Dentin
 - D. Both (a) and (b)
11. **The cells in the center of the enamel organ are densely packed and form:**
 - A. The enamel knot
 - B. Strips of cloth
 - C. Surgical knot
 - D. All of the above
12. **Which statement is false?**
 - A. A vertical extension of the enamel knot is formed by enamel cord
 - B. The function of the enamel cord and knot is to act as a reservoir of the dividing cells for the growing enamel organ
 - C. The epithelial enamel organ, the dental papilla and the dental sac are the formative tissues for entire tooth and its supporting structures
 - D. None of the above
13. **The size of the ameloblast cell is:**
 - A. 4 to 5 microns in diameter and about 40 microns in length
 - B. 6 to 8 microns in diameter and about 60 microns in length
 - C. 6 to 8 microns in diameter and about 40 microns in length
 - D. 1 to 2 microns in diameter and about 60 microns in length
14. **The basement membrane that separates the enamel organ and the dental papilla just prior to dentin formation is called the:**
 - A. Advanced bell stage
 - B. Membrana preformativa
 - C. Cap stage
 - D. Enamel knot
15. **Hertwig's root sheath consists of:**
 - A. The outer and inner enamel epithelium only
 - B. The stratum intermedium
 - C. The stellate reticulum
 - D. All of the above
16. **The remnants of epithelial root sheath found in periodontal ligament are called:**
 - A. Enamel pearls
 - B. Enamel knots
 - C. Rests of Malassez
 - D. Epithelial diaphragm
17. **If cells of the epithelial root sheath remain adherent to dentin surface, they may differentiate into ameloblasts and produce enamel. Such droplets of enamel are called:**
 - A. Epithelial pearls
 - B. Enamel pearls
 - C. Rests of Malassez
 - D. Epithelial diaphragm
18. **Development of accessory root canal is due to:**
 - A. The broken continuity of Hertwig's root sheath
 - B. The tongue-like extension of the horizontal diaphragm development
 - C. The apposition of dentin and cementum to the apex of the root
 - D. None of the above

19. Which of the following statements is/are true?
- A. Teeth may develop in abnormal location
 - B. A lack of initiation results in the absence of either single or multiple teeth
 - C. A lack of initiation mostly affects the permanent upper lateral incisor, 3rd molar and lower 2nd premolar
 - D. All of the above
20. Which of the following statements is/are true?
- A. Enamel does not form in the absence of dentin
 - B. In vitamin A deficiency the ameloblast fails to differentiate properly
 - C. In vitamin A deficiency, atypical dentin (osteodentin) is formed
 - D. All of the above
21. Retarded eruption of teeth occurs in persons with:
- A. Hypopituitarism and Hypothyroidism
 - B. Hyperpituitarism
 - C. Hyperthyroidism
 - D. None of the above
22. Shape and size of teeth is determined by which stage?
- A. Initiation
 - B. Histodifferentiation
 - C. Morphodifferentiation
 - D. Apposition
23. Supernumerary teeth are formed due to abnormality in:
- A. Initiation
 - B. Proliferation
 - C. Histodifferentiation
 - D. Morphodifferentiation
24. The final products of the embryonic dental papilla are:
- A. Pulp, dentin and cementum only
 - B. Pulp and dentin only
 - C. Pulp, dentin, cementum and the periodontal ligament
 - D. None of the above
25. After formation of the deciduous tooth germ, the dental lamina:
- A. Becomes atrophic and dormant
 - B. Forms the salivary glands
 - C. Degenerates
 - D. Proliferates lingually to each deciduous tooth germ producing permanent tooth buds
26. The extension of the dental lamina lingual to each deciduous tooth germ is termed the:
- A. Successional dental lamina
 - B. Linguoalveolar sulcus
 - C. Lip furrow band
 - D. Secondary tooth bud
27. The proliferation of the dental lamina beyond the tooth germ of the second deciduous molar is the origin of the:
- A. Major salivary glands
 - B. Peridens
 - C. Tooth germs of the permanent molars
 - D. Distomolars
28. The three permanent molars develop at:
- A. Eight months of fetal life
 - B. At birth, 2½ to 3 years and 7 to 10 years after birth respectively
 - C. Four months of fetal life
 - D. Eight months fetal life, 3 years after birth and 6 years after birth respectively
29. Every tooth germ passes through the following stages:
- A. Dental lamina and bell stage
 - B. Dental lamina, histodifferentiation and apposition of dental tissues
 - C. Dental lamina and bud stage, cap stage, bell stage and apposition of dental tissues
 - D. None of the above
30. The bell stage of tooth development refers to:
- A. Dental lamina
 - B. Apposition of dental tissues
 - C. Proliferation
 - D. Histodifferentiation and morphodifferentiation
31. In the cap stage the following cell types can be recognized in the enamel organ:
- A. Outer enamel epithelium, inner enamel epithelium and stratum intermedium
 - B. Outer enamel epithelium and inner enamel epithelium
 - C. Stellate reticulum and dental papilla
 - D. Outer enamel epithelium, inner enamel epithelium and stellate reticulum
32. The stellate reticulum (enamel pulp) contains:
- A. No intercellular fluid
 - B. Poor quantity of mucopolysaccharide and rich amount of Tomes' fibers
 - C. A large quantity of intercellular fluid
 - D. A small quantity of intercellular fluid

33. **The cap stage contains the following transitory structures:**
- Enamel knot, enamel cord and enamel niche
 - Enamel matrix
 - Stratum intermedium
 - Cervical loop
34. **The inner enamel epithelium develops into the:**
- Stellate reticulum
 - Ameloblasts
 - Odontoblasts
 - Dental papilla
35. **The stratum intermedium:**
- Is highly gelatinous and rich in mucopolysaccharides
 - Does not provide a reserve source of cells for the stratum intermedium
 - Lies between the inner enamel epithelium and stellate reticulum
 - Plays no role in enamel calcification
36. **Prior to formation of enamel matrix the stellate reticulum of the enamel organ:**
- Proliferates
 - Degenerates
 - Becomes considerably narrowed
 - Becomes considerably widened
37. **Remnants of the dental lamina:**
- Do not persist throughout life
 - Decrease in number but persist throughout life
 - Remain constant in number with age
 - Increase in number with age
38. **After dentin and enamel formation have reached the cemento-enamel junction, the cervical loop:**
- Degenerates and disappears
 - Forms the cell-free zone
 - Forms the membrana preformativa
 - Becomes transformed into the epithelial sheath of Hertwig
39. **When the continuity of Hertwig's sheath is destroyed following the beginning of dentin formation the result is?**
- Formation of epithelial rests of Malassez
 - Formation of cementoblasts
 - Development of epithelial diaphragm
 - Differentiation of odontoblasts
40. **Odontoblasts:**
- Form the lateral foramina of the pulp
 - Differentiate only where Hertwig's sheath is present
 - Form interglobular dentin
 - Can differentiate in the absence of Hertwig's sheath
41. **In three-rooted teeth:**
- The diaphragm surrounds three cervical openings
 - Four epithelial fingers grow and fuse
 - Three epithelial fingers grow and fuse and the diaphragm surrounds three cervical openings
 - The diaphragm surrounds four cervical openings
42. **In developing tooth during formation of enamel and primary dentin:**
- The enamel is formed faster than the dentin
 - The enamel is formed slower than the dentin
 - The enamel is formed more cervically than the dentin
 - The ameloblasts and odontoblasts move toward the pulp
43. **The shape of the root is determined by cells from the:**
- Dental papilla
 - Dental organ
 - Dental sac
 - Vestibular lamina
44. **Hertwig's epithelial root sheath induces the formation of:**
- Enamel
 - Coronal dentin
 - Odontoblasts
 - Ameloblasts
45. **During sixth week of embryonic life development of which structure is seen:**
- Hard palate
 - Soft palate
 - Tooth
 - Oral epithelium
46. **The formation of the Hertwig's epithelial root sheath occurs by:**
- Dental lamina
 - Enamel organ
 - Stellate reticulum
 - None of the above

47. The Hertwig's epithelial root sheath helps to:
- Form crown of tooth
 - Form outer and inner enamel epithelium
 - Mould the shape of roots
 - All of the above
48. The droplets of enamel found in area of furcation of the roots of permanent molars are:
- Enamel organ
 - Enamel cracks
 - Enamel pearls
 - Enamel spindle
49. Dentin is formed from:
- Enamel organ
 - Dental papilla
 - Dental sac
 - None of the above
50. Shape of the crown of teeth is controlled by:
- Genes
 - Signaling molecules of genes
 - Growth factors
 - All of the above
51. Which of the following dental tissues is laid down first?
- Enamel
 - Dentin
 - Cementum
 - All the above simultaneously

ANSWERS

1. D 2. D 3. A 4. C 5. A 6. C
 7. A 8. D 9. C 10. D 11. A 12. D
 13. A 14. B 15. A 16. C 17. B 18. A
 19. D 20. D 21. A 22. C 23. A 24. B
 25. D 26. A 27. C 28. B 29. C 30. D
 31. D 32. C 33. A 34. B 35. C 36. C
 37. B 38. D 39. A 40. B 41. C 42. B
 43. B 44. C 45. C 46. B 47. C 48. C
 49. B 50. D 51. B

CHAPTER-4 ENAMEL

1. On the cusps of human molars and premolars, maximum thickness of enamel is about:
- 0 to 1 mm
 - 2 to 3 mm
 - 4 to 5 mm
 - 4 to 6 mm
2. The specific gravity of enamel is:
- 2.4
 - 2.6
 - 2.8
 - 3.0
3. Which of the following statements is/are true?
- Enamel acts like a semipermeable membrane
 - Enamel mainly consists of inorganic material (96%) and small amount of organic substance and water (4%)
 - During development histologic staining reactions of Enamel matrix resemble keratinizing epidermis
 - All of the above
4. Which of the following statements is/are true?
- Enamel is composed of enamel rods, rod sheath and interprismatic substances
 - The number of enamel rods is about 5 million in lower lateral incisors and 12 million in the upper first molar
 - The length of enamel rods is greater than thickness of enamel because of oblique direction and wavy course of the rods.
 - All of the above
5. The diameter of the enamel rods increases from dentino-enamel junction towards the surface of enamel in a ratio of about:
- 1:2
 - 1:3
 - 1:4
 - 1:5

