Must-knows

Resolution Smallest distance at which two points are still seen as separate; d = 0.61λ / NA. Shorter wavelength or higher NA gives finer resolution.

Limits Eye ~0.2 mm; light microscope ~0.2 µm; TEM ~1–3 nm in tissue sections (theoretical far less).

Magnification Total = objective × eyepiece (40× × 10× = 400×). Magnifying past the resolution limit is empty magnification: no new detail.

Immersion oil Oil (n ≈ 1.515, like glass) between coverslip and 100× objective raises NA to ~1.4, so resolution improves.

Units 1 mm = 1000 µm; 1 µm = 1000 nm. RBC ~7.5 µm; ribosome ~25 nm (TEM only).

Bright-field parts Light source, condenser (with diaphragm), stage, objective, eyepiece; image from light absorbed by stained tissue.

Electron microscopy Electrons have a far shorter wavelength than light; lenses are electromagnetic coils; vacuum column; image seen on a screen or camera.

METHOD Light microscopes

Bright-field Principle transmitted light absorbed by stained section Best for routine stained (H&E) sections

Phase contrast Principle converts differences in refractive index into light and dark Best for living, unstained cells (cell culture)

Polarising Principle polariser + analyser; birefringent structures shine Best for collagen, skeletal muscle A band, crystals, amyloid (Congo red)

Fluorescence Principle UV/blue excitation; longer-wavelength emission seen Best for fluorochrome-labelled antibodies, DNA dyes

Confocal (laser scanning) Principle pinhole rejects out-of-focus light; point scanning Best for optical sections and 3D reconstruction of thick tissue

METHOD Electron microscopes

Transmission (TEM) Image formed from electrons passing through the section Specimen ultrathin section 40–90 nm, heavy-metal stained Shows ultrastructure: organelles, ribosomes, membranes

Scanning (SEM) Image formed from secondary electrons from the surface Specimen whole specimen coated with gold/metal Shows 3D surface of cells and tissues

MORE Resolution: what decides it

Wavelength (λ) Effect shorter λ gives finer d Note blue/UV better than red; electrons best

Numerical aperture (NA = n sin α) Effect higher NA gives finer d Note dry objective ≤0.95; oil ~1.4

Magnification Effect does not change d Note beyond ~1000× NA is empty magnification

Must-knows

Routine sequence Fixation → dehydration → clearing → embedding (infiltration) → sectioning → mounting → staining → coverslipping.

Fixation Stops autolysis and bacterial decay, preserves structure; formalin (buffered 4% formaldehyde) cross-links proteins via methylene bridges.

EM fixation Glutaraldehyde (protein cross-links) then osmium tetroxide (fixes and stains lipid membranes); embed in epoxy resin.

Dehydration and clearing Graded alcohols (70% → 100%) remove water; xylene mixes with both alcohol and paraffin, so wax can enter.

Embedding Paraffin (~56–60 °C) or resin infiltrates the tissue and gives it a firm consistency for thin sectioning.

Section thickness Paraffin 3–10 µm (microtome); semithin resin ~1 µm (toluidine blue); ultrathin 40–90 nm (ultramicrotome, glass/diamond knife).

Frozen sections Cryostat; no alcohol or xylene, so lipids and enzymes survive; minutes-long intraoperative diagnosis.

H&E Haematoxylin (basic, with mordant) stains acidic structures blue-purple: nuclei, rER-rich cytoplasm. Eosin (acidic) stains basic proteins pink: cytoplasm, collagen.

Immunohistochemistry Labelled antibody shows where an antigen (protein) lies; direct or indirect (labelled secondary antibody amplifies signal).

MORE Steps of paraffin processing

1 Fixation Agent 10% neutral buffered formalin Purpose prevent autolysis, preserve structure

2 Dehydration Agent graded alcohols (ethanol 70→100%) Purpose remove water

3 Clearing Agent xylene (or toluene) Purpose replace alcohol; miscible with paraffin; tissue turns translucent

4 Infiltration and embedding Agent molten paraffin wax, set in a block Purpose firm support for sectioning

5 Sectioning Agent rotary microtome, steel knife Purpose sections 3–10 µm

8 Staining Agent e.g. H&E Purpose contrast for structures

METHOD Light vs electron microscopy preparation

Fixative Light microscopy buffered formalin TEM glutaraldehyde, then osmium tetroxide

Embedding medium Light microscopy paraffin TEM epoxy resin (Epon, Araldite)

Instrument Light microscopy microtome TEM ultramicrotome

Knife Light microscopy steel TEM glass or diamond

Thickness Light microscopy 3–10 µm TEM 40–90 nm (semithin survey 0.5–1 µm)

Stain / contrast Light microscopy dyes (H&E) TEM heavy metals: uranyl acetate, lead citrate

METHOD Routine and special stains

Haematoxylin Shows nuclei, rER, acidic matrix (basophilic) Colour blue-purple

Eosin Shows cytoplasm, collagen, RBCs (acidophilic) Colour pink-red

Masson's trichrome Shows collagen / muscle and cytoplasm / nuclei Colour blue or green / red / black

MORE Dye binding terms

Basophilic Meaning binds basic dye; rich in acidic groups Examples nuclei (DNA phosphate), plasma-cell cytoplasm (rRNA of rER), cartilage matrix

Acidophilic (eosinophilic) Meaning binds acidic dye; rich in basic proteins Examples cytoplasm, mitochondria, collagen, RBCs

Mnemonics

Mnemonic Fix, Dehydrate, Clear, Embed, Cut, Stain = order of routine paraffin processing

Must-knows

Four basic tissues Epithelial, connective, muscle and nervous tissue. Histology = tissues and how they build organs; 'histo' = tissue or web.

