

Degree Course in Dentistry and Dental Prosthetics 2021/2022

Integrated Course: Histology

CFU Number: 8

SSD Course: BIO/17

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PREREQUISITES

Although there are no propaedeutic requirements, basic concepts of cell biology are required.

LEARNING OBJECTIVES

Aim of the integrated course of Histology is to provide students with knowledge on the physiological organization and development of cells and tissues. During the Histology lectures fundamentals of cytology are discussed for the full understanding of the organization of tissues and their development. The organization of cells and extracellular matrix and their association in the organization of the different tissues is illustrated and discussed, together with the standard histological procedures, including microscopy approaches (optics, fluorescence and electronics), as a tool for the study of structure and development of the human organism.

LEARNING OUTCOMES

Knowledge and understanding

At the end of the course of Histology the student should have acquired:

- Knowledge on the structure of the various tissues forming the human organism
- Knowledge on the histological organization of the various human organs
- Ability to recognize the morphology of tissues and the cells that compose them, from a morphological and functional point of view
- Ability to recognize and describe the microscopic organization of tissues and organs, and the processes leading to their formation during development
- Ability to synthesize and correlate the various topics.

Applying knowledge and understanding

At the end of the course the student should be able to:

- Recognize and contextualize microscopic information.
- Understand the relationship between cells and the extracellular matrix underlying the organization of the different tissues of the multicellular organism.

- Apply the knowledge of histology and embryology to the subsequent study of physiology, anatomy, pathological anatomy and clinical disciplines.

Communication skills

At the end of the course the student should be able to use a correct scientific terminology to identify, at the microscopic level, the different types of cells and tissues present in the human body and the mechanisms of their formation during embryonic development.

Making judgements

At the end of the course the student should autonomously interpret histological slides and describe the processes of embryogenesis.

Learning skills

At the end of the course, the student should have acquired independent method for studying and updating through different kind of literature or through scientific literature.

COURSE SYLLABUS

Citology

Methods to study cells and tissues. Notes on the use of optical (bright field, phase contrast, interference, fluorescence, confocal) and electronic (TEM and SEM) microscopes; unit of measurement and resolving power (Abbe's formula). Procedures for preparing of specimens for optical (paraffin and freezing) and electron microscopy. Cell cultures. Autoradiography and electrophoresis. Main histomorphological and histochemical staining procedure. Principles of immunolocalization of cellular molecules and organelles. The fractional centrifugation.

The plasma membrane. Molecular structure and organization of the plasma membrane: the fluid mosaic model; membrane lipids and their properties; extrinsic and intrinsic proteins: antigenic properties, function as adhesive proteins, function as receptors and their role in signal transduction. Transport of small molecules and water across the plasma membrane: simple diffusion, facilitated diffusion, active transport and osmosis. Principal morphological ((freeze-fracture) and molecular (immunolocalization and electrophoresis of proteins) study methods. Composition and functions of the glycocalyx.

The cytoplasmic organelles. Composition of the cytosol and of the various cytoplasmic inclusions (glycogen granules and lipid droplets). The smooth endoplasmic reticulum: structure, role in lipid metabolism, detoxification processes, in glycogenolysis and in the accumulation of calcium. Ultrastructural organization and function of the granular endoplasmic reticulum. Main steps in the translation process and differences between the synthesis of proteins destined for the cytosol and that of secretory, membrane or lysosomal proteins. Post-translational modifications of proteins: glycosylation, hydroxylation and role of molecular chaperones. COP protein-coated transport vesicles. Specificity of vesicular transport and fusion processes: v-SNARE and t-SNARE proteins. Golgi complex: ultrastructure, biosynthetic processes and sorting of the molecules synthesized in the endoplasmic reticulum. Constitutive and regulated cell secretion: regulatory mechanisms. Endocytosis. Internalization of soluble molecules by caveole: pinocytosis, transcytosis, interactions of caveolins with signal molecules. Receptor Mediated Endocytosis: Clathrin-coated vesicles. Endosomes and the different sorting pathways of specific ligands. Lysosomes: biogenesis,

morphology, hydrolytic enzymes. Phagocytosis and autophagy. The peroxisomes: structure and functions

Mechanisms for the degradation of cytoplasmic proteins: the ubiquitin-proteasome system and the aggresome.