6. In cross sections of human enamel, many rods resemble:
 - A. Triangle
 - B. Fish scales
 - C. Skin scales
 - D. Cow horn
7. Which of the following statements is/are false?
 - A. The average thickness of the crystal of human enamel is about 30 nanometers
 - B. The average width of the crystal of human enamel is about 90 nanometer
 - C. Both of the above
 - D. None of the above
8. The striations are more pronounced in enamel that is:
 - A. Sufficiently calcified
 - B. Insufficiently calcified
 - C. Fully calcified
 - D. Not at all calcified
9. The enamel rods are segmented because the enamel matrix is formed in rhythmic manner. In humans these segments seem to be of a uniform length of about:
 - A. 2 microns
 - B. 4 microns
 - C. 6 microns
 - D. 8 microns
10. The arrangement of enamel rods in the permanent teeth, in the cervical region, deviates from the horizontal in an:
 - A. Apical direction
 - B. Occlusal direction
 - C. Incisal direction
 - D. All of the above
11. In an oblique plane the bundles of rods seem to intertwine more irregularly in the region of cusps or incisal edge, this optical appearance of enamel is called:
 - A. Gnarled enamel
 - B. Striation
 - C. Hunter-Schreger bands
 - D. Incremental lines of Retzius
12. The change in the direction of enamel rods is responsible for appearance of the:
 - A. Gnarled enamel
 - B. Hunter-Schreger bands
 - C. Both of the above
 - D. None of the above
13. The Brownish bands i.e. the successive apposition of layer of enamel during the formation of crown is known as:
 - A. Gnarled enamel
 - B. Incremental line of Retzius
 - C. Hunter-Schreger bands
 - D. All of the above
14. In transverse sections of a tooth, the incremental line of Retzius appears as:
 - A. Concentric circle
 - B. Oblique line
 - C. Zigzag line
 - D. Cracks
15. Perikymata are transverse, wave-like grooves, believed to be the external manifestation of the:
 - A. Striae of Retzius
 - B. Gnarled enamel
 - C. Hunter-Schreger bands
 - D. All of the above
16. Perikymata are absent in the:
 - A. Postnatal cervical part
 - B. All permanent teeth
 - C. All deciduous teeth
 - D. Occlusal part of the deciduous teeth
17. The enamel of deciduous teeth develops partly before and partly after birth, the boundary between the two portions is known as:
 - A. Basal lamina
 - B. Incremental line
 - C. Neonatal line
 - D. Postnatal line
18. A delicate membrane which covers the entire crown of the newly erupted tooth is called as:
 - A. Primary enamel cuticle
 - B. Nasmyth's membrane
 - C. Pellicle
 - D. Both (a) and (b)
19. Thin, leaf-like structures that extend from enamel surface towards the dentino-enamel junction are called:
 - A. Enamel tufts
 - B. Gnarled enamel
 - C. Enamel lamellae
 - D. Enamel spindle

20. Which of the following statements is/are true about enamel lamellae?
- Type A lamellae are composed of poorly calcified rod segments
 - Type B lamellae consist of degenerated cells
 - Type C lamellae arise in erupted teeth where cracks are filled with organic matter (more common)
 - All of the above
21. Enamel tufts which arise at the dentinoenamel junction and reach into enamel to about 1/5th to 1/3rd of its thickness are:
- Hypocalcified enamel rods and interprismatic substances
 - Hypercalcified enamel rods only
 - Hypomineralized enamel rods only
 - Similar to surface structures
22. In the mature dental tissues, the only tissue whose cells are lost is:
- Dentin
 - Pulp
 - Cementum
 - Enamel
23. Which statement about enamel prism rod is not correct?
- The prisms run from the dentinoenamel junction to the crown surface
 - The maximal width of a prism in horizontal section is 5 μm
 - Prisms are wider near the dentinoenamel junction than near the crown surface
 - The lengths of all enamel prisms are equal
24. The enamel organ:
- Promotes the differentiation of the dental papilla
 - Does not form the enamel
 - Does not narrow after the first layer of dentin has formed
 - Does not promote the differentiation of the dentin
25. After the enamel is completely formed and has undergone maturation:
- The ameloblasts form the primary enamel cuticle
 - The ameloblasts become much longer
 - The enamel organ becomes a widened layer of stratified squamous epithelium
 - None of the above
26. During enamel maturation:
- Calcification ceases
 - Additional calcification occurs until complete calcification is present
 - The ameloblasts play no role
 - Additional enamel matrix is produced
27. The maturation of the enamel starts at:
- Any point between occlusal surface and cervical region
 - Occlusal surface of the crown and progresses toward cervical region
 - Cervical region of the crown
 - None of the above
28. The formation of dentin:
- Decreases the blood supply
 - Is not based on tissue interdependence
 - Is apparently a necessary stimulus for the formation of enamel
 - Is not a necessary stimulus for the formation of enamel
29. The perikymata are:
- Elevations between the imbrication lines of Pickerill
 - Present in the cementum
 - Present in the enamel
 - Present in the dentin
30. The incremental lines of Retzius are seen in ground transverse sections:
- As white bands not in concentric rings
 - As brown bands not in concentric rings
 - As concentric circles in the enamel which resemble rings in a tree trunk
 - Running inward from the dentinoenamel junction
31. The enamel rod sheath:
- Has cross striations
 - Has a lesser amount of inorganic matter than the enamel rod
 - Is a more calcified enamel peripheral shell around each enamel rod
 - None of the above
32. The enamel rods in the incisal and cuspal areas are:
- Vertical in direction
 - Horizontal in direction
 - Inclined apically
 - Oblique and then horizontal in direction

- 33. Enamel rods:**
- A. Have only a concave surface in cross section
 - B. Increase in thickness from the dentinoenamel junction to the surface of enamel
 - C. Are always the same thickness regardless of location
 - D. Decrease in thickness from the dentinoenamel junction to the enamel surface
- 34. Gnarled enamel is:**
- A. Present only at the cemento enamel junction
 - B. Present only at the dentinoenamel junction
 - C. Present only in deciduous teeth
 - D. Due to markedly wavy and irregular enamel rods at the cusps or incisal edges
- 35. Ionic interchanges between matured/erupted enamel and saliva:**
- A. Occur throughout life
 - B. Occur only for two years following eruption
 - C. Cease to occur in young adult life
 - D. Do not occur
- 36. Matured enamel is subjected to:**
- A. Erosion and attrition only
 - B. Caries, erosion and attrition
 - C. Caries and attrition only
 - D. Caries and erosion only
- 37. Enamel contains:**
- A. 96% organic substance
 - B. 1.5% inorganic substance
 - C. 100% organic substance
 - D. None of the above
- 38. Enamel contains:**
- A. 96% water
 - B. 1% water
 - C. 5% water
 - D. None of the above
- 39. Enamel is:**
- A. The only tissue whose formation does not cease
 - B. Made up of 100% inorganic material
 - C. The only calcified tissue in mammals of epithelial origin
 - D. None of the above
- 40. Enamel lamellae:**
- A. Are club-like swellings extending into the enamel for short distance
 - B. Extend for short distances from the dentinoenamel junction toward the enamel surface
 - C. Represent a pathway for proteolytic bacteria to the dentinoenamel junction
 - D. Contain less organic matter than the enamel proper
- 41. Enamel tufts:**
- A. Form a pathway for proteolytic bacteria to extend to the dentinoenamel junction
 - B. Contain less organic material than normal enamel
 - C. Contain highly calcified enamel rods and interprismatic substance
 - D. Are areas of imperfect calcification extending for short distances from the dentinoenamel junction toward the enamel surface
- 42. The Hunter-Schreger bands are:**
- A. Not visible in ground sections
 - B. An illusion in reflected light
 - C. Due to alternating directions of successive groups of enamel rods in reflected light
 - D. Not visible in reflected light
- 43. The uptake of fluorine by enamel:**
- A. Fails to taper off
 - B. Is the same during the first years following enamel formation and in later years
 - C. Is very slow during the first years following enamel formation
 - D. Is very rapid during the first years following enamel formation
- 44. Nitrogen is present in the surface enamel in:**
- A. Greater concentration in young teeth
 - B. Greater concentration in older teeth
 - C. Lower concentration in older teeth
 - D. Lower concentration at the dentinoenamel junction
- 45. The addition of 1 PPM of fluorine (F) to the drinking water causes:**
- A. An increment of about 300 PPM in surface enamel
 - B. A decrement of about 100 PPM in surface enamel
 - C. An increase in aluminum in the surface enamel
 - D. No change in F concentration in surface enamel

46. The principal blood supply of ameloblasts during most of enamel formation is from:
- Enamel
 - Dental pulp
 - Reduced dental organ
 - Dentin
47. In cross section enamel prisms approximately have an average thickness of:
- 3 microns
 - 300 angstroms
 - 3 angstroms
 - 30 microns
48. The long axis of enamel crystals is:
- Parallel to the prism axis
 - Parallel to the tooth surface
 - Perpendicular to the tooth surface
 - Both (a) and (c)
49. The gnarled enamel is observed in:
- In cervical area
 - Only in the canines
 - In carious areas
 - In cuspal areas
50. The enamel spindle is:
- Part of Tomes' enamel process
 - The distal part of an odontoblastic process in enamel
 - An area with a high collagen content
 - A hypercalcified area
51. Hunter Schreger bands appear due to:
- Daily growth rhythms
 - Increased organic content in some areas
 - Dietary change after birth
 - Changes in the enamel prism orientation from one group of prisms to the next group of prisms
52. Tomes' enamel process:
- Remains in the mature enamel as the ameloblastic process
 - Extends into the dentin
 - Forms at the proximal end of the ameloblast
 - Is apical to the terminal bar/area at the secretory apical end of ameloblast
53. Enamel that can withstand severe masticatory force is:
- Root enamel
 - Cervical enamel
 - Gnarled enamel
 - Enamel lamellae.
54. Poorly calcified enamel is present in:
- Perikymata
 - Gnarled enamel
 - Enamel lamellae
 - Enamel prisms.
55. In the human body the hardest calcified tissue is:
- Dentin
 - Cementum
 - Bone
 - Enamel
56. The percentage of the inorganic component of the enamel is:
- 96%
 - 95%
 - 92%
 - 90%
57. The dentinoenamel junction is in the form of:
- A scalloped line with convexities towards the enamel.
 - A scalloped line with the convexities of the scallops towards dentin
 - A straight line
 - None of the above.
58. Direction of the enamel rods in the deciduous and permanent teeth is:
- Same throughout
 - Different at cervical third
 - Different at occlusal and incisal thirds
 - None of the above
59. For the control of dental caries fluoride level in drinking water should be:
- 8 PPM
 - 4 PPM
 - 2 PPM
 - 1 PPM
60. Enamel tufts:
- Arise from the surface of enamel and reach at dentinoenamel junction
 - Arise at the dentinoenamel junction and proceed into the enamel towards surface
 - Arise at dentin pulp junction and reach upto enamel
 - Arise from the surface of enamel and reach upto the pulp.