Origin from all three germ layers Epithelia arise from ectoderm, mesoderm and endoderm (e.g. epidermis, endothelium, gut lining).

Defining features Tightly packed polyhedral cells with little extracellular matrix, polarised (apical, lateral, basal domains), resting on a basement membrane. Avascular: fed by diffusion from capillaries in the lamina propria.

Functions Covering, lining, protection (epidermis, urothelium), absorption (intestine), secretion (stomach, glands), sensation, transport (cilia), contractility (myoepithelial cells).

Naming rule Number of layers (simple / stratified) + shape of the cells of the most superficial layer (squamous, cuboidal, columnar). Stratified squamous is further split by keratinisation.

Pseudostratified is simple Every cell rests on the basement membrane but not every cell reaches the lumen; nuclei lie at several levels so it looks layered. Short basal cells are stem cells.

Apical specialisations Microvilli (actin core, absorption), stereocilia (long microvilli, epididymis), cilia (9+2 axoneme on basal bodies, motile), glycocalyx (carbohydrate coat).

Goblet cell Unicellular exocrine gland secreting mucin (heavily glycosylated protein): PAS-positive, pale in H&E; apical mucin mass pushes the nucleus to the basal pole.

Stratified squamous: keratinised vs not Non-keratinised: living nucleated surface cells on moist surfaces (mouth, oesophagus, vagina, cornea). Keratinised: dead anucleate keratin-filled surface cells (epidermis).

Transitional epithelium (urothelium) Lines renal calyces, pelvis, ureter, bladder, proximal urethra. Relaxed 4–5 layers with dome-shaped (umbrella) surface cells, some binucleate; stretched 2–3 layers, flat surface cells.

Gland development Surface epithelium proliferates into the connective tissue. Exocrine: connection persists as a duct. Endocrine: connection is lost, product enters capillaries.

Modes of secretion Merocrine = exocytosis, no cytoplasm lost (most common). Apocrine = product plus apical cytoplasm (mammary lipid). Holocrine = whole cell breaks down (sebaceous).

TYPES Classification of covering epithelia

Simple squamous Layers and cell shape one layer, flat cells, flat nuclei bulging into lumen Where endothelium (vessels, heart), mesothelium (pleura, peritoneum, pericardium), lung alveoli, parietal layer of Bowman's capsule, thin limb of loop of Henle, corneal endothelium, inner and middle ear Function exchange (gases, fluid), filtration, lubrication, low friction

Simple cuboidal Layers and cell shape one layer, cells as tall as wide, round central nucleus Where kidney tubules, thyroid follicles, small exocrine ducts, ovary surface Function secretion, absorption, barrier

Simple columnar Layers and cell shape one layer, tall cells, oval nuclei near the basal pole Where stomach surface and gastric pits, small intestine and colon (with goblet cells), gallbladder (microvilli, no goblet cells); ciliated form in uterine tube Function absorption, secretion, protection

Pseudostratified columnar Layers and cell shape one layer, all cells on basement membrane, nuclei at several levels Where trachea and bronchi (ciliated, with goblet cells = respiratory epithelium), nasal cavity; epididymis and ductus deferens (stereocilia) Function secretion, moving mucus by cilia, protection

Stratified squamous non-keratinised Layers and cell shape many layers, cuboidal basal to flat nucleated living surface cells Where oral cavity, oesophagus, vagina, ectocervix, anal canal, corneal epithelium, inner lips Function protection on moist surfaces, some secretion

TYPES Classification of covering epithelia (cont.)

Stratified squamous keratinised Layers and cell shape many layers, surface cells dead, anucleate, filled with keratin Where epidermis of skin Function protection against abrasion, water loss, microbes

Stratified columnar Layers and cell shape several layers, superficial cells columnar Where conjunctiva, large excretory ducts, parts of male urethra Function protection, secretion

Transitional (urothelium) Layers and cell shape relaxed 4–5 layers (basal cuboidal, intermediate polygonal, dome-shaped umbrella surface cells); stretched 2–3 layers, flat surface cells Where renal calyces and pelvis, ureter, bladder, proximal urethra Function protection, distension, barrier to urine

CELLS Cell shapes

Squamous Proportions width greater than height Nucleus flat, elongated Example endothelium, alveoli

Cuboidal Proportions height ≈ width ≈ depth Nucleus round, central Example thyroid follicle, kidney tubule

Columnar Proportions height greater than width Nucleus oval, near basal pole Example intestine, stomach

SURFACE Apical surface specialisations

Microvilli Core / composition actin filaments anchored in terminal web Size and motility ≈1 µm long, 0.08 µm wide; non-motile; brush (striated) border Where enterocytes, proximal convoluted tubule

Stereocilia Core / composition actin filaments (very long microvilli, not cilia) Size and motility long, non-motile; branched in epididymis Where epididymis, ductus deferens; sensory hair cells of inner ear