Mitochondria: morphology, distribution and replication. Mitochondrial genome. Localization and function of mitochondrial enzymatic complexes: main aspects of the Krebs cycle and oxidative phosphorylation. Role of mitochondria in calcium homeostasis, in apoptosis and in the synthesis of steroid hormones.

The cytoskeleton. Microtubules, microfilaments and intermediate filaments: molecular organization, distribution in the cell and in the different cell types. The function of the cytoskeleton in specific processes such as cell motility, phagocytosis, endocytosis, exocytosis, vesicle movement. Proteins associated with microtubules (kinesins and dyneins) and microfilaments (proteins that bind actin).

The centrosome. The membrane cytoskeleton. Vibrating lashes: structure and function. The primary edge.

The nucleus. Structure of the interphase nucleus. Exchanges between nucleus and cytoplasm. Composition of interphase chromatin and nuclear matrix. Euchromatin and heterochromatin. The nucleosomes. Histones, non-histone regulatory proteins. The nucleolus: molecular structure and organization; the synthesis of rRNA and the assembly of ribosomal subunits. The nuclear envelope: differences between the cytoplasmic and nuclear surfaces, structure and function of nuclear pores, importins and exports, associated regulatory molecules, the nuclear lamina.

Notes on cell division and the phases of the cell cycle. Notes on the formation of chromosomes (their structure) and the mitotic apparatus. Notes on the stages of mitosis.

Cellular interactions General principles of paracrine, autocrine, endocrine and juxtacrine cellular interactions. Cell-cell and cell-matrix adhesive systems. The intercellular junction structures: structural and molecular organization of the occluding junctions, of the anchoring junctions (adherent zonules and desmosomes) and of the communicating junctions (gap junctions). The integrins. Podosomes and focal adhesions. Hemidesmosomes.

Histology

Methods for the morphological study of tissues

Optical microscopes (transmitted light, phase contrast, interferential, fluorescence and confocal optical microscopes) and electronic microscopes (TEM and SEM), basic principles (resolving power and magnification) and use; preparation of a histological preparation for the optical (paraffin preparation) and electronic microscope (ultra-fine sections; freezing / fracture); the main stains for optical and electronic microscopy, histochemistry and immunohistochemistry.

THE EPITHELIAL TISSUES

Generalities (organization, vascularization and innervation) and embryonic derivation. Characteristics of epithelial cells [shape, polarity, specializations of the apical surface (common cilia and primary cilia (notes on eyelashes), microvilli, stereocilia), specializations of the lateral surface [adhesive molecules and junction complexes (ultrastructural and molecular organization of the occluding junction, adherent, desmosome), the gap junctions], specializations of the basal surface (structural and molecular organization, function of the basal lamina), the cytoskeleton, epithelial stem cells (general characteristics of stem cells of adult tissues).

Classification of epithelial tissues (lining and glandular; notes on sensory and special epithelia).

Surface epithelia. General characteristics, classification, distribution and hints of functions. Mucous membranes (intestinal epithelium, airway epithelium, bladder epithelium), serosa, skin and endothelium [notes on the structure of capillaries (continuous, fenestrated and sinusoid capillaries, passage of molecules and cells through the endothelium) and of blood vessels]. Epidermis (keratinocytes and corneification, melanocytes and melanogenesis, Langerhans cells and Merkel cells).

Glandular epithelia. Embryonic derivation and histological organization of the exocrine and endocrine glands (parenchyma and stroma). Exocrine glands (position, organization, types of adenomeres and excretory ducts), classification and secretions (oxyntic cells and Paneth cells). Myoepithelial cells. Examples of exocrine glands: skin glands (sebaceous, sweat and mammary glands), major salivary glands, pancreas. Endocrine glands. General characteristics (concept of cellular signaling, autocrine, paracrine, endocrine and juxtacrine, exosomes) and histological organization (cordonal, follicular and interstitial). Hormones (polypeptides, amino acids and steroids). Examples of histological organization and functions of endocrine glands: pituitary, thyroid, adrenal, pancreas.