61. The angle between the diazones and parazonies is approximately:
- 10 degree
 - 40 degree
 - 70 degree
 - 100 degree
62. The mean rate of enamel formation is:
- 1.0 micron per day
 - 3.5 micron per day
 - 6.5 micron per day
 - 10.0 micron per day
63. Which of the following is present at dentinoenamel junction and help in cell signaling?
- Ameloblastin
 - Enamelin
 - Tuftelin
 - None of the above
64. In transition stage following changes take place:
- Reduced height of ameloblast
 - Secretion of enamel stops completely
 - Removal of amelogenin starts
 - All of the above
65. What is the thickness of the layer of prismless enamel found in primary teeth?
- 25 microns
 - 50 microns
 - 75 microns
 - 100 microns

ANSWERS

- | | | | | | |
|-------|-------|-------|-------|-------|-------|
| 1. B | 2. C | 3. D | 4. D | 5. A | 6. B |
| 7. D | 8. B | 9. B | 10. A | 11. A | 12. C |
| 13. B | 14. A | 15. A | 16. D | 17. C | 18. D |
| 19. C | 20. D | 21. A | 22. D | 23. D | 24. A |
| 25. A | 26. B | 27. B | 28. C | 29. C | 30. C |
| 31. B | 32. A | 33. B | 34. D | 35. A | 36. B |
| 37. D | 38. D | 39. C | 40. C | 41. D | 42. C |
| 43. D | 44. B | 45. A | 46. C | 47. B | 48. D |
| 49. D | 50. B | 51. D | 52. D | 53. C | 54. C |
| 55. D | 56. A | 57. B | 58. B | 59. D | 60. B |
| 61. B | 62. B | 63. C | 64. D | 65. A | |

CHAPTER-5 DENTIN

1. Which of the following statements is/are true about Dentinoenamel junction (DEJ)?
- The surface of dentin at the DEJ is pitted
 - This relation assumes the firm hold of the enamel cap on dentin surface
 - DEJ appears as a scalloped line
 - All of the above
2. Which of the following statements is/are true about Dentinoenamel junction (DEJ). DEJ is observed a?
- Hypermineralized zone about 30 microns thick"
 - Hypomineralized zone about 30 microns thick"
 - Hypermineralized zone about 60 microns thick"
 - Hypomineralized zone about 60 microns thick"
3. Odontoblastic process which passes across the dentinoenamel junction into enamel is called:
- Gnarled enamel
 - Enamel spindle
 - Enamel lamellae
 - Enamel tufts
4. Enamel spindle in ground section of dried teeth appears dark in:
- Transmitted light
 - Reflected light
 - Polarized light
 - All of the above
5. In ground section of dried teeth, which of the following appear light in transmitted light?
- Enamel spindle
 - Dead tracts
 - Sclerotic dentin
 - All of the above
6. Dental lamellae may also be predisposing locations for caries because they contain:
- Much organic material
 - Less organic material
 - Much inorganic material
 - None of the above

7. At the free border of the enamel organ, the outer and inner enamel epithelial layers are continuous and are reflected into one another as the:
- Facial loop
 - Buccal loop
 - Cervical loop
 - Terminal bar
8. Which of the following statements is/are true?
- Distal ends of enamel rods are not in direct contact with the dentin
 - The projection of the ameloblasts into enamel matrix has been named Tomes' process
 - Tomes' process contains rough endoplasmic reticulum and mitochondria
 - All of the above
9. Which of the following statements is/are false?
- At least two ameloblasts are involved in the synthesis of each enamel rod
 - The bulk of the 'head' of each enamel rod is formed by one ameloblast while the contribution of 3 ameloblasts is to the component of the tail of each rod.
 - Each ameloblast contributes to four different rods
 - None of the above
10. The dentin is formed:
- Slightly before the enamel
 - Slightly after the enamel
 - Slightly after the cementum
 - Slightly after periodontal ligament
11. Physically and chemically the dentin closely resembles:
- Enamel
 - Cementum
 - Bone
 - Amalgam
12. The formula of hydroxyapatite is/are:
- $3\text{Ca}_3(\text{PO}_4)_2 \cdot \text{Ca}(\text{OH})_2$
 - $[\text{Ca}_{10}(\text{OH})_2(\text{PO}_4)_6]$
 - $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$
 - Both (a) and (b)
13. Hydroxyapatite crystal of dentin is:
- Much smaller than enamel
 - Much larger than enamel
 - Same as enamel
 - None of the above
14. The ratio between outer and inner surfaces of dentin is about:
- 1:1
 - 1:2
 - 2:1
 - 5:1
15. The ratio between the number of tubules per unit area on the pulpal and outer surfaces of dentin is about:
- 1:1
 - 1:2
 - 4:1
 - 1:4
16. The main body of dentin is:
- Peritubular dentin
 - Intertubular dentin
 - Pre-dentin
 - Tomes' fibers
17. The first formed dentin which is not mineralized is:
- Peritubular
 - Intertubular
 - Pre-dentin
 - Odontoblastic process
18. The dentin that immediately surrounds the dentinal tubules is:
- Peritubular dentin
 - Intertubular dentin
 - Pre-dentin
 - First formed dentin
19. The cytoplasmic extension of the odontoblast into dentinal tubules is called as:
- Odontoblastic process
 - Tomes' fibers
 - None of the above
 - Both (a) and (b)
20. The odontoblastic processes are composed of:
- Microtubules 20 microns in diameter
 - Small filaments 5 to 7.5 microns in diameter
 - Mitochondria
 - All of the above
21. Secondary dentin is:
- First formed dentin
 - Dentin formed before root completion
 - Dentin formed after root completion
 - Circumpulpal dentin
22. The incremental lines of Von Ebner are in:
- Enamel
 - Dentin
 - Bone
 - Cementum
23. Some of the incremental lines are accentuated because of disturbances in the matrix and mineralization process and are known as Contour lines (Owen), and are found in:
- Enamel
 - Dentin
 - Bone
 - Cementum
24. Contour lines (Owen) are:
- Hypocalcified bands
 - Hypercalcified bands
 - Non-mineralized bands
 - None of the above

25. Sometimes mineralization of dentin begins in globular area; these hypomineralized zones are known as:
 A. Interglobular dentin B. Granular layer
 C. Peritubular dentin D. Intertubular dentin
26. Tomes' granular layer is caused by a coalescing and looping of the terminal portions of the dentinal tubules found in:
 A. Enamel B. Crown dentin
 C. Root dentin D. Cementum
27. Due to extensive abrasion, erosion, caries or operative procedures, the odontoblast deposits dentin which is called:
 A. Reparative dentin B. Secondary dentin
 C. Tertiary dentin D. (a) and (c) are correct
28. Dentin areas characterized by degenerated odontoblastic processes which appear white in reflected light are called as:
 A. Dead tracts B. Sclerotic Dentin
 C. Transparent dentin D. Mantle dentin
29. Dead tracts are mostly found in:
 A. Only deciduous teeth
 B. Only incisors
 C. Older teeth
 D. Carious teeth
30. Which of the following statements is/are true about sclerotic dentin?
 A. It leads to protective changes in the dentin itself
 B. The mineralization is very similar to peritubular dentin
 C. It appears dark in reflected light
 D. All of the above
31. How much initial increment of dentin is formed during dentinogenesis until the crown is formed?
 A. 3 mm/day B. 4 mm/day
 C. 5 mm/day D. 6 mm/day
32. Korff's fibers are found in:
 A. Primary dentin B. Secondary dentin
 C. Sclerotic dentin D. Transparent dentin
33. What is the rate of reparative dentin formation after cavity preparation in dentin?
 A. 4 microns per day B. 6 microns per day
 C. 8 microns per day D. 10 microns per day
34. The apatite crystals of dentin are:
 A. 300 times larger than those formed in enamel
 B. 300 times smaller than those formed in enamel
 C. 3000 times smaller than those formed in enamel
 D. 3000 times larger than those formed in enamel
35. How many living cells are damaged when 1 mm² of dentin is exposed?
 A. About 10,000 living cells
 B. About 20,000 living cells
 C. About 30,000 living cells
 D. About 40,000 living cells
36. The rapid penetration and spread of caries in the dentin is the result of the:
 A. Tubule system in the dentin
 B. Canaliculi system in the dentin
 C. Haversian system in the dentin
 D. All of the above
37. The sulcular epithelium is:
 A. Keratinized B. Nonkeratinized
 C. Orthokeratinized D. Parakeratinized
38. The remnant of the primary enamel cuticle after eruption is referred to as:
 A. Nasmyth's membrane
 B. Pellicle
 C. Primary attachment epithelium
 D. All of the above
39. The separation of primary attachment epithelium from the enamel is termed as:
 A. Active eruption
 B. Passive eruption
 C. Fast eruption
 D. Functional eruption
40. The clinical crown is the part of the tooth which is:
 A. Covered by enamel
 B. Covered by gingiva
 C. Exposed in the oral cavity
 D. None of the above
41. Compared with intertubular dentin, peritubular dentin is characterized by having:
 A. Less inorganic salt content and a heavier collagenous matrix
 B. More inorganic salt content and a finer collagenous matrix
 C. More fluid content and a different refractive index
 D. Fewer fibers and being more stainable
42. The most important property of clinical significance of dentin of the tooth is that it:
 A. Is softer than enamel
 B. Protects the pulp
 C. Supports the enamel
 D. Is more resilient than enamel

43. **Growth in height of the alveolar process is dependant upon:**
- A. Growth of alveolar process with the eruption of teeth.
 - B. Condylar growth
 - C. Growth of the upper face
 - D. Growth of the cranial base
44. **When teeth fail to develop in the jaw, the alveolar process?**
- A. Shows partial growth
 - B. Fails to form
 - C. Continues to grow
 - D. Shows excessive growth
45. **The differentiation of odontoblasts and formation of dentin and enamel first begins at the:**
- A. Region of the cervical loop
 - B. Incisal edge or tip of cusps
 - C. Midway between the incisal edge and cervical loop
 - D. None of the above
46. **Growth centers in tooth development are located at:**
- A. Any region between the incisal edge and cervical loop
 - B. The incisal edge and tip of the cusps
 - C. The region of the cervical loop
 - D. Dentinoenamel junction under the cusp tip
47. **In the formation of dentin:**
- A. Odontoblasts fail to recede towards the pulp
 - B. Korff's fibers form the odontoblasts
 - C. Korff's fibers are initially deposited dentin at the cusp tips
 - D. Korff's fibers become homogenized and form predentin
48. **Predentin consists of:**
- A. Mineral salts and cementing substance
 - B. Cementing substance
 - C. Calcospherites
 - D. None of the above
49. **When odontoblasts recede towards the pulp, they leave a part of their cytoplasm termed?**
- A. Korff's fibers
 - B. Tomes' process within the formed dentin
 - C. Membrana preformativa
 - D. None of the above
50. **Dentin consists of:**
- A. Cells, collagen fibers and cementing substance
 - B. Cementing substance
 - C. Intercellular substances
 - D. None of the above
51. **Which is highly calcified dentin?**
- A. Interglobular dentin
 - B. Tomes' granular layer
 - C. Peritubular dentin
 - D. Intertubular dentin
52. **Dentin in which the tubules are calcified is termed as:**
- A. Physiological secondary dentin
 - B. Dead tract dentin
 - C. Interglobular dentin
 - D. Sclerotic dentin
53. **Korff's fibers are:**
- A. The same as enamel spindles
 - B. The same as the enamel tufts
 - C. Related to reticular fibers
 - D. Intracellular
54. **Important characteristic of mantle dentin is presence of:**
- A. Enamel spindles
 - B. Hertwig's sheath
 - C. Elastin fibers
 - D. Korff's fibers
55. **Initially the neonatal line is:**
- A. Eliminated by fluoridated water
 - B. Hypercalcified
 - C. Found in all teeth
 - D. Hypocalcified
56. **The percentage of the organic and inorganic components of dentin is:**
- A. 35% organic and water and 65% inorganic
 - B. 35% inorganic and 65% organic and water
 - C. 60% organic and water and 40% inorganic
 - D. 50% organic and water and 50% inorganic
57. **The odontoblastic process is surrounded by the ring shaped transparent zone called as:**
- A. Tubular dentin
 - B. Peritubular dentin
 - C. Intertubular dentin
 - D. Ring dentin
58. **The diameters of the dentinal tubules are:**
- A. Smaller near the pulpal cavity
 - B. Larger at their outer ends
 - C. Larger near the pulp cavity
 - D. Are same from pulp cavity to the outer ends.