Cilia Core / composition axoneme of 9 peripheral doublets + central pair (9+2) of microtubules, dynein arms; arise from basal bodies Size and motility 5–10 µm long, 0.2 µm wide; motile, beat in one direction Where respiratory tract, uterine tube

Glycocalyx Core / composition carbohydrate chains of membrane glycoproteins and glycolipids Size and motility thin coat on apical membrane Where all cells, prominent on enterocytes

SURFACE Epidermis layers (keratinised stratified squamous), deep to superficial

Stratum basale Cells one layer of cuboidal to low columnar cells on basement membrane Key feature mitosis (stem cells)

Stratum spinosum Cells several layers of polygonal cells Key feature desmosomes give 'spiny' (prickle) look

Stratum granulosum Cells 3–5 layers of flattened cells Key feature keratohyalin granules

Stratum lucidum Cells thin layer of flat eosinophilic cells Key feature only in thick skin (palms, soles); eleidin, pale/bright

Stratum corneum Cells many layers of dead anucleate flat cells Key feature filled with keratin

TYPES Gland classification

Ducts Type Exocrine Feature duct to surface or lumen Example sweat, sebaceous, salivary

Ducts Type Endocrine Feature ductless, secretes hormones into blood Example thyroid, adrenal, pituitary

Cell number Type Unicellular Feature single secretory cell Example goblet cell

Cell number Type Multicellular Feature many cells, secretory portion + duct Example sweat, salivary glands

Duct Type Simple Feature unbranched duct Example sweat gland

Duct Type Compound Feature branched duct Example salivary glands, pancreas

Secretory portion Type Tubular Feature tube of about duct diameter (simple, coiled or branched) Example intestinal crypts; sweat gland (coiled); gastric glands (branched)

Secretory portion Type Acinar (alveolar) Feature rounded sac-like end piece Example sebaceous gland (branched acinar), exocrine pancreas (compound acinar)

MORE Modes of secretion

Merocrine (eccrine) How product leaves exocytosis of vesicles at apical surface Cell fate intact, keeps secreting Example most glands: eccrine sweat, salivary, pancreas

Apocrine How product leaves product released with part of apical cytoplasm Cell fate apical region reforms Example lipid of mammary gland

Holocrine How product leaves whole cell fills with product and breaks down Cell fate dies; replaced by mitosis of peripheral basal cells Example sebaceous gland

Mnemonics

Mnemonic Come Let's Get Sun Burned = Epidermis superficial to deep: corneum, lucidum, granulosum, spinosum, basale

Mnemonic Mero = merely exocytosis, Apo = apex, Holo = whole = Merocrine loses nothing, apocrine loses apical cytoplasm, holocrine loses the whole cell

Must-knows

Three components Cells + extracellular matrix (ground substance and fibres). Matrix dominates; cells are scattered in it.

Origin: mesenchyme All connective tissues arise from embryonic mesenchyme (mostly mesoderm). Fibroblasts, chondroblasts, osteoblasts, adipocytes and haematopoietic stem cells descend from it.

Functions Support and binding, nutrition of avascular epithelia by diffusion through ground substance, defence (immune cells), protection, fat storage, repair.

Loose vs dense Loose: many cells, much ground substance, thin loosely woven fibres. Dense: few cells, little ground substance, thick collagen bundles; regular (one direction) or irregular (many directions).

Resident vs wandering cells Resident (fixed): fibroblasts, myofibroblasts, macrophages, adipocytes, mast cells, mesenchymal cells. Wandering (from blood): lymphocytes, plasma cells, neutrophils, eosinophils, basophils, monocytes.

Fibroblast Commonest cell; synthesises collagen, elastin, GAGs, proteoglycans and glycoproteins. Active form: large, pale oval nucleus, basophilic cytoplasm; resting fibrocyte: smaller, dark slim nucleus.

Macrophages and mast cells Macrophages come from blood monocytes; phagocytic, antigen-presenting. Mast cells: granules of histamine and heparin, metachromatic; release on IgE cross-linking (immediate hypersensitivity).

Ground substance Hydrated, viscous gel of GAGs, proteoglycans and multiadhesive glycoproteins; lubricant and barrier to microorganisms; fine granular in TEM, lost in routine sections.

Collagen hierarchy Tropocollagen (3 alpha chains, triple helix, 300 nm) -> fibril (64-67 nm cross-banding) -> fibre -> bundle. Type I is the commonest (~90% of body collagen).

Elastic and reticular fibres Elastic: elastin core + fibrillin microfibrils, thin branching network; orcein / resorcin-fuchsin. Reticular: type III collagen, argyrophilic (silver, black) and PAS-positive, made by fibroblasts (by reticular cells in lymphoid organs and marrow).

White vs brown fat White: unilocular, one droplet, peripheral flattened nucleus (signet ring), energy store. Brown: multilocular, central round nucleus, many mitochondria with UCP1 (thermogenin) -> heat.