THE CONNECTIVE TISSUES

General characteristics, embryonic origin, classification, distribution and functions.

The connective tissues proper (classification, distribution and functions): loose (areolar) and dense (regular and irregular). Mesenchymal stem cells. Resident cells (fibroblasts, reticular cells, macrophages [phagocytosis (opsonins, Toll-type receptors, complement, such as APC cells, the macrophage system), mast cells, adipocytes]. Integrins and interactions with molecules of the intercellular substance. Intercellular substance (amorphous matrix and protein fibers). Amorphous matrix (GAGs, proteoglycans and glycoproteins) and interstitial fluid. Protein fibers. Collagens (classification: fibrillar, laminar / reticular and FACIT; and their molecular organization in particular of collagen I, synthesis and fibrillogenesis). Elastin and elastic fibers (molecular characteristics of elastin, fibrillins and their synthesis). Notes on some pathologies due to proteoglycans, collagenopathies and elastinopathies. Connective tissues proper with special properties [adipose (white and brown), reticular, elastic, pigmented, mucous].

The supporting connective tissues. Cartilage tissues.

Hyaline cartilage (embryonic origin, histological organization, distribution and growth: characteristics of coloring, perichondrium, chondroblasts / chondrocytes, intercellular substance and aggrecans, collagen fibers). Special types of hyaline cartilage (metaphyseal / seriata / conjugation and articular cartilage). Elastic cartilage (embryonic origin, histological organization, distribution and growth: characteristics of coloring, perichondrium, chondroblasts / chondrocytes, intercellular substance). Fibrous cartilage (embryonic origin, histological organization, distribution and growth: characteristics of coloring, perichondrium, chondroblasts / chondrocytes, intercellular substance).

Bone tissues.

1. General characteristics of their histological composition and organization (types of bones, vascularity, innervation). Cells (osteoprogenitor cells, osteoblasts, osteocytes, osteoclasts). Intercellular substance (mineralized matrix, collagen fibers, glycoproteins). Bone tissue lamellar (compact or spongy) and not lamellar. Preparation of a histological preparation of bone tissue

(demineralization or section due to wear). Periosteum and endosteum. Osteoblasts (synthesis of molecules of the intercellular substance, their role in the formation of osteoclasts-RANK / RANKL, in maintaining the "niche" of hematopoietic stem cells and in mineralization). Osteocytes. Osteoclasts [their origin, cytological (podosome) and functional (hydrogen ion pump, lysosome) characteristics. Role of osteoblasts and osteoclasts in calcium metabolism (parathyroid hormone, calcitonin). Notes on osteoporosis (estrogens, RANKL and OPG).

2. Ossification. Direct ossification. Indirect ossification. Example of ossification of the bones of the face and skull (chondrocranium and neurocranium). Example of indirect ossification of a long bone of the axial skeleton (primary ossification center and secondary ossification centers). Growth in length (metaphyseal cartilage) and width and ossification of a bone. Formation of an osteone. Mechanisms of mineralization (role of osteoblasts, mineralization vesicles, collagen fibers and proteoglycans).

Blood and lymph.