59. The most peripheral part of the primary dentin is:
 A. Intertubular dentin
 B. Predentin
 C. Mantle dentin
 D. Circumpulpal dentin
60. Tertiary dentin is:
 A. Reparative dentin' B. Response dentin
 C. Reactive dentin D. All of the above
61. When calcospherites fail to fuse during calcification of dentin matrix:
 A. Transparent dentin is formed
 B. Irregular dentin is formed
 C. Interglobular dentin is formed
 D. Sclerotic dentin is formed.
62. Odontoblast cell bodies are present in the:
 A. Enamel B. Dentin
 C. Pulp D. Cementum
63. Which of the following gene is implicated for odontoblastic differentiation?
 A. Msx -1 B. Msx-2
 C. Alx-3 D. MAPIB
64. Which of the following gene is implicated for dentin mineralization?
 A. Msx 1 B. PHEX
 C. Msx-2 D. Alx-3
65. von Korff's fibers contain:
 A. Type I Collagen
 B. Type II Collagen
 C. Type III Collagen
 D. None of the above
66. Reparative dentin is laid down by:
 A. Newly differentiated odontoblasts
 B. Old odontoblasts survived after injury
 C. Both the above
 D. None of the above.
67. Reactionary or regenerated dentin is laid down by:
 A. Odontoblasts survived by any injury
 B. Newly differentiated odontoblasts
 C. Both the above
 D. None of the above
68. Dentin phosphophoryn is not present in:
 A. Primary dentin B. Secondary dentin
 C. Tertiary dentin D. None of the above
69. Important factor/s in mineralization of dentin is/ are:
 A. Odontoblast secret dentin phosphoprotein and osteonectin
 B. Osteopoutin promote mineralization of dentin
 C. Both the above.
 D. None of the above
70. Which statement is not correct?
 A. Fluoride increases the hardness of dentin if incorporated during dentinogenesis
 B. Ridiculer dentin is less mineralized and slowly formed in comparison to coronal dentin
 C. Deficiency of Vitamin D does affect the formation of dentin
 D. Deficiency of Vitamin D does not affect the formation of teeth.
71. The average diameter of coronal dentinal tubules near the pulp is:
 A. 0.2-0.5 microns B. 2-3 microns
 C. 0.2 -0.3 microns D. 4-7 microns

ANSWERS

- | | | | | | |
|-------|-------|-------|-------|-------|-------|
| 1. D | 2. A | 3. B | 4. A | 5. C | 6. A |
| 7. C | 8. D | 9. D | 10. A | 11. C | 12. D |
| 13. A | 14. D | 15. C | 16. B | 17. C | 18. A |
| 19. D | 20. D | 21. C | 22. B | 23. B | 24. A |
| 25. A | 26. C | 27. D | 28. A | 29. C | 30. D |
| 31. B | 32. A | 33. A | 34. B | 35. C | 36. A |
| 37. B | 38. A | 39. B | 40. C | 41. B | 42. D |
| 43. A | 44. A | 45. B | 46. D | 47. C | 48. D |
| 49. D | 50. D | 51. C | 52. D | 53. C | 54. D |
| 55. D | 56. A | 57. B | 58. C | 59. C | 60. D |
| 61. C | 62. C | 63. D | 64. B | 65. C | 66. A |
| 67. A | 68. C | 69. C | 70. C | 71. B | |

CHAPTER-6 DENTAL PULP

1. How many pulp organs are normally found in every person?
 A. 22 B. 32
 C. 52 D. 62

2. **The total volume of all the permanent pulp organs is:**
 - A. 0.02 cc
 - B. 0.006 cc
 - C. 0.068 cc
 - D. 0.38 cc
3. **The total volume of pulp organ of maxillary first molar is:**
 - A. 0.006 cc
 - B. 0.007 cc
 - C. 0.068 cc
 - D. 0.38 cc
4. **The total volume of pulp organ of mandibular central incisor is:**
 - A. 0.006 cc
 - B. 0.007 cc
 - C. 0.012 cc
 - D. 0.014 cc
5. **The average diameter of the apical foramen of the mandibular and maxillary teeth in adults is:**
 - A. 0.3 mm and 0.4 mm respectively
 - B. 0.6 mm and 0.7 mm respectively
 - C. 0.8 mm and 0.9 mm respectively
 - D. 0.8 mm and 0.6 mm respectively
6. **Weil's zone of pulp is a:**
 - A. Cell-rich zone
 - B. Cell-free zone
 - C. Fibroblast zone
 - D. Mesenchymal cell zone
7. **The most numerous cell type in the pulp is:**
 - A. Fibroblast
 - B. Cementoblast
 - C. Odontoblast
 - D. Defense cell
8. **In the young pulp the cells, which divide and are active in protein synthesis are known as:**
 - A. Fibroblasts
 - B. Fibrocytes
 - C. Odontoblasts
 - D. Histiocytes
9. **The primary cells in the very young pulp, which may become odontoblasts, fibroblasts or macrophages when needed, are:**
 - A. Undifferentiated mesenchyme cells
 - B. Defense cells
 - C. Plasma cells
 - D. Rough endoplasmic reticulum
10. **The anastomosis which occurs in pulp is:**
 - A. Venous-venous
 - B. Arteriole-venous
 - C. Both (a) and (b)
 - D. None of the above
11. **The second most prominent cell in the pulp is:**
 - A. Fibroblast
 - B. Odontoblast
 - C. Cementoblast
 - D. Neutrophil
12. **Lymph vessels draining the pulp and periodontal ligament of mandibular anterior teeth pass to the:**
 - A. Submental lymph nodes
 - B. Submandibular lymph nodes
 - C. Deep cervical lymph nodes
 - D. All of the above
13. **Sensation of pain in pulp is mediated by:**
 - A. Large myelinated fibers
 - B. Large unmyelinated fibers
 - C. Small unmyelinated fibers
 - D. None of the above
14. **The peripheral axons form a network of nerves located adjacent to the cell rich zone in pulp known as:**
 - A. 5 HT
 - B. Plexus of Rashkow
 - C. Zone of Weil
 - D. Brachial plexus
15. **Sensory response in the pulp can differentiate between:**
 - A. Heat and pain
 - B. Touch and heat
 - C. Pressure and chemicals
 - D. None of the above
16. **Which of the following statements is/are true?**
 - A. Pulpal pressure is among the highest of the body tissues
 - B. Sensory nerves in the pulp respond with pain to all stimuli
 - C. Function of pulp is inductive, formative, nutritive, protective and defensive
 - D. All of the above
17. **The average length of time a deciduous pulp functions in the oral cavity is only about:**
 - A. 8.3 years
 - B. 10.3 years
 - C. 11.3 years
 - D. 13.3 years
18. **The maximum life of the deciduous pulp including both prenatal and postnatal times of development and the period of regression is approximately:**
 - A. 8.3 years
 - B. 9.6 years
 - C. 10.6 years
 - D. 12.6 years
19. **Which of the following statements is/are true about regressive changes of pulp?**
 - A. Mitochondria and endoplasmic reticulum are reduced in number and size
 - B. Fibrosis of pulp
 - C. Pulp stones and diffuse calcifications of pulps
 - D. All of the above

20. Calcification in the walls of blood vessels in aging pulp is found most often in the region near the:
- Coronal portion of root
 - CEJ of root
 - Apical foramen
 - Pulp chamber
21. Calcification of thrombi in blood vessels is called:
- True denticle
 - Diffuse calcification
 - Phlebolith
 - All of the above
22. The structures similar to dentin in that they exhibit dental tubuli containing the process of odontoblasts, are rarely found and located close to the apical foramen, are:
- True denticles
 - False denticles
 - Cementicles
 - None of the above
23. Which of the following statements is/are true?
- Diffuse calcifications are usually found in the root canal and less often in coronal pulp
 - Denticles are seen more frequently in the coronal pulp
 - 90% of teeth in persons over the age of 50 years contain calcification of some type
 - All of the above
24. The tooth pulp is initially called the:
- Predentin
 - Dental papilla
 - Subpulpal dentin
 - Pulp polyp
25. At the location of future incisor the development of the dental pulp begins at about:
- 4th week of embryonic life
 - 6th week of embryonic life
 - 8th week of embryonic life
 - 9th week of embryonic life
26. The cell of the dental pulp that has the potential for giving rise to several distinctly different types of cells is the:
- Endothelial cell
 - Mesenchymal cell
 - Schwann cell
 - Macrophage
27. After completion of root formation, the remainder of the dental papilla becomes the:
- Hertwig's sheath
 - Dental pulp
 - Dental follicle
 - Enamel organ
28. The cells that line the pulp chambers of newly erupted teeth are:
- Ameloblasts
 - Odontoblasts
 - Fibroblasts
 - Cementoblasts
29. The function of non myelinated sympathetic nerve endings in the pulp is to:
- Form a plexus in the cell free zone of Weil
 - Elicit only a pain response
 - Alter the blood flow to the pulp
 - Enter the dentinal tubules
30. The coronal pulp has:
- Two surfaces
 - Four surfaces
 - Five surfaces
 - Six surfaces
31. The majority of the nerves that enter the pulp are:
- Myelinated
 - Non-myelinated
 - Both of the above
 - None of the above
32. Dental pulp functions:
- To protect the tooth by its phagocytic cells
 - As a source of odontoblasts
 - To provide sensitivity to heat, cold and pressure.
 - All of the above
33. Wandering cells of pulp are:
- Lymphoid wandering cells
 - Histiocytes
 - Fibroblasts
 - Undifferentiated mesenchymal cells.
34. True denticles:
- Have dental tubules and the processes of the odontoblasts.
 - Are usually located close to the apical foramen.
 - May be induced by fragments of epithelial root sheath
 - All of the above.
35. Which of the following statements is / are true?
- The free denticles are entirely surrounded by pulp tissue.
 - Attached denticles are partly fused with the dentin.
 - Embedded denticles are entirely surrounded by dentin.
 - All are correct.
36. The incidence and size of pulp stone:
- Decrease with age
 - Increase with age
 - There is no effect of age
 - None of the above
37. The vitality of the pulp depends upon:
- Nerve supply
 - Blood supply
 - Both of the above
 - None of the above