CELLS Cells of connective tissue proper

Fibroblast Origin / resident or wandering mesenchyme; resident How it looks elongated, pale oval nucleus with nucleolus; basophilic cytoplasm with RER and Golgi Function makes fibres and ground substance

Fibrocyte Origin / resident or wandering inactive fibroblast; resident How it looks smaller, spindle-shaped, dark slim nucleus, little cytoplasm Function maintenance; reactivates in repair

Macrophage Origin / resident or wandering blood monocyte; resident (histiocyte) How it looks irregular surface with processes, indented nucleus, many lysosomes Function phagocytosis, antigen presentation, cytokines

Mast cell Origin / resident or wandering bone-marrow progenitor; resident How it looks large oval, central nucleus hidden by many basophilic metachromatic granules; near small vessels Function releases histamine, heparin, leukotrienes, ECF-A

Adipocyte Origin / resident or wandering mesenchyme; resident How it looks white: one droplet, peripheral nucleus; brown: many droplets, central nucleus Function fat storage, insulation, heat (brown), leptin

Plasma cell Origin / resident or wandering B lymphocyte; wandering How it looks oval, eccentric nucleus with clock-face chromatin, basophilic cytoplasm, pale Golgi halo Function secretes antibodies

CELLS Fibres of the extracellular matrix

Collagen Made of type I collagen (tropocollagen triple helix) Light / EM look thick, unbranched, wavy bundles; EM: fibrils with 64-67 nm banding Stain eosin pink; blue with Mallory/trichrome Property / where great tensile strength, little stretch; tendon, dermis, bone

CELLS Fibres of the extracellular matrix (cont.)

Reticular Made of type III collagen, heavily glycosylated Light / EM look thin, branched delicate network Stain silver (black, argyrophilic), PAS+ Property / where supporting stroma of lymph nodes, spleen, marrow, liver, basement membranes

Elastic Made of elastin core + fibrillin microfibrils Light / EM look thin, straight, branching, form networks or fenestrated sheets Stain orcein, resorcin-fuchsin, Verhoeff Property / where stretch and recoil; arteries, lung, elastic ligaments

MATRIX Collagen types (main ones)

I Form thick fibrils, fibres Where skin (dermis), bone, tendon, ligaments, dentin, fibrocartilage

II Form thin fibrils Where hyaline and elastic cartilage, vitreous body

III Form reticular fibres Where skin, lung, blood vessels, lymphoid organs, smooth muscle

IV Form sheet-like network Where basal lamina

TYPES Classification of connective tissues

Embryonic CT Ground substance viscous gel, few fibres Members mesenchyme; mucous (gelatinous) tissue

Connective tissue proper Ground substance semi-liquid gel Members loose (areolar); dense regular; dense irregular

Specialised: adipose, reticular, elastic Ground substance gel Members white (unilocular) and brown (multilocular) fat; reticular tissue; elastic tissue

Cartilage Ground substance firm, rubbery Members hyaline, elastic, fibrocartilage

Bone Ground substance calcified, very hard Members woven (immature), lamellar (mature): compact and spongy

Blood and lymph Ground substance liquid (plasma) Members blood cells and platelets in plasma

TYPES Types of connective tissue proper and related tissues

Loose (areolar) Structure many cells of all types, much ground substance, thin collagen + elastic fibres in all directions Where lamina propria, papillary dermis, hypodermis, around vessels, mesentery Function binding, diffusion, defence, oedema site

Dense irregular Structure thick collagen bundles interwoven in many planes, few fibroblasts Where reticular dermis, organ capsules, submucosa of gut Function resists stretch from many directions

Dense regular Structure parallel collagen bundles with rows of flattened fibroblast (tenocyte) nuclei Where tendons, ligaments, aponeuroses, cornea stroma Function resists pull in one direction

White adipose Structure unilocular cells in lobules separated by CT septa, rich capillaries Where subcutaneous, visceral (omentum, mesentery), buttocks, thighs, breast Function energy store, insulation, padding, endocrine

Brown adipose Structure multilocular cells, very vascular, sympathetic nerves Where newborn: interscapular, neck; adult: supraclavicular, paravertebral, mediastinal, para-aortic, suprarenal Function non-shivering heat

TYPES White vs brown adipose tissue

Lipid droplets White (yellow, unilocular) one large droplet fills the cell Brown (multilocular) many small droplets

Nucleus White (yellow, unilocular) flattened, peripheral (signet ring) Brown (multilocular) round, central

Mitochondria White (yellow, unilocular) few Brown (multilocular) very many, with UCP1 (thermogenin)

Function White (yellow, unilocular) energy store, insulation, padding, leptin secretion Brown (multilocular) heat production (uncoupled oxidation)

Distribution White (yellow, unilocular) most fat in adults: subcutaneous, visceral; in newborns too (e.g. buttocks) Brown (multilocular) newborns (~2-5% of body weight); adults: supraclavicular, paravertebral, mediastinal

H&E look White (yellow, unilocular) empty polygonal spaces with thin pink rims Brown (multilocular) pale, foamy (vacuolated) cytoplasm, many capillaries

Mnemonics

Mnemonic Type 1: bONE; Type 2: carTWOlage; Type 3: reTHREEcular (reticular fibres, vessels); Type 4: floor (basement membrane) = Main collagen types and where they are

Must-knows

Three cartilage types Hyaline, elastic and fibrocartilage; told apart by the kind and amount of fibres in the matrix. Hyaline is the commonest and forms most of the fetal skeleton.

Cartilage is avascular No vessels, nerves or lymphatics; chondrocytes are fed by diffusion from perichondrial capillaries (or synovial fluid). Low metabolic rate, so it heals poorly.

Perichondrium Dense connective tissue sheath: outer fibrous layer (collagen I, fibroblasts), inner chondrogenic layer (chondroblast precursors). Absent from articular cartilage and fibrocartilage.