Blood (composition and functions). Plasma (serum) and corpuscular elements (hematocrit, buffy coat, blood smear). Red blood cells [structural (size, shape and cytoskeleton) and functional characteristics (numbers, hemoglobin, transport of oxygen and carbon dioxide, hemolysis processes, notes on blood groups]. Notes on haemolytic pathologies (cytoskeletal anomalies, favism and sickle cell anemia) . Platelets [structural characteristics (size, chromomer, hyalomer, shape and cytoskeleton, granules) and functional characteristics (numbers, plug and thrombus)]. Activation of platelets (adhesion to collagen, exocytosis granules, exposure of phospholipids and thrombin and fibrinogen receptors, aggregation) . Notes on the intrinsic and extrinsic blood coagulation. White blood cells (types and numbers, the leukocyte formula, diapedesis). Neutrophils (how to recognize them in a blood smear, MO and TEM characteristics; "kamikaze" and "effect functions" spiderman ", the" respiratory burst "). Eosinophils (how to recognize them in a blood smear, characteristics at MO and TEM; functions, the basic protein ica major). Basophils (how to recognize them in a blood smear, MO and TEM characteristics; functions). Monocytes (how to recognize them in a blood smear, characteristics at OM and TEM). Lymphocytes (how to recognize them in a blood smear, characteristics at OM and TEM; functions).

Lymph (composition and function).

Lympho-hematopoietic tissues. Lymphoid tissues (notes on the structure and functions of the thymus, lymph nodes, spleen and bone marrow).

Hemopoiesis [the hematopoietic stem cell, hematopoietic cytokines, progenitors and precursors of differentiative cell lines, erythropoiesis, granulocytopenia, monocytopenia, thrombopoiesis (mechanisms of platelet release), lymphopoiesis].

The immune system. Innate and acquired immunity. Notes on the functions of B, T and NK lymphocytes (concept of antigen, antibodies, complement, clonal selection, gene rearrangement, immune memory, APC cells, the HLA system, the different classes of T lymphocytes).

Tooth tissues

Mineralized tissues: enamel, dentin, cement. Soft tissues: paradontium and pulp.

MUSCLE TISSUES

General characteristics, embryonic origin, classification, distribution.

Skeletal muscle tissue. General organization of a muscle. Muscle fibers cytological (shape and size, colorability, bands and striae) and ultrastructural characteristics. Myofibrils and contractile

myofilaments, the sarcomere. The actin filaments (molecular organization); myosin filaments (molecular organization); filaments and accessory proteins of the sarcomere (titin, nebulin, oscurin, protein C, myomesin); the dystrophin and the costamer. The triads and the sarcoplasmic reticulum. Neuromuscular synapse, motor plate, functioning of a cholinergic synapse. The molecular mechanism of muscle contraction (action potential, T tubules, DHPR and ryanodine receptors, calcium ions and troponins / tropomyosin, the myosin heads interaction cycle, ATP, binding sites on actin filaments). Satellite cells. Structural, metabolic and functional heterogeneity of muscle fibers (type I, type IIa, IIb and IIx fibers). Outline of muscle fiber growth factors IGF-1 and myostatin, testosterone and anabolic steroids.

Smooth muscle tissue. General organization of the tissue (laminae, small muscles, myoepithelial cells). Cytological (shape and size) and ultrastructural characteristics of the smooth muscle cell (caveole, gap junctions, dense bodies, contractile filaments). Organization of contractile filaments (caldesmone, calponina, dense bodies, intermediate filaments of the cytoskeleton). Characteristics and stimuli (nervous, hormonal, mechanical, NO) of the contraction. Unitary and multi-unit musculature. Synapses "en passant". Molecular mechanism of contraction (calmodulin, MLCK, myosin light chain phosphorylation, caldesmone / calponin).

Heart muscle tissue. General tissue organization, cardiomyocyte network and intercalary discs. Cytological (shape and size) and ultrastructural characteristics of the cardiomyocyte (intercalary discs, contractile filaments, sarcomere, dyads, mitochondria). Characteristics of cardiomyocyte contraction (sodium channels and spontaneous onset of action potential, DHPR and ryanodine channels, calcium ion and sarcomere). Special cardiomyocytes of the sinoatrial node and Purkinje cells.

NERVOUS TISSUE

General characteristics, embryonic origin, classification, distribution. CNS and SNP. The autonomic nervous system.

General organization of the tissue (neuronal networks, synapses, glia, gray matter and white matter). Connective lining (the meninges) and vascularization (the blood-brain barrier). Special histological methods for the study of nervous tissue from "black" Golgi to "Clarity" staining.