38. When compared with a very young pulp the more aged pulp contains?
- Fewer cells and fewer collagen fibers
 - More cells and fewer collagen fibers
 - Fewer cells and more collagen fibers
 - More cells and more collagen fibers
39. Which cell type is usually not found in dental pulp?
- Small lymphocytes and plasma cells
 - Fat cells
 - Histiocytes and macrophages
 - Fibroblasts and undifferentiated mesenchymal cells.
40. In an inflamed pulp usually there are certain cells which are associated with small blood vessels having large and prominent nucleus, these are:
- Lymphocytes
 - Mast cells
 - Histiocytes
 - Fibroblasts
41. The blood flow in vessels of the pulp as compared to most of the other organs of the body is:
- Very slow
 - Faster
 - Almost nil
 - Equal
42. The proof of presence of lymph capillaries in the pulp are on the basis of:
- Absence of basal lamina adjacent to the endothelium.
 - Absence of red blood cells and presence of lymphocytes in certain vessels.
 - Injected fine particulate substances subsequently found in some thin walled vessels other than blood vessels
 - All of the above.
43. Which of the following fibers are not found in dental pulp:
- Argyrophilic fibers
 - Collagen fibers
 - Elastic fibers
 - None.
44. Which statement is wrong:
- In pulp fibroblasts help in the inflammatory and healing process by the secretion of growth factors and cytokines.
 - In pulp fibroblasts help in the inflammatory and healing process by the secretion of colony stimulating factors, FC F -2 and VE GF.
 - Both the above.
 - None of the above.
45. Dendritic cells in pulp are present in the areas affected by:
- Caries
 - Attrition
 - Restorative procedure
 - All of the above
46. Which statements is/are correct:
- Plasma cells in the pulp produce antibodies
 - Pulpal stem cells are pluripotent cells.
 - Both of above
 - None of the above.
47. Which of the following statement is wrong?
- In pulp nerve ending are located close to the odontoblast
 - In pulp nerve endings are located away from the odontoblasts.
 - In mild pulpal injury molecules like kinetin, actin and myosin produce dense skeletal network.
 - Injured cells in the pulp release the calcium which help in migration of cell.
48. In which week of intrauterine life nerve fibers in the dental follicle are first seen:
- Sixth week
 - Eleventh week
 - Eighteenth week
 - Twenty fourth week
49. In which week of intrauterine life nerve fibres in the dental papilla are first seen:
- Sixth week
 - Twelveth week
 - Eighteenth week
 - Twentyfourth week.
50. Which of the following material form thick dentin bridge and produce less inflammation, necrosis and hyperemia?
- Zinc oxide Eugenol
 - Calcium hydroxide
 - Mineral trioxide aggregate
 - Polyantibiotic paste
51. Pulp capping material of future are bioactive molecules like:
- Bone morphogenic protein
 - Purified dentin protein
 - Both the above
 - None of the above.
52. Which of the following check the vitality of the tooth by recording the pulpal blood flow?
- Laser doppler flowmetry
 - Transmitted light photoplethysmography.
 - Both the above
 - None of the above.

ANSWERS

- | | | | | | |
|-------|-------|-------|-------|-------|-------|
| 1. C | 2. D | 3. C | 4. A | 5. A | 6. B |
| 7. A | 8. B | 9. A | 10. C | 11. B | 12. A |
| 13. A | 14. B | 15. D | 16. D | 17. A | 18. B |
| 19. D | 20. C | 21. C | 22. A | 23. D | 24. B |
| 25. C | 26. B | 27. B | 28. B | 29. C | 30. D |
| 31. B | 32. D | 33. B | 34. A | 35. D | 36. B |
| 37. B | 38. C | 39. B | 40. C | 41. B | 42. D |
| 43. C | 44. C | 45. D | 46. C | 47. B | 48. B |
| 49. C | 50. C | 51. C | 52. C | | |

CHAPTER-7
CEMENTUM

- Cementum was first demonstrated microscopically in:**
 - 1835
 - 1935
 - 1895
 - 1995
- The cementum is:**
 - Vascular
 - Avascular
 - Attached to pulp
 - None of the above
- Which of the following statements is/are true?**
 - Young cementum is lighter in color than dentin
 - Cementum is permeable
 - Cementum has the highest fluoride content of all the mineralized tissues
 - All of the above
- Epithelial rests of Malassez are found in the:**
 - Dentin
 - Periodontal ligament
 - Cementum
 - Enamel
- The uncalcified matrix of cementum is called:**
 - Cementoid
 - True denticles
 - False denticles
 - Free denticles
- Which of the following statements is/are true?**
 - Acellular cementum is often missing on the apical 3rd of the root
 - Cementum is thinnest at the cemento-enamel junction
 - Cementum is thickest at the apex
 - All of the above
- The incremental lines of cementum are:**
 - Poorly mineralized
 - Highly mineralized
 - Irregularly mineralized
 - None of the above
- In approximately 60% of teeth:**
 - Cementum overlaps the enamel at the cervical end for a short distance
 - Cementum and enamel do not meet
 - Cementum meets in a sharp line to enamel
 - A connective tissue attachment to tooth is possible without cementum
- Absence of cementum is found in:**
 - Hypophosphatasia
 - Hyperparathyroidism
 - Hyperthyroidism
 - Mongolism
- Which of the following statements is/are true?**
 - Cementum is not resorbed under normal conditions
 - Deposition of cementum in an apical area can compensate for loss of tooth substance by occlusal wear
 - Hypercementosis is abnormal thickening of cementum
 - All of the above
- Orthodontic tooth movement is made possible because:**
 - Cementum is more resistant to resorption than bone
 - Bone is more resistant to resorption than cementum
 - Bone is poorly vascularized than cementum
 - Cementum is poorly vascularized than bone
- Regarding functional repair of cementum which of the following statements is/are correct?**
 - There is tendency to reestablish the former outline of root surface
 - Only a thin layer of cementum is deposited on the surface of deep resorption and root outline is not reconstructed
 - The outline of alveolar bone in these cases follows that of root surface
 - Both (b) and (c)

13. **Compact bone and cellular cementum are similar as they contain:**
 A. Canaliculi and incremental lines
 B. Lacunae and elastic fibers
 C. Collagen fibers and blood vessels
 D. Sharpey's fibers and elastic fibers
14. **Sharpey's fibers:**
 A. Arise from Hertwig's sheath
 B. Arise from the epithelial rests of Malassez
 C. Arise from the epithelial diaphragm
 D. Are collagen fibers of the dental follicle embedded in the cementum
15. **Cementoid is:**
 A. The alveolar bone
 B. The central zone of fibers
 C. The uncalcified cementum
 D. The calcified matrix of cementum
16. **Cementum formation is:**
 A. Not a continuous process
 B. Always results in cellular cementum
 C. A continuous process
 D. Present in the dental follicle
17. **Cementum is formed from:**
 A. Endoderm B. The dental organ
 C. Ectoderm D. The dental sac
18. **Cellular cementum is thickest:**
 A. Around the root apex
 B. At cemento-enamel junction
 C. At middle one third of the root.
 D. At coronal one third of the root.
19. **Cementum is thinnest at:**
 A. Apical third of root
 B. Middle third of root
 C. Cemento-enamel junction
 D. Apical foramen
20. **Acellular cementum is thickest at:**
 A. Apical foramen
 B. Coronal one third of root
 C. Middle one third of root
 D. Apical one third of root
21. **Cementum overlaps the cervical end of enamel in a relatively sharp line in:**
 A. 30% of teeth B. 20% of teeth
 C. 10% of teeth D. None
22. **If functional qualities of cementum improve by its overgrowth it is called as:**
 A. Hypoplasia B. Hyperplasia
 C. Hypertrophy D. Any of the above
23. **In a nonfunctioning teeth there is:**
 A. Thickening of the cementum
 B. Thinning of the cementum
 C. No change in cementum
 D. None of the above
24. **Cementoblasts are of the following types:**
 A. Cementoblasts producing cellular cementum
 B. Cementoblasts producing acellular cementum
 C. Both the above
 D. None of the above.
25. **Cementoblasts are:**
 A. Cuboidal cells B. Round cells
 C. Tall columnar cells. D. Spider like cells.

ANSWERS

1. A 2. B 3. D 4. B 5. A 6. D
 7. B 8. A 9. A 10. D 11. A 12. D
 13. A 14. D 15. C 16. C 17. D 18. A
 19. C 20. B 21. D 22. C 23. A 24. C
 25. A

CHAPTER-8

PERIODONTAL LIGAMENT

1. **The periodontium comprises of how many connective tissues?**
 A. 2 B. 3
 C. 4 D. 5
2. **The functions of periodontal ligament is/are:**
 A. Support and nutrition
 B. Synthesis and resorption
 C. Proprioception
 D. All of the above
3. **The majority of the fibers of the periodontal ligament are:**
 A. Collagen
 B. Variety of micromolecules
 C. Mesenchymal
 A. All of the above

4. **The mandible consists of a series of bones united by sutures found in:**
 - A. Birds
 - B. Orthopodes
 - C. Reptiles
 - D. Mammals
5. **Which of the following statements is/are true?**
 - A. In the reptiles the teeth are ankylosed to the bone
 - B. In the mammals teeth are suspended in their socket by ligament
 - C. In the reptiles growth of the mandibular body in height occurs in mandibular sutures
 - D. All of the above
6. **Osteoclasts are rich in:**
 - A. Alkaline phosphatase
 - B. Acid phosphatase
 - C. Both of the above
 - D. None of the above
7. **The mast cells are characterized by numerous cytoplasmic granules. The granules have been shown to contain:**
 - A. Heparin
 - B. Histamine
 - C. Serotonin
 - D. All of the above
8. **Periodontal ligament appears to be made up of:**
 - A. Type I and Type II collagen
 - B. Type I and Type III collagen
 - C. Type II and Type III collagen
 - D. Only Type III collagen
9. **The fibers in human periodontal ligament are made up of:**
 - A. Collagen
 - B. Oxytalan
 - C. Both of the above
 - D. None of the above
10. **The fiber bundle that is most numerous and constitutes the main attachment of the tooth is:**
 - A. Alveolar crest group
 - B. Horizontal group
 - C. Oblique group
 - D. Apical group
11. **What do you know about intermediate plexus in periodontal ligament?**
 - A. It may appear as fibers arising from cementum and bone joined in the midregion of the periodontal space
 - B. It provides a site where rapid remodeling of fibers occurs
 - C. It is an artifact arising out of the plane of section and may move from one bundle to the other
 - D. All of the above
12. **A particular glycoprotein which occurs in filamentous form in the periodontal ligament is called:**
 - A. Fibronectin
 - B. Proline
 - C. Hydroxyproline
 - D. Chitin
13. **The blood supply of periodontal ligament is derived from:**
 - A. Branches from apical vessels that supply dental pulp
 - B. Branches from intra-alveolar vessels
 - C. Branches from gingival vessels
 - D. All of the above
14. **Which vitamin is essential for collagen synthesis?**
 - A. Vitamin A
 - B. Vitamin B
 - C. Vitamin C
 - D. Vitamin D
15. **Measurements of a larger number of periodontal ligament range from:**
 - A. 0.01 - 0.02 mm
 - B. 0.01 - 0.03 mm
 - C. 0.015 - 0.038 mm
 - D. 0.15 - 0.38 mm
16. **The thickness of the periodontal ligament is:**
 - A. Less in functionless and embedded teeth
 - B. More in teeth that are under excessive occlusal stresses
 - C. More in functionless teeth
 - D. (a) and (b) are correct
17. **The periodontal ligament is synthesized from:**
 - A. Cementum
 - B. Alveolar bone
 - C. Secondary cementum
 - D. Middle portion of the dental follicle
18. **The cementum, periodontal ligament and alveolar bone are derived from the:**
 - A. Dental follicle
 - B. Cementoid
 - C. Cementoblasts
 - D. Sharpey's fibers
19. **The dental follicle is a condensation of:**
 - A. Mesenchymal cells surrounding the enamel organ and dental papilla
 - B. Cells around the dental papilla
 - C. Cells around the enamel organ
 - D. Ectodermal cells surrounding the enamel organ and dental papilla
20. **The fibers forming the periodontal ligament are:**
 - A. Reticular
 - B. Collagen
 - C. Elastin
 - D. Keratin