Chondrocytes and isogenous groups Chondroblasts trapped in matrix become chondrocytes, one per lacuna; mitosis yields isogenous groups. Superficial cells flattened parallel to the surface, deep cells larger and rounder.

Bone = cells + mineralised matrix Cells: osteoprogenitors, osteoblasts, osteocytes, osteoclasts. Matrix: type I collagen (organic) with hydroxyapatite $Ca_{10}(PO_4)_6(OH)_2$ laid along and between the fibrils.

Osteoblast lineage Osteoprogenitor -> osteoblast (secretes osteoid toward the bone surface) -> osteocyte in a lacuna, or flattened bone lining cell.

Osteocyte network One osteocyte per lacuna; processes in canaliculi joined by gap junctions pass nutrients and signals; fluid flow in the lacunocanalicular system lets them sense load.

Osteoclast Huge (up to ~100 um), up to ~50 nuclei, acidophilic; fused monocyte-macrophage cells. Resorbs bone in Howship lacunae via a ruffled border sealed off by a clear zone.

Woven vs lamellar bone Woven (primary): irregular collagen, many osteocytes, less mineral; first in development and repair, persists at sutures, tooth sockets, tendon insertions. Lamellar (secondary) forms adult compact and spongy bone.

Osteon (Haversian system) Structural unit of compact bone: 4-20 concentric lamellae around a central canal, bounded by a cement line; collagen changes direction in neighbouring lamellae. Volkmann canals link central canals crosswise.

Periosteum and endosteum Periosteum: outer fibrous + inner osteogenic layer, anchored by Sharpey fibres, absent at articular surfaces. Endosteum: thin single cell layer lining marrow cavity, trabeculae and canals.

TYPES Cartilage types

Hyaline Matrix fibres fine type II collagen fibrils, masked by ground substance Perichondrium yes (not on articular surfaces) Where articular surfaces, costal, nasal, larynx (thyroid, cricoid, most of arytenoid), trachea, bronchi, epiphyseal plates, fetal skeleton Key features glassy, homogeneous, basophilic; isogenous groups; calcifies with age

Elastic Matrix fibres dense elastic fibre network + type II collagen Perichondrium yes Where auricle, wall of external acoustic meatus, auditory tube, epiglottis, corniculate and cuneiform cartilages, vocal process of arytenoid Key features yellowish fresh; flexible support; elastic fibres shown by orcein (brown) or resorcin-fuchsin; does not calcify

Fibrocartilage Matrix fibres thick type I collagen bundles + some type II Perichondrium no Where intervertebral disc (anulus fibrosus), pubic symphysis, menisci, articular discs (temporomandibular, sternoclavicular), some tendon and ligament insertions Key features small chondrocytes single or in rows between bundles; mostly acidophilic; withstands compression and tension

CELLS Cartilage cells and matrix regions

Perichondrium, fibrous layer Description outer dense connective tissue, type I collagen, fibroblasts Note carries the capillaries that feed the cartilage

Perichondrium, chondrogenic layer Description inner cellular layer of chondroprogenitor cells Note source of appositional growth

Chondroblast Description matrix-secreting cell at the surface, not yet in a lacuna Note becomes a chondrocyte once enclosed

Chondrocyte Description cell in a lacuna; flattened superficially, large and round deep Note shrinks from the lacunar wall in paraffin sections

CELLS Cartilage cells and matrix regions (cont.)

Isogenous group Description cluster of chondrocytes from one parent cell Note sign of interstitial growth

Territorial matrix Description basophilic matrix around each isogenous group Note high sulfated GAG content

Interterritorial matrix Description matrix between groups; fewer proteoglycans, type II collagen Note stains paler

CELLS Bone cells

Osteoprogenitor cell Origin mesenchymal stem cells Look flattened, spindle-shaped, pale oval nucleus, scant cytoplasm Where inner periosteum, endosteum Function divides; differentiates into osteoblasts

Osteoblast Origin osteoprogenitor cell Look cuboidal to columnar, basophilic (much RER), in a single layer Where bone surfaces where bone forms Function secretes osteoid (type I collagen, proteoglycans, osteocalcin) toward the bone; starts mineralisation (alkaline phosphatase, matrix vesicles)

Osteocyte Origin osteoblast enclosed in matrix Look less RER, smaller Golgi, condensed chromatin; many processes Where one per lacuna; processes in canaliculi Function maintains matrix; senses load; gap junctions with neighbours

Osteoclast Origin fusion of monocyte-macrophage precursors (haematopoietic) Look giant (up to ~100 um), up to ~50 nuclei, acidophilic Where Howship lacunae on bone surfaces Function resorbs bone: H+ pumps dissolve mineral, cathepsin K and MMPs digest collagen

MATRIX Bone matrix composition

Organic (osteoid) Components ~90% type I collagen; proteoglycans; glycoproteins osteocalcin, osteonectin, osteopontin, bone sialoprotein Gives bone tensile strength, flexibility

Inorganic Components hydroxyapatite $Ca_{10}(PO_4)_6(OH)_2$ along and between collagen fibrils; also carbonate, citrate, Mg, Na, K Gives bone hardness, stiffness

TYPES Bone tissue types

Woven (primary) Structure irregular collagen, many osteocytes, lower mineral (less dense on X-ray) Where embryo, fracture repair; adult sutures, tooth sockets, tendon insertions