Cytological (shape and size, plates of Nissl, cytoskeleton, Golgi) and ultrastructural characteristics (neurotubules and neurofilaments, actin filaments, REG) of the neuron. Morphological and functional classification of neurons. Dendrites. The axon and its coatings (the myelin sheath and the concept of nerve fiber). Structure, ultrastructure and molecular organization of the myelin sheath. The axoplasmic flow. Regenerative capacity of the axon. General principles of the functioning of a neuron (the resting potential, the action potential and the propagation of the nerve impulse). The electrical and chemical synapses. Interneuronal chemical synapses (synaptogenesis, types and structure, pre- and postsynaptic density, neurotransmitters, neuropeptides). Histological structure of the nerves and ganglia.

The glia (astrocytes, oligodendrocytes, ependyma, microglia, Schwann cells, satellite cells). Notes on neuronal stem cells.

EMBRYOLOGY

The meiotic division. Timing and modalities of meiosis in spermatogenesis and oogenesis. Aneuploidy. Testis structure: albuginea tunic, lobules, seminiferous tubules, rectus tubules and rete

testis. Localization and function of Sertoli cells, Leydig cells and myoid cells. Male genital tract and related glands. The seminiferous epithelium. Spermatogenesis: the mitotic phase, the meiotic phase and spermiogenesis. Structure of spermatozoa. Cycle and wave of the seminiferous epithelium. The Immune Privilege of the Testis. Hormonal control of spermatogenesis. Testicular tumors.

Structure of the ovary: cortical area and medullary area. Folliculogenesis: the primordial, primary, secondary, antral, dominant, ovulatory follicle. The zona pellucida. The cells of the theca. Ovulation. The corpus luteum. Ovarian cycle and uterine cycle. Hormonal control of the ovarian cycle and follicular selection. Mutual influence between somatic cells and the oocyte: growth and maturation of the oocyte and proliferation and maturation of follicular cells. The sperm. Capacitation and acrosomal reaction of spermatozoa. Fertilization. Activation of the oocyte: calcium, cortical reaction, completion of meiosis (MPF and cytostatic factor). Parthenogenesis and imprinting. Birth control methods. Assisted reproduction techniques. Generalities of prenatal development: embryonic and fetal period and sensitivity to teratogenic agents. Diagnosis and prenatal therapy.

First week of development. The segmentation. Cloning. Embryonic stem cells (ES), adult stem cells and induced pluripotent cells (iPSCs). Preimplantation genetic diagnosis. IVF and ICSI. Activation of the embryo genome. Inactivation of the X chromosome. Imprinting and hydatiform mole.

Second week of development: Embryonic implantation. Differentiation of the trophoblast: cytotrophoblast, syncytium trophoblast and lacunar system. HCG and pregnancy test. Amniotic cavity and yolk sac. Formation of the extraembryonic mesoderm. Chorionic or extra-embryonic coelomic cavity. Decidual reaction. Ectopic implant.

Third week of development: gastrulation and formation of the three embryonic sheets. Primary epithelial-mesenchymal transition. Buccopharyngeal and cloacal membrane. Allantoid diverticulum. Development of the notochord, induction of the neural plaque (BMP inhibitors: cord, noggin and follistatin) and neurulation. Neural tube and neural crest. The paraxial, intermediate and lateral mesoderm. The intra-embryonic coelom. Formation of extra- and intra-embryonic angioblastic islands. Cardiogenic area and transverse septum. Regression of the primitive line and the sacro-coccygeal teratoma. Determination of the body axes: AVE, Hensen's node, notochord, primitive line and postero-anterior, dorso-ventral and left-right decreasing gradient of factors of the TGFbeta family (nodal and BMP). Differentiation of neural crests and importance of BMP, WNT and FGF.

Fourth week of development: lateral and cephalo-caudal folding and cylindrical definition of the body. Development of the ectoderm, mesoderm and endoderm. Homeotic genes and the body pattern along the anterior-posterior axis. Molecular mechanisms of somitogenesis (clock and wavefront model).