21. The largest group of periodontal fibers are:
 A. The apical fibers
 B. The horizontal fibers
 C. The oblique fibers
 D. The transseptal fibers
22. The apical group of periodontal fibers originates from the:
 A. Cervical portion of the tooth
 B. Crest of the alveolar bone
 C. Ends of the roots
 D. Mid root region.
23. The nerves present in the periodontal ligament are:
 A. Nonmyelinated B. Myelinated
 C. None D. Both
24. The principal fibers of the periodontal ligament are attached to:
 A. Dentin and cementum
 B. Cementum and basal bone
 C. Alveolar bone proper and cementum
 D. Supporting alveolar bone and cementum
25. Which of the following is not a function of periodontal ligament?
 A. Supportive B. Nutritive
 C. Sensory D. Defensive
26. The periodontal ligament is thinnest in:
 A. Apical third of the root
 B. Middle region of the root
 C. Coronal third of the root
 D. Apical foramen
27. In a non- functioning tooth the periodontal ligament is:
 A. Thick B. Thin
 C. Either of the above D. None of the above
28. The periodontal ligament fibers, which hold the tooth in socket and oppose lateral forces are:
 A. Gingival fibers
 B. Alveolar crest fibers
 C. Apical fibers
 D. Oblique fibers
29. Width of the periodontal ligament is:
 A. 5 to 15 micron B. 25 to 50 micron
 C. 0.15 to 0.38 mm D. 0.60 to 0.85 mm
30. Function/s of the fibroblasts is/are:
 A. Formation and remodeling of periodontal ligament fibres
 B. To maintain the width of periodontal ligament
 C. Both the above
 D. None of the above
31. Mesenchymal stem cells in periodontal ligament perform the following functions:
 A. Tissue homeostasis
 B. Source of renewable progenitor cells
 C. Both of above
 D. None of the above
32. The average diameter of the arterioles in periodontal ligament is:
 A. 5 micron B. 10 micron
 C. 15 micron D. 20 micron
33. Mean average diameter of venous channels in periodontal ligament is:
 A. 18 micron B. 28 micron
 C. 38 micron D. 48 micron
34. Desmodont is another name for:
 A. The tooth with one wall pocket.
 B. Tooth with three walled pocket
 C. Periodontal ligament “
 D. Dehiscence

ANSWERS

1. C 2. D 3. A 4. C 5. D 6. B
 7. D 8. B 9. C 10. C 11. D 12. A
 13. D 14. C 15. D 16. D 17. D 18. A
 19. C 20. B 21. C 22. C 23. D 24. C
 25. D 26. B 27. B 28. B 29. C 30. C
 31. C 32. D 33. B 34. C

CHAPTER-9

ORAL MUCOUS MEMBRANE

1. The main function of saliva is to:
 A. Digest the food
 B. Kill the bacteria
 C. Clean the surfaces of teeth
 D. Lubricate the food and facilitate its swallowing

2. **Masticatory mucosa is the mucosa covering:**
 - A. Gingiva
 - B. Alveolar mucosa
 - C. Floor of mouth
 - D. Soft palate
3. **Which of the following is keratinized area?**
 - A. Vermilion border of lip
 - B. Alveolar mucosa
 - C. Floor of oral cavity
 - D. Inferior surface of tongue
4. **The connective tissue component of oral mucosa is termed the:**
 - A. Basal layer
 - B. Basement membrane
 - C. Lamina propria
 - D. Submucous layer
5. **A common feature of all epithelial cells is that they contain keratin. The analogous components of connective tissue cells are called:**
 - A. Desmin
 - B. Vimentin
 - C. Neural filaments
 - D. None of the above
6. **Lamina lucida contains:**
 - A. Laminin
 - B. Bullous pemphigoid antigen
 - C. Antigen bound by the antibody KF-1
 - D. Both (a) and (b)
7. **The percentage of cell membrane occupied by hemidesmosomes is highest in basal cells of:**
 - A. Gingiva and palate
 - B. Alveolar mucosa
 - C. Buccal mucosa
 - D. Tongue
8. **Out of the four layers which cells are the most active in protein synthesis?**
 - A. Basal cells
 - B. Spinous cells
 - C. Granular cells
 - D. Corneum cells
9. **Odland body is found in:**
 - A. Spinous cell layers
 - B. Granular cell layers
 - C. Both (a) and (b)
 - D. None of the above
10. **The lamina propria, a layer of dense connective tissue, is:**
 - A. Thicker in anterior than in posterior parts of palate
 - B. Thicker in posterior than in anterior part of palate
 - C. Not found in posterior part of palate
 - D. Not found in anterior part of palate
11. **Gingiva most often is:**
 - A. Nonkeratinized
 - B. Keratinized
 - C. Orthokeratinized
 - D. Parakeratinized
12. **Which of the following statements is/are correct?**
 - A. In the absence of sulcus there is no free gingiva
 - B. The disappearance of stippling is an indication of gingivitis
 - C. In younger females the connective tissue of gingiva is more finely textured than in the males
 - D. All of the above
13. **In a three-dimensional view, the interdental papilla of posterior teeth is tent shaped, whereas in anterior teeth its shape is:**
 - A. Pyramidal
 - B. Rectangular
 - C. Elliptical
 - D. Square
14. **The gingival 'col' is more vulnerable to periodontal disease because it is:**
 - A. Keratinized
 - B. Parakeratinized
 - C. Nonkeratinized
 - D. Orthokeratinized
15. **The accessory fibers that extend interproximally between adjacent teeth are known as:**
 - A. Transseptal fibers
 - B. Dentogingival fibers
 - C. Alveologingival fibers
 - D. Circular fibers
16. **The pigmentation is more abundant at the:**
 - A. Buccal gingiva
 - B. Labial gingiva
 - C. Base of the interdental papilla
 - D. Lingual gingiva
17. **The gingiva is:**
 - A. 75% parakeratinized
 - B. 15% keratinized
 - C. 10% nonkeratinized
 - D. All of the above
18. **The lymph supply of gingiva is:**
 - A. Submental
 - B. Submandibular
 - C. Jugulo-omohyoid
 - D. Only (a) and (b)
19. **Vermilion border, which is the transitional zone between the skin of the lip and the mucous membrane of the lip, is also known as:**
 - A. White zone
 - B. Red zone
 - C. Reducing zone
 - D. Violet zone
20. **The sebaceous glands lateral to the corner of the mouth and often seen opposite the molars are called:**
 - A. Fordyce's spot
 - B. Hutchinson spot
 - C. Intestinal polyposis
 - D. Miller's spot

21. Which papillae of tongue are keratinized and do not contain taste buds?
- Fungiform papillae
 - Filiform papillae
 - Vallate papillae
 - (b) and (c)
22. Which of the following statements is/are true?
- Sweet taste is perceived at the tip of the tongue
 - Salty taste is perceived at the lateral border of the tongue
 - Bitter and sour taste are perceived on the palate and in the posterior part of the tongue
 - All of the above
23. Which statement about oral mucosa in general is not correct?
- Oral mucosa is defined as moist lining of the oral cavity that is in continuation with the exterior surface of skin.
 - Oral mucosa is situated anatomically between the skin and intestinal mucosa. Hence, it shows some properties of both
 - Oral mucosa shows regional structural modifications according to the stress and workload borne by it.
 - The epithelium of the oral mucosa is always orthokeratinized
24. The gingival sulcus is formed by the:
- Tooth surface and epithelial covering of attached gingiva
 - Tooth surface and epithelial covering of free gingiva
 - Free gingival groove and mucogingival junction
 - Epithelial covering of the free and attached gingiva
25. The reduced enamel epithelium:
- Is about 40 microns thick
 - Produces the primary enamel cuticle
 - Does not protect the enamel until tooth eruption
 - Produces the primary attachment epithelium
26. After tooth eruption, the reduced enamel epithelium:
- Causes shrinkage of the stratum reticulum
 - Promotes the differentiation of dentin
 - Forms the epithelial attachment
 - Forms the secondary enamel cuticle
27. The last organic material secreted by the ameloblast is:
- Primary enamel cuticle
 - Secondary cuticle
 - Enamel tufts
 - Tomes' process
28. The oral epithelium is attached to the enamel via:
- Reticular fibers
 - Collagen fibers
 - Hemidesmosomes
 - Elastic fibers
29. Oral Epithelium is:
- Nervous tissue
 - Muscle tissue
 - Connective tissue
 - Avascular tissue
30. The lamina propria of the oral mucous membrane contains:
- Ectoderm
 - Bone
 - Keratin
 - Blood vessels
31. Majority of taste buds are found on the:
- Filiform papillae
 - Fungiform papillae
 - Circumvallate papillae and the adjacent trench wall.
 - All of the above
32. Glands of von Ebner empty their contents into:
- Fungiform papillae
 - Circumvallate trench
 - Filiform papilla
 - None of the above
33. The gingival sulcus is bounded by:
- Free gingival groove and the junction of mucosa and gingiva
 - Surface of the tooth and the attached gingiva
 - Epithelial covering of the free gingiva and the tooth surface
 - Epithelial covering of the free gingiva and the attached gingiva
34. The specialised mucosa is present on:
- Lips and cheeks
 - Gingiva and hard palate
 - Dorsum of tongue and taste buds
 - Floor of mouth and soft palate
35. The connective tissue component of the oral mucosa is known as:
- Submucosa
 - Basal lamina
 - Lamina propria
 - Dermis