Lamellar (secondary) Structure collagen in parallel sheets (lamellae, 3-7 um) Where all adult compact and spongy bone

Compact (cortical) Structure dense, osteons + circumferential and interstitial lamellae Where shafts of long bones, outer and inner tables of flat bones

Spongy (cancellous) Structure network of trabeculae with marrow spaces; usually no osteons; fed from marrow Where epiphyses, metaphyses, diploe, vertebral bodies

MORE Structures of compact bone

Osteon (Haversian system) Description 4-20 concentric lamellae around a central canal, parallel to the long axis

Concentric lamellae Description collagen parallel within a lamella, at a different angle in the next

Central (Haversian) canal Description lengthwise canal with vessels, nerves, loose connective tissue; lined by endosteum

Perforating (Volkmann) canal Description crosswise canal joining central canals and the surfaces; not ringed by lamellae

Lacunae Description spaces holding osteocytes; in compact and spongy bone

Canaliculi Description fine channels running radially between lacunae and to the central canal

Interstitial lamellae Description angular remnants of older osteons partly resorbed in remodelling

Outer circumferential lamellae Description around the whole shaft, deep to the periosteum

Inner circumferential lamellae Description around the shaft at the endosteal surface, facing the medullary cavity

MORE Coverings and marrow

Periosteum, fibrous layer Make-up outer dense connective tissue, type I collagen, fibroblasts Note covers bone except articular surfaces

Periosteum, osteogenic layer Make-up inner cellular layer, osteoprogenitor cells Note bone growth in width and fracture repair

Sharpey (perforating) fibres Make-up type I collagen bundles from periosteum into outer circumferential lamellae Note anchor periosteum, tendons, ligaments

Endosteum Make-up thin, mostly one layer of osteoprogenitor and lining cells Note lines marrow cavity, trabeculae, central and perforating canals

Red marrow Make-up haematopoietic cells on a stroma of reticular cells and fibres Note high activity; spongy bone of flat bones, vertebrae, epiphyses

Yellow marrow Make-up mostly adipocytes Note low activity; adult diaphyses; can revert to red marrow

Mnemonics

Mnemonic Osteoblasts Build, osteoClasts Chew (Carve) = blasts form bone, clasts resorb it

Must-knows

Blood is a connective tissue Cells (formed elements) suspended in a fluid matrix, plasma. Its fibres, fibrin threads, appear only when blood clots. pH 7.35–7.45; about 38 °C.

Volume About 5 L in an adult (men 5–6 L, women 4–5 L), roughly 7–8% of body weight.

Centrifuged blood Plasma ~55%, packed erythrocytes ~45% (haematocrit), buffy coat <1% (leukocytes + platelets) between them.

Erythrocyte Biconcave disc ~7.5 µm, no nucleus or organelles; ~97% of dry content haemoglobin, plus carbonic anhydrase. ATP from anaerobic glycolysis, so it uses none of the O2 it carries. Lives ~120 days.

Haemoglobin HbA = 2 α + 2 β globin chains, each with a haem whose Fe2+ binds one O2: up to 4 O2 per molecule, reversibly.

Neutrophil Commonest leukocyte (50–70%), 12–15 µm, 3–5 nuclear lobes; pale specific granules + azurophilic granules. Phagocytoses bacteria; blood half-life ~6–7 h, 1–4 days in tissue, then apoptosis.

Eosinophil and basophil Eosinophil: bilobed nucleus, orange-red granules; fights helminths, phagocytoses antigen–antibody complexes, falls with corticosteroids. Basophil: rarest (<1%), dark granules with heparin + histamine, mast-cell-like.

Monocyte Largest leukocyte (12–20 µm), indented/kidney-shaped nucleus, grey-blue cytoplasm. Leaves blood after 1–3 days and becomes a macrophage (mononuclear phagocyte system, incl. osteoclasts); does not return.

Lymphocytes Small 6–8 µm, round dark nucleus, thin blue rim. B cells mature in marrow → plasma cells (humoral immunity); T cells mature in thymus (cellular immunity, most blood lymphocytes); NK cells kill infected/tumour cells without prior sensitisation.

Platelets Anucleate fragments of megakaryocyte cytoplasm, 2–4 µm, 150 000–400 000/µL, ~10 days. Peripheral hyalomere (marginal microtubule bundle) + central granulomere (granules, mitochondria, glycogen).

Haemopoietic hierarchy Pluripotent stem cell → common myeloid progenitor (erythrocytes, platelets, granulocytes, monocytes, mast cells) or common lymphoid progenitor (B, T, NK). Red marrow is the adult site.

MORE Composition of blood

Plasma — Amount ~55% of blood volume — Note water ~92%, proteins ~7%, other solutes ~1% (by weight)

Erythrocytes (haematocrit) — Amount ~45% of volume; ~4–6 million/µL — Note women ~3.9–5.5, men ~4.1–6.0 million/µL

Buffy coat — Amount <1% of volume — Note leukocytes + platelets

Leukocytes — Amount 4 500–11 000/µL — Note granulocytes (neutrophil, eosinophil, basophil) and agranulocytes (lymphocyte, monocyte)

Platelets — Amount 150 000–400 000/µL — Note fragments, not cells

MORE Plasma proteins

Albumin (most abundant) — Made by liver — Function main source of colloid osmotic (oncotic) pressure; carrier of hormones, drugs, fatty acids

α and β globulins — Made by liver — Function carriers of iron (transferrin), haemoglobin (haptoglobin), lipids; add to oncotic pressure

MORE Plasma proteins (cont.)