The neuroectoderm: development of the neural tube. Eminence or caudal gem. Three to five brain vesicles. Derivatives of the walls and cavities of the five vesicles. Development of the pituitary gland. The spinal cord and the formation of the equine tail. Derivatives of the neuroepithelium: neurons and glia of the central nervous system. Radial glial cells. Differentiation of microglia cells. The peripheral nervous system and other derivatives of neural crest cells. Importance of BMP and SHH in the differential development of neurons in the spinal cord. NGF and neurotrophic factors. Malformations: rachisis, spina bifida occulta, meningocele, meningomyelocele.

Development of the face and neck. Development and derivatives of the frontal process, arches, furrows, and pharyngeal pockets. Stomodeum and buccopharyngeal membrane. Embryonic derivation of the cartilages, bones, muscles and nerves of the face. Development of the primitive and definitive palate, tongue and thyroid. Tooth development: four stages. Malformations: cervical sinus, cleft lip (cleft lip), cleft palate, cleft lip and palate

The embryonic appendages: placenta, allantoid, yolk sac, umbilical cord. Development, structure and function of the placenta. Placental hormones. The placental circulation. Decidual reaction: basal, capsular, parietal decidua. Angiogenesis and changes in the vascular wall. Immunological tolerance. Amnios and amniotic fluid. The dizygotic and monozygotic twins. Malformations: Polyhydramnios and oligohydramnios. Fetal erythroblastosis and the Rh factor.

Practical Histology

Through the use of the optic microscope students will have to identify histological specimens, describe their organization, and correlate structure with function, at cellular and tissue level. Histological specimens to be studied are:

- Simple boundary epithelium: squamous (mesothelium, endothelium), cuboidal (glandular ducts) and columnar (intestine) epithelia.
- Stratified boundary epithelium: squamous (esophagus) and keratinized squamous (epidermis) epithelia
- Pseudostratified epithelium (trachea)
- Transitional epithelium (ureter)
- Glandular epithelium: intraepithelial, unicellular glands (the goblet cell); examples of exocrine (salivary glands) and endocrine (thyroid, parathyroids) glands; exo/endocrine gland: the pancreas.
- Connective tissue: loose connective tissue (trachea, intestine and esophagus); dense irregular connective tissue (the skin); dense regular connective tissue (tendons).
- Supportive connective tissues: tracheal cartilage and compact bone (ground and H&E)
- Blood smear
- Lympho-epithelial tissue of the thymus
- Skeletal, cardiac and smooth muscle tissues
- Nerve tissue: section of the spinal cord

COURSE STRUCTURE

The Histology course is structured in 68 hours of frontal teaching (divided into lessons of 2 or 4 hours according to the academic calendar) and 12 hours of lab practice, consisting of microscopic analysis of several histologic preparations, including sections of epithelium, lining and glandular, connective tissue proper, adipose tissue, cartilage, bone, blood and lymphatic organs, muscle tissues (skeletal, cardiac and smooth muscles) and nervous tissue (nerve and spinal cord).

COURSE GRADE DETERMINATION

The acquisition of the expected learning results is evaluated through the exam. The exam consists in a written test, followed by an oral examination. The written test consists of about 60 questions, divided in multiple-choice questions, true/false questions and associations. The oral exam includes the identification of one/two histologic slides and questions on cytology, histology and embryology.

OPTIONAL ACTIVITIES

In addition to the didactic activity, the student will be given the opportunity to take advantage of tutoring activities, upon request, and to attend seminars.

READING MATERIALS

- Ross M.H. and W. Pawlina: Histology a text and atlas, sixth edition. Wolters Kluwer/Lippincott Williams and Wilkins.
- Schoenwolf, Bleyl, Bauer and Francis-West: Larsen's Human Embryology, 5th edition.
- Moore, Persaud, Torchia. The Developing Human: Clinically Oriented Embryology. Elsevier