36. **The basement membrane of oral mucosa:**
 A. Can be seen by light microscope
 B. Is present at the interface of epithelium and connective tissue
 C. Has a width of 1-4 nm
 D. All of the above
37. **The epithelium of the oral mucous membrane is:**
 A. Stratified columnar
 B. Simple squamous epithelium
 C. Stratified squamous
 D. Non-stratified squamous
38. **The epithelium present in gingiva and hard palate is:**
 A. Parakeratinized
 B. Nonkeratinized
 C. Keratinized
 D. Any of the above
39. **The epithelium of cheek and sublingual tissue is:**
 A. Nonkeratinized B. Parakeratinized
 C. Keratinized D. None of the above
40. **Of the four layers, the cells most active in protein synthesis are of:**
 A. Stratum corneum
 B. Stratum granulosum
 C. Stratum spinosum
 D. Stratum basale
41. **The mucous membrane of the soft palate is:**
 A. Parakeratinized
 B. Nonkeratinized
 C. Keratinized
 D. Any of the above
42. **Masticatory mucosa is not present on:**
 A. Palatal fauces
 B. Attached gingiva
 C. Dorsum of the tongue
 D. Floor of the mouth
43. **Keratinosomes (Odland body) help in:**
 A. Exchange of fluids
 B. Nutrition
 C. Sensory perception
 D. Formation of intercellular agglutinating material
44. **Jacobson's organ is:**
 A. Also known as vomeronasal organ
 B. Ellipsoidal structure lined with olfactory epithelium
 C. Considered as auxiliary olfactory sense organ
 D. All the above are correct
45. **The disappearance of the stippling from the gingiva indicates:**
 A. Trauma
 B. Old age
 C. Progressive gingivitis
 D. Is a normal feature
46. **The most numerous group of gingival fibers is:**
 A. Dentoperiosteal B. Dentogingival
 C. Alveologingival D. Circular
47. **The interdental ligament is formed by:**
 A. Dento-gingival fibers
 B. Dentoperiosteal fibers
 C. Transseptal fibers
 D. Alveologingival fibers
48. **A common feature of melanocytes, Langerhans cells and Merkel cell is that they all:**
 A. Produce melanin
 B. Have a low or no desmosomal attachment with surrounding keratinocytes
 C. Are dendritic
 D. Are pressure sensitive cells
49. **The Langerhans cells are:**
 A. Found in upper layers of skin and the mucosal epithelium
 B. Of hematopoietic origin
 C. Involved in the immune response
 D. All of the above
50. **Merkel cells help in:**
 A. Nutritive function
 B. Sensory function
 C. Neurosensory activities
 D. Olfactory function
51. **Number of the circumvallate papilla ranges from:**
 A. 20 to 25 B. 15 to 20
 C. 8 to 10 D. 4 to 5
52. **The papilla responsible to recognize sour taste is:**
 A. Vallate papilla B. Foliate papilla
 C. Fungiform papilla D. Filiform papilla
53. **Which of the followings is located at the angle of the V-shaped terminal groove:**
 A. Foramen ovale B. Foramen cecum
 C. Foramen magnum D. None of the above
54. **Bitter and Sour taste sensations are mediated by:**
 A. Chorda tympani
 B. Hypoglossal nerve
 C. Glossopharyngeal nerve
 D. Intermediocardial nerve

55. In oral mucous membrane the thickness of lamina lucida is:
 A. 5 to 10 nm B. 20 to 40 nm
 C. 50 to 70 nm D. 70 to 90 nm
56. Merkel cells are commonly observed in:
 A. Masticatory mucosa
 B. Lining mucosa
 C. Both the above
 D. None of the above
57. Anticancer drugs affect oral mucosa and cause:
 A. Discoloration B. Thickening
 C. Ulcers D. Blisters
58. In which part of oral cavity mucous membrane is the thinnest?
 A. Soft palate B. Labial mucosa
 C. Floor of mouth D. Buccal mucosa
3. The mandible makes its appearance as a bilateral structure in the 6th week of fetal life as thin plate of bone:
 A. Mesial to Meckel's cartilage
 B. Lateral to Meckel's cartilage
 C. Mesial and lateral to Meckel's cartilage
 D. Mesial and some distance from Meckel's cartilage
4. Spongy bone is least found in region of:
 A. Anterior teeth
 B. Premolar teeth
 C. Molar teeth
 D. Upper posterior teeth only
5. The interdental and interradicular septa contain the perforating canals of:
 A. Alock's
 B. Zuckerk and Hirschfeld
 C. Dorello's
 D. Gartner's

ANSWERS

1. D 2. A 3. A 4. C 5. B 6. D
 7. A 8. B 9. C 10. A 11. D 12. D
 13. A 14. C 15. A 16. C 17. D 18. D
 19. B 20. A 21. B 22. D 23. D 24. B
 25. D 26. C 27. A 28. C 29. D 30. D
 31. C 32. B 33. C 34. C 35. C 36. D
 37. C 38. C 39. A 40. C 41. B 42. C
 43. D 44. D 45. C 46. B 47. C 48. B
 49. D 50. C 51. C 52. B 53. B 54. C
 55. B 56. A 57. C 58. C

CHAPTER-10

BONE AND ALVEOLUS

1. In the beginning of the second month of fetal life the skull consists of the:
 A. Chondrocranium
 B. Desmocranium
 C. Appendicular skeleton
 D. All of the above
2. Which bone develops in desmocranium?
 A. Frontal bone
 B. Parietal bone
 C. Greater wing of sphenoid bone
 D. All of the above
3. The mandible makes its appearance as a bilateral structure in the 6th week of fetal life as thin plate of bone:
 A. Mesial to Meckel's cartilage
 B. Lateral to Meckel's cartilage
 C. Mesial and lateral to Meckel's cartilage
 D. Mesial and some distance from Meckel's cartilage
4. Spongy bone is least found in region of:
 A. Anterior teeth
 B. Premolar teeth
 C. Molar teeth
 D. Upper posterior teeth only
5. The interdental and interradicular septa contain the perforating canals of:
 A. Alock's
 B. Zuckerk and Hirschfeld
 C. Dorello's
 D. Gartner's
6. Which enzyme participates in the deposition of hydroxyapatite crystals in bone?
 A. Alkaline phosphatase
 B. Adenosine triphosphatase
 C. Pyrophosphatases
 D. All of the above
7. Osteoclasts are multinucleated giant cells formed by monocytes and are found in bay-like depression in the bone called:
 A. Haversian canal
 B. Volkman canal
 C. Howship's lacunae
 D. HIRSCHFELD canal
8. During orthodontic tooth movement on the pressure side there is an increase in the level of:
 A. Odontoblast
 B. Cementoblast
 C. Alkaline phosphatase
 D. Cyclic adenosine monophosphate
9. The body of the mandible is formed by the:
 A. Conversion of cartilage directly into bone
 B. Ordinary endochondral ossification similar to that of most long bones
 C. Intramembranous ossification similar to that of the bones of the cranial vault
 D. None of the above

10. **The upper jaw:**
 A. Is formed by fusion of the maxilla and premaxilla
 B. Develops from the otic capsule
 C. Is formed from Meckel's cartilage
 D. Develops from the nasolacrimal groove
11. **The mandible ossifies:**
 A. From one center in the midline of the mandible
 B. From two centers located medially to Meckel's cartilage
 C. As an endomembranous bone
 D. None of the above
12. **Which of the following is vascularized?**
 A. Cementum B. Enamel
 C. Bone D. Calculus
13. **The area of alveolar bone where Sharpey's fibers are embedded is called:**
 A. Lamellar bone
 B. Bundle bone
 C. Intramembranous bone
 D. Haversian bone
14. **Mark the correct statements about bundle bone. It:**
 A. Lines the socket in the teeth that are subjected to stress
 B. Anchors the Sharpey's fibers of the periodontal ligament
 C. Contains fewer matrix collagen fibrils than typical bone
 D. All of the above are correct
15. **The alveolar bone:**
 A. Consists of a compact layer and a cancellous layer
 B. Can be demarcated from the bone of the jaw
 C. Is known as periodontal plate
 D. Increases in size to compensate for loss of permanent teeth.
16. **The bone consists of:**
 A. 65% organic and 35% inorganic material
 B. 65% inorganic and 35% organic
 C. 70% organic and 30% inorganic
 D. 50% organic and 50% inorganic part
17. **The cells present inside the Howships lacunae are:**
 A. Osteocytes B. Osteoclasts
 C. Odontoblasts D. Osteoblasts

ANSWERS

1. D 2. D 3. C 4. A 5. B 6. D
 7. C 8. D 9. D 10. A 11. B 12. C
 13. B 14. D 15. A 16. A 17. B

CHAPTER-11 SALIVARY GLANDS

1. **The salivary glands are:**
 A. Exocrine B. Endocrine
 C. Holocrine D. None of the above
2. **The "basket cells" are also known as:**
 A. Myoepithelial cells
 B. Endothelial cells
 C. Parenchymal cells
 D. None of the above
3. **Myoepithelial cells are abundant in:**
 A. Sweat glands B. Mammary glands
 C. Both (a) and (b) D. None of the above
4. **Which of the following statements is/are true?**
 A. Parotid gland is purely serous
 B. Submandibular gland is mixed but predominantly serous in nature
 C. Sublingual gland is mixed but predominantly mucous in nature
 D. All of the above
5. **The three bilaterally paired major salivary glands are located:**
 A. Extraorally B. Intraorally
 C. In tongue D. In neck
6. **The parotid glands open through the:**
 A. Stensen's duct B. Wharton's duct
 C. Bartholin's duct D. Blanden duct
7. **The parotid gland duct opens into the oral cavity at the position:**
 A. On the floor of the mouth
 B. At the side of the lingual frenum
 C. At the caruncula
 D. On the buccal mucosa opposite the maxillary 2nd molar

8. Which statements is/are true?
 A. The parotid gland is purely serous
 B. In infants, few mucous secretory units may be found in parotid gland
 C. Both (a) and (b)
 D. None of the above
9. Which gland(s) is/are pure serous in nature?
 A. Parotid and von Ebner's glands
 B. Palatine glands only
 C. Glossopalatine glands
 D. Lingual glands
10. Which gland(s) is/are purely mucous?
 A. Palatine glands
 B. Glossopalatine glands
 C. Posterior lingual mucous glands
 D. All of the above
11. The primordia of the parotid and submandibular glands appear during 6th week of fetal life, whereas the primordium of sublingual glands appears after:
 A. 3-4th weeks of fetal life
 B. 7-8th weeks of fetal life
 C. 9-10th weeks of fetal life
 D. 10-12 weeks of fetal life
12. The minor salivary glands begin their development in fetal life during:
 A. 1st month B. 2nd month
 C. 3rd month D. 4th month
13. The total volume of saliva-secreted daily is approximately?
 A. 750 ml B. 1.5 liter
 C. 2 liter D. 3 liter
14. The largest amount of saliva is produced by:
 A. Submandibular gland
 B. Sublingual gland
 C. Parotid gland
 D. Lingual glands
15. The pH of whole saliva is:
 A. 1.2 - 2.4 B. 3.0 - 5.6
 C. 6.7 - 7.4 D. 7.0 - 8.2
16. The predominant salivary immunoglobulin is:
 A. IgA B. IgG
 C. IgE D. IgM
17. Human parotid gland produces a hormone, which is known as:
 A. Menotropins B. Parotin
 C. Serotonin D. Prohormone
18. Salivary glands are not found in:
 A. Anterior part of hard palate
 B. Posterior part of hard palate
 C. In mandible posterior to 3rd molar teeth
 D. Nasopalatine canal
19. The severance of the duct of minor salivary gland and pooling of saliva in the tissues is called as:
 A. Ranula B. Mucocele
 C. Congenital epulis D. Sialadenitis
20. What is the use of Sialochemistry?
 A. Determination of the quantity and composition of the saliva
 B. Determination of ovulation time
 C. Both (a) and (b)
 D. None of the above
21. Which of the following statement is/are true about the connective tissue elements of the salivary gland?
 A. The connective tissue forms a distinct capsule around major glands but not around minor glands.
 B. The connective tissue elements carry the vascular and nerve supply of the gland.
 C. From the capsule connective tissue septa penetrate the gland subdividing it into lobules.
 D. All of the above are correct
22. 'Bartholin's duct' is a secretory duct of:
 A. Parotid gland
 B. Sublingual gland
 C. Submandibular gland
 D. Palatine glands
23. The antibacterial proteins present in saliva is/ are:
 A. Lysozymes B. Lactoferrins
 C. Both of the above D. None of the above
24. The major glands may become enlarged during:
 A. Protein deficiency B. Alcoholism
 C. Pregnancy D. All of the above
25. Salivary flow is reduced in:
 A. Xerostomia
 B. Sjögren's syndrome
 C. Inflammation of glands
 D. All of the above
26. Secretory granules of salivary glands are:
 A. One mm in diameter
 B. Five mm in diameter
 C. Ten mm in diameter
 D. Fifteen mm in diameter

27. Submandibular salivary gland secrete:

- A. 10 percent of whole saliva.
- B. 30 percent of whole saliva.
- C. 60 percent of whole saliva
- D. 90 percent of whole saliva.