γ globulins (immunoglobulins) Made by plasma cells Function antibodies; the one major plasma protein not made by liver

Fibrinogen (smallest share of the main groups, ~4–7%) Made by liver Function converted to fibrin threads of the clot

CELLS Leukocytes

Neutrophil (granulocyte) Diameter 12–15 µm % of leukocytes 50–70% Nucleus 3–5 lobes; drumstick (Barr body) in women; band = unsegmented young form Cytoplasm / granules pale lilac-pink specific granules (lysozyme, lactoferrin, collagenase, alkaline phosphatase) + azurophilic lysosomes Life blood ~6–7 h, tissue 1–4 days Main role phagocytosis of bacteria; H_2O_2 and other oxidants

Eosinophil (granulocyte) Diameter 12–15 µm % of leukocytes 1–4% Nucleus bilobed Cytoplasm / granules large orange-red specific granules with crystalloid core (major basic protein, eosinophil peroxidase, histaminase) Life 1–2 weeks Main role kills helminths; phagocytoses antigen–antibody complexes; rises in allergy, falls with corticosteroids

Basophil (granulocyte) Diameter 12–15 µm % of leukocytes 0.5–1% (rarest) Nucleus bi- or irregularly lobed, hidden by granules Cytoplasm / granules coarse dark blue-purple granules: heparin, histamine, leukotrienes (SRS-A) Life several months Main role vasodilation and allergy (IgE receptors); supplements mast cells

Lymphocyte (agranulocyte) Diameter small 6–8 µm, large up to 18 µm % of leukocytes 20–30% Nucleus round, dense, heterochromatic, nearly fills cell Cytoplasm / granules thin rim of pale blue cytoplasm (free ribosomes), few azurophilic granules Life days to years Main role specific immunity

Monocyte (agranulocyte) Diameter 12–20 µm (largest) % of leukocytes 3–8% Nucleus oval, indented or kidney-shaped, eccentric, finer paler chromatin Cytoplasm / granules grey-blue, fine azurophilic granules (lysosomes), often vacuolated Life blood 1–3 days, then months as macrophage Main role becomes macrophage; removes debris and microbes with lysosomal enzymes; antigen presentation

MORE Bone marrow

Red marrow Feature haemopoietic; all marrow at birth; in adults in flat bones (skull, ribs, sternum, pelvis), vertebrae and proximal epiphyses of femur and humerus

Yellow marrow Feature mostly adipocytes; replaces red marrow with age; reverts to red after severe bleeding or hypoxia

Stroma Feature reticular cells + reticular fibres (type III collagen), macrophages

Parenchyma Feature haemopoietic cords between sinusoids

Sinusoids Feature discontinuous endothelium; new cells cross the wall to enter blood

MORE Haemopoietic lineages

Erythropoiesis Progenitor common myeloid → MEP (CFU-E) Stages proerythroblast → basophilic → polychromatophilic → orthochromatophilic erythroblast → reticulocyte Product erythrocyte

Granulopoiesis Progenitor common myeloid → GMP (CFU-G) Stages myeloblast → promyelocyte → myelocyte → metamyelocyte → band → segmented Product neutrophil, eosinophil, basophil

Monocytopoiesis Progenitor common myeloid → GMP (CFU-M) Stages monoblast → promonocyte → monocyte Product monocyte → macrophage, osteoclast, myeloid dendritic cell

Thrombopoiesis Progenitor common myeloid → MEP (CFU-Meg) Stages megakaryoblast → promegakaryocyte → megakaryocyte (endomitosis, polyploid) Product platelets (cytoplasm fragments)

Lymphopoiesis Progenitor common lymphoid Stages lymphoblast → prolymphocyte → lymphocyte; T precursors mature in thymus Product B, T, NK cells; B → plasma cell

Mnemonics

Mnemonic	Never Let Monkeys Eat Bananas = Leukocytes from commonest to rarest: neutrophil, lymphocyte, monocyte, eosinophil, basophil
Mnemonic	60, 30, 6, 3, 1 = Approximate differential count (%) in the same order

Must-knows

Three muscle types Skeletal (striated, voluntary), cardiac (striated, involuntary), smooth (no striations, involuntary). Striation = sarcomeres of neighbouring myofibrils lying in register.

Muscle vocabulary Sarcolemma = cell membrane; sarcoplasm = cytoplasm; sarcoplasmic reticulum (SR) = smooth ER storing Ca2+; a skeletal 'fibre' = one whole multinucleated cell.

Origin and development Mostly mesoderm. Skeletal fibre = syncytium from fusion of many myoblasts into a myotube (central nuclear chain), whose nuclei then move to the periphery.

Satellite cells Quiescent mononuclear myogenic stem cells between sarcolemma and external lamina; on injury they proliferate and fuse to repair or form fibres.

Sarcomere Z disc to Z disc, ~2.5 um at rest = one A band + half of each flanking I band. The contractile unit of striated muscle.

Sliding filament model Filaments keep their length; thin filaments slide toward the M line. Sarcomere, I band and H zone shorten; A band constant.

Triad vs diad Skeletal: triad = T tubule + 2 terminal cisternae, at each A-I junction (2 per sarcomere). Cardiac: diad = T tubule + 1 cistern, at the Z line.