28. Which is the predominant factor in the formation of the alveolar process?

- A. Eruption of teeth
- B. Normal process of growth
- C. Lengthening of the condyle.
- D. Overall growth of the bodies of the maxilla and the mandible.

ANSWERS

1. A 2. A 3. C 4. D 5. A 6. A
 7. D 8. C 9. A 10. D 11. B 12. C
 13. A 14. A 15. C 16. A 17. B 18. A
 19. B 20. C 21. D 22. B 23. C 24. D
 25. D 26. A 27. C 28. A

CHAPTER-12

ERUPTION OF TEETH AND PHYSIOLOGIC TEETH MOVEMENTS

1. The permanent incisors and canine first develop:
 - A. Labial to deciduous tooth germs
 - B. Lingual to the deciduous tooth germs
 - C. Mesial to deciduous tooth germs
 - D. Distal to deciduous tooth germs
2. The upper permanent molars develop in tuberosity of the maxilla and in the beginning their occlusal surface faces:
 - A. Labially B. Lingually
 - C. Mesially D. Distally
3. At first, the occlusal surface of the permanent mandibular molars faces:
 - A. Mesially B. Distally
 - C. Lingually D. Labially
4. Successional teeth possess an additional anatomic feature, the gubernacular canal and its contents, the gubernacular cord. The function of gubernacular cord is:
 - A. Guiding the permanent tooth as it erupts
 - B. Blood supply to tooth
 - C. Blood supply to bone
 - D. Nutrition to tooth
5. Removal of the root:
 - A. Prevents eruption of tooth
 - B. Does not prevent eruption
 - C. Causes regeneration of root
 - D. None of the above
6. Which ligament has a key role in maintaining tooth position?
 - A. Horizontal group ligament
 - B. Apical group ligament
 - C. Inter-radicular group ligament
 - D. Transseptal ligament
7. Clinically as the teeth break through the oral mucosa, there is often some pain, slight fever, and general malaise, all signs of an inflammatory process. In infants, these symptoms are popularly called:
 - A. Teething
 - B. Biting
 - C. Erythroblastosis fetalis
 - D. Erythema multiforme
8. Resorption of the roots of the deciduous incisors and canines begins on their:
 - A. Mesial surface B. Distal surface
 - C. Labial surface D. Lingual surface
9. The most likely cause of tooth eruption is:
 - A. The growing root
 - B. Vascular pressure
 - C. The developing periodontal ligament
 - D. Bone growth
10. The transseptal ligament connects:
 - A. Cementum to bone
 - B. Bone to bone
 - C. Gingiva to cementum
 - D. Cementum of one tooth to the cementum of adjacent tooth
11. The actual eruptive movements occur mainly:
 - A. In a horizontal direction
 - B. In a rotational direction
 - C. In an axial direction
 - D. In multiple directions

12. Which one of the following events does not take place during the active phase of eruption?
- Bone deposition and resorption on the crypt wall.
 - Root formation
 - Organization of a periodontal ligament from the dental follicle.
 - Gradual separation of the attachment epithelium from the enamel surface.
13. Prior to actual eruption in the jaw bone, the tooth:
- Rotates
 - Moves in an apical direction
 - Moves in a horizontal direction
 - Moves in multiple directions
14. In the teeth with deciduous predecessors which canal is present that has an influence on eruptive tooth movement:
- Accessory canal
 - Gubernacular canal
 - Incisive canal
 - Nasolacrimal canal
15. Eruption of teeth:
- Is localized genetically controlled
 - Is programmed event
 - Both the above
 - None of the above.
16. When the tooth is in the bony crypt the eruption rate is:
- One micron per day
 - Five micron per day
 - Ten micron per day
 - Fifteen micron per day.
17. Final position of the tooth in the oral cavity is determined by the following factors:
- Pressure exerted by the tongue and cheeks
 - Pressure exerted by the adjacent teeth
 - Both of above
 - None of the above.

ANSWERS

1. B 2. D 3. A 4. A 5. B 6. D
 7. A 8. D 9. C 10. D 11. C 12. D
 13. D 14. B 15. C 16. A 17. C

CHAPTER-13

SHEDDING OF DECIDUOUS TEETH

- Resorption of the roots of the deciduous incisors and canines begins on their:**
 - Mesial surface
 - Distal surface
 - Labial surface
 - Lingual surface
- Resorption of the roots of deciduous molars often first begins on their:**
 - Outer surface
 - Inner surface
 - Mesial surface
 - Distal surface
- A characteristic feature of the odontoclast is high level of activity of the enzyme which is known as:**
 - Alkaline phosphatase
 - Acid phosphatase
 - Pyrophosphatase
 - Hyaluronidase
- When a successional tooth germ is missing, shedding of deciduous tooth is:**
 - Premature
 - Normal
 - Delayed
 - Never
- Sometimes part of the roots of deciduous teeth are not in the path of erupting permanent teeth and may escape resorption. They are most frequently found in association with the permanent:**
 - Incisors
 - Canine
 - Premolars
 - Molars
- Retained deciduous teeth are most often the upper:**
 - Central incisor
 - Lateral incisor
 - Canine
 - Molar
- For the removal of the dental hard tissues the cells responsible are:**
 - Odontoclast
 - Osteoclasts
 - Osteocytes
 - Chondroblast
- A high level of enzyme acid phosphatase is a characteristic feature of:**
 - Mast cells
 - Osteoclast
 - Osteocytes
 - Chondrocyte
- The result of premature loss of deciduous teeth is:**
 - Delayed eruption of successor
 - Earlier eruption of successor
 - No effect on eruption
 - Eruption does not occur

10. Pathologic root resorption may be caused by the pressure exerted by:

- A. Tumors B. Cysts
C. Both D. None of the above

11. Relative to primary mandibular incisors, permanent mandibular incisors erupt:

- A. Lingually B. Facially
C. Distally D. Mesially

ANSWERS

1. D 2. B 3. B 4. C 5. C 6. B
7. A 8. B 9. C 10. C 11. A

CHAPTER-14

TEMPOROMANDIBULAR JOINT

1. The condylar cartilage is:

- A. Both primary and secondary cartilages
B. A primary cartilage present prior to ossification of the mandible
C. A secondary cartilage not present prior to ossification of the mandible
D. A secondary cartilage present prior to ossification of the mandible

2. The amount of synovial fluid:

- A. Increases with age
B. Decreases with age
C. No affect with age
D. Decreases in winters.

ANSWERS

1. C 2. B

CHAPTER-15

MAXILLARY SINUS

1. The maxillary sinus communicates with the environment by:

- A. Superior nasal meatus
B. Middle nasal meatus
C. Middle nasal meatus and the nasal vestibule
D. Inferior nasal meatus

2. Maxillary sinus epithelium is:

- A. Stratified and columnar
B. Pseudostratified columnar and ciliated
C. Squamous and nonciliated
D. Glandular

3. The secretory cells present in the sub-epithelial glands of maxillary sinus are:

- A. Mucous cells B. Serous cells
C. Both of the above D. None of the above

4. All are the functions of the maxillary sinus except:

- A. Protect the internal structures against exposure to cold air
B. Contribute resonance to voice
C. Production of bactericidal lysozyme
D. Helps in mastication

ANSWERS

1. C 2. B 3. C 4. D

CHAPTER-18

ADVANCED TECHNIQUES IN THE STUDY OF ORAL TISSUES

1. Which statement(s) is/are true?

- A. Hyaluronic acid predominates in the loose connective tissues
B. Hyaluronic acid has high capacity to bind water and is responsible for transport and diffusion of metabolic substances across tissue
C. Bacterial infections may occur as a result of the hydrolytic action of the bacterial enzyme hyaluronidase
D. All of the above

2. The organic components of bone are mainly:

- A. 93% type I collagen
B. Hydroxyapatite
C. 90% type II collagen
D. 90% type IV collagen

3. Which is considered to be one of the most ideal fixatives?

- A. Sodium hypochlorite
B. Formaldehyde
C. Acetaldehyde
D. H₂SO₄

4. Formaldehyde as a fixative is generally used as a:
 A. 10 % solution B. 20 % solution
 C. 30 % solution D. 40 % solution
5. Rossman's fluid contains:
 A. Formaldehyde
 B. Alcohol
 C. Picric acid and acetic acid
 D. All of the above
6. Carnoy's mixture, is composed of:
 A. Ethyl alcohol B. Acetic acid
 C. Chloroform D. All of the above
7. Feulgen's reaction is used for visualizing:
 A. DNA B. RNA
 C. Chromosome D. Microsome
8. The best known and frequently used technique for detection of carbohydrate grouping is:
 A. Versene technique B. PAS technique
 C. Carnoy's mixture D. All of the above
9. Which is nonreactive with PAS method?
 A. Developing bone B. Resorbing bone
 C. Resorbing dentin D. Enamel matrix
10. Which type of collagen is absent in normal adult dentin?
 A. Type I B. Type II
 C. Type III D. All of the above
11. The localization of type III collagen in dentin is found in:
 A. Dentinogenesis imperfecta type II
 B. Osteogenesis imperfecta
 C. None of the above
 D. Both (a) and (b)
12. In the developing molar and incisor teeth, alkaline phosphatase is present in the:
 A. Outer enamel epithelium
 B. Inner enamel epithelium
 C. Stellate reticulum
 D. Stratum intermedium
13. The lack of mast cells is found in:
 A. Tongue B. Gingiva
 C. ANUG D. All of the above
14. Elevation of which enzyme is considered to assist in the diagnosis of cancer?
 A. Amylase
 B. Glucose 6-phosphate dehydrogenase
 C. Aminopeptidase
 D. Cytochrome oxidase
15. Which of the techniques is/ are used for quantitative analysis of histochemical reactions:
 A. Microphotocell counter
 B. Double beam recording
 C. Both of the above
 D. None of the above
16. Scanning electron microscope:
 A. Is used in analysis of changes in the bone architecture
 B. Can produce three dimensional image
 C. Can elucidate the chemical substances uptake by metabolic pathways of different tissues in different regions of cytoplasm.
 D. Can be used for none of the above.
17. Rossman's fluid is used for visualization of:
 A. Glycogen B. Glycoproteins
 C. Proteoglycans D. All of the above
18. β -Glucuronides is important for:
 A. Cell proliferation
 B. Conjugation of steroid hormone
 C. In hydrolysis of conjugated glucuronides
 D. All of the above
19. Post fixation is a secondary fixation done on:
 A. Lipid rich tissues
 B. Freeze dried tissues
 C. Both the above
 D. None of the above

ANSWERS

1. D 2. A 3. B 4. A 5. D 6. D
 7. A 8. B 9. D 10. C 11. D 12. D
 13. C 14. B 15. C 16. A 17. D 18. D
 19. C