Ca2+ controls contraction ACh at the motor end plate -> action potential along sarcolemma and T tubules -> SR releases Ca2+ -> Ca2+ binds troponin C -> tropomyosin shifts, uncovering the myosin-binding sites on actin -> cross-bridge cycling.

Intercalated discs Step-like dark transverse lines at cardiac cell ends, at the level of a Z line. Transverse part: fasciae adherentes + desmosomes; lateral part: gap junctions (functional syncytium).

Smooth muscle contractile apparatus No myofibrils or sarcomeres; actin and myosin form an oblique lattice anchored to dense bodies (cytoplasmic) and dense plaques (sarcolemmal), linked by intermediate filaments.

TYPES Skeletal vs cardiac vs smooth muscle

Cell shape Skeletal long unbranched cylinder Cardiac short, branched, joined end to end Smooth spindle (fusiform), tapering ends

Nuclei Skeletal many, flattened, peripheral, deep to sarcolemma Cardiac 1 (or 2), central, oval Smooth 1, central, elongated (cigar); corkscrew when contracted

Striations Skeletal yes Cardiac yes Smooth no

Myofibrils / sarcomeres Skeletal yes Cardiac yes Smooth no; oblique lattice on dense bodies

Cell junctions Skeletal none between fibres Cardiac intercalated discs (fascia adherens, desmosome, gap junction) Smooth gap junctions

Control Skeletal voluntary (somatic motor nerve) Cardiac involuntary, intrinsic pacemaker + autonomic Smooth involuntary: autonomic, hormones, stretch

Contraction Skeletal strong, quick, discontinuous Cardiac strong, quick, rhythmic, continuous Smooth weak, slow, sustained; fatigue-resistant

MORE Organisation of skeletal muscle

Whole muscle What it is many fascicles Wrapping / contents epimysium (dense CT) Note continues into tendon; deep fascia superficial to it

Fascicle What it is bundle of fibres Wrapping / contents perimysium Note larger arteries, veins, nerves run here

Muscle fibre What it is one multinucleated cell Wrapping / contents endomysium (reticular fibres, capillaries, nerve twigs) + external lamina Note nuclei between fibres = fibroblasts, endothelium

SURFACE Bands and lines of the sarcomere (cont.)

Z disc (line) Contains alpha-actinin; anchors thin filaments, titin Appearance thin dark line bisecting I band In contraction Z discs move closer

I band Contains thin filaments only Appearance light; isotropic in polarised light In contraction narrows; almost gone at full contraction

A band Contains thick filaments (+ overlapping thin) Appearance dark; anisotropic (birefringent) In contraction constant length (= thick filament)

H zone Contains thick filaments only (no thin-filament overlap); its centre is the bare zone, without myosin heads Appearance paler zone in middle of A band In contraction narrows; disappears at full contraction

Sarcomere Contains Z to Z: A band + 2 half I bands Appearance ~2.5 um resting In contraction shortens

MORE Proteins of the striated contractile apparatus

Actin (G -> F) Location thin filament: 2 F-actin strands in a helix Role has myosin-binding sites

Tropomyosin Location in grooves of F-actin helix Role covers myosin-binding sites at rest

Troponin (TnT, TnC, TnI) Location on tropomyosin, thin filament Role TnT binds tropomyosin, TnC binds Ca2+, TnI inhibits actin-myosin binding

Myosin II Location thick filament (~200-500 molecules, bipolar) Role 2 heavy + 4 light chains; head = actin-binding site + ATPase; hinge lets head pivot

Titin Location Z disc to M line, along thick filament Role elastic spring; centres thick filament; passive elasticity

MORE Cardiac muscle specialisations

Fascia adherens Where transverse part of intercalated disc Feature anchors actin of terminal sarcomere Function transmits pull cell to cell

Desmosome (macula adherens) Where transverse part of intercalated disc Feature anchors intermediate filaments Function keeps cells from pulling apart

Gap junction Where lateral part of intercalated disc Feature ionic channels Function spreads excitation: functional syncytium

CELLS Smooth muscle cell structure

Dense bodies Feature cytoplasmic; contain alpha-actinin (like Z discs) Function anchor thin filaments

Thin and thick filaments Feature oblique crisscross lattice; thick filaments side-polar Function contraction; cell shortens and bulges, nucleus corkscrews

METHOD Identifying muscle in sections

Skeletal Longitudinal section long parallel fibres, cross-striations, many flattened peripheral nuclei Transverse section large polygonal profiles of even size, peripheral nuclei, none central Clue myofibrils as fine dots in TS

Cardiac Longitudinal section branching fibres, central nuclei, step-like intercalated discs Transverse section irregular profiles of varied size, single central nucleus in pale area; many capillaries Clue pale perinuclear zone

Smooth Longitudinal section overlapping spindles, elongated central nuclei, wavy when contracted Transverse section small round profiles, only some with a nucleus; little CT Clue no striations

Mnemonics

Mnemonic H zone, I band shrink; A stays = In contraction H and I narrow, A band constant ("HI shrink, A is Always the same")

Mnemonic dArk = Anisotropic; lIght = Isotropic = A band dark and anisotropic, I band light and isotropic

Mnemonic Epi-Peri-Endo = Wrappings from surface to single fibre: epimysium, perimysium, endomysium