Ultrastructural Pathology
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	Adapted from a handout by: Dr. Dave Fritz, DACVP
1	Introduction
2	1. Describe normal features; this allows you to- 2. Name the tissue or cells 3. Use appropriate EM vocabulary
3	Morphologic description- Should include (in the following order): 1. Tissue 2. Pathologic changes 3. Presence and description of infectious agents, loss or normal structures, etc. 4. Severity may be difficult to access
4	**GENERAL CELLULAR ANATOMY**
5	Cell junctions: 1. Tight junction (zonula occludens)=promotes proper flow of nutrients and ions from lumen through cell and out of basolateral membrane. also maintains concentrations between lumen and extracellular fluid. 2. Intermediate junction=Intercellular space is wider than normal. This area contains gap junctions, which allow intercellular communication and desmosomes. Spot desmosomes (macular adherens) anchor cell to cell, hemidesmosomes anchor **to basal lamina, and belt (zonula adherens) desmosomes join the terminal web.**
6	Note cell junctions and membrane interdigitations (another method of cellular attachment).
7	Close-up of cell attachments, note tonofilaments.
8	Adhesion plagues also found on mesothelium, lymphocytes, macrophages, etc. desmosomes between macrophages in a fathead minnow with mycobacteriosis.
9	Microvilli with delimiting membrane and parallel sheaves of actin filaments. These filaments insert into the myosin filaments of the terminal web. These are not motile but can shorten.
10	TEM of microvilli in longitudinal and cross-section. Contrast to cilia.
11	Cilia from bronchus. Axonemes or substructure of microtubules is composed of 9 outer dimers and 2 inner singlets. The basal body has 9 outer triplets and no inner tubules. Microtubules are 23 nm in diameter.
12	Nucleus with double nuclear membrane (continuous with surrounding rER), nuclear pores and nucleolus. Heterochromatin is unused and junk chromatin which is connected to the proteins in the nuclear membrane by lamins. Nuclear pores are used to pass assembled ribosomes and messenger RNA and are ATP-dependent.
13	Nucleolus composed of filamentous portion (rRNA transcription), granular portion (assembly of preribosomal components), and the pars amorpha (lucent area).
14	Mitochondria: double membrane, cristae (site of the cytochrome chains), matrix (site of anaerobic respiration), and matrix granules. Mitochondria may be descended from primitive eukaryotes as they possess their own DNA, can replicate, and synthesize some of their own proteins. Their membranes also have pores to allow passage of small molecules.
15	Shape of cristae varies with cell type. Cholesterol metabolizing cells (i.e. Adrenal cortical cells) have vesicular or tubular cristae. Tissues with high oxidative demands have plush, long cristae (skeletal muscle) compared to those with low

	oxidative demands (hepatocytes). These mitochondria with vesicular cristae are from neoplastic adrenal cortical cells. Mitochondria tend to aggregate at the sites in the cell where energy requirements are the highest.
16	Rough endoplasmic reticulum in parallel arrays. Inner cisternae and outer membranes studded with 20-25 nm ribosomes. Cells that produce abundant protein have large amounts of rER (plasma cells, gastric chief cells, pancreatic cells, etc.).
17	Smooth endoplasmic reticulum is tangled and disorganized. This photo is from a mouse being given phenobarbital treatment. SER is the site of the p450 cytochromes and is associated with steroid synthesis and detoxification processes.
18	Golgi apparatus: Composed of stacked arrays of c-shaped cisternae with a concave surface facing the nucleus, and often in close association with RER. The convex (cis) or forming face of the golgi coalesce with transitional vesicles containing proteins from the rER. The proteins pass through the stacks toward the concave (trans) or maturing face in coated vesicles specific to each stack. They are ultimately glycosylated and then released in secretory vesicles.
19	Two types of lysosomes are identifiable due to contents. Primary lysosomes are homogenous, while secondary lysosomes have dark residual bodies. In hepatocytes, peroxisomes, which participate in lipid metabolism and the detoxification of substances such as phenols and alcohol) look much like primary lysosomes but may have a darker core.
20	Autophagosome
21	EM of liver with glycogen particles. Alpha particles (rosette formation of 80-100 nm) and beta particles (monoparticulate form of 10-30 nm). Similar in size to ribosomes. Glycogen not well preserved in EM processing. Note mitochondria with few, short cristae (generally seen in inert mitochondria) and lysosome with single membrane.
22	Lipid vacuole in hepatocyte. Usually <u>not</u> membrane bound. Osmium may precipitate around periphery (as this case). Lipid is usually dense, routine fixation will wash lipid out leaving a lucent area.
23	Collagen with periodicity of 64 nm. Note the normal variation in diameter in cross-section.
24	Fibrin with periodicity of 23 nm. Difficult to see in routine preps – usually just appears in mats.
25	**CELL INJURY**
26	Normal cell with cilium, microvilli, tight junction, organelles, infolding of basal Membrane with interposed mitochondria.
27	Mild swelling of cisternae of ER and perinuclear space.
28	EM of dilated ER and perinuclear space (carbon tetrachloride intoxication).
29	Membrane appendages lost with blebbing of membrane into lumen. Swelling of ER, perinuclear space, and mitochondria with condensation of matrix, and loss of Organelle polarity. Margination of nuclear chromatin.
30	EM with swollen mitochondria with condensed matrix and dilated ER with free ribosomes. (The swelling of the cristae results in the condensation of the matrix.)
31	Chromatinolysis, swelling of golgi apparatus, high amplitude swelling of

	Mitochondria and formation of membranous whorls around these mitochondria.
32	EM of high amplitude mitochondrial swelling and loss of membrane integrity. All of the organelles swell due to fluid imbibition in the organelles.
33	Loss of membrane integrity (plasma and nuclear), high amplitude swelling of All mitochondria with calcium densities, and release of cell from basal lamina.
34	Degeneration/necrosis vs. apoptosis. Swelling vs. shrinking, degenerate Organelles vs. normal organelles, disrupted lytic chromatin vs. marginated, Condensed chromatin crescents, membrane lysis vs. membrane bound. The FAS ligand, TNF-alpha, and lymphotoxin are all potent activators of apoptosis.
35	Apoptosis in mouse cell. The formation of crescents in the nucleus is characteristic.
36	Phagocytized apoptotic bodies in rat prostate carcinoma. Apoptotic bodies are often found in macrophages.
37	Yellowstone River, MT
38	**GASTROINTESTINAL**
39	Gastric parietal cell. Produces HCL, note secretory canaliculi, extensive Microvilli, abundant mitochondria and extensive tubuloreticular system. Note the presence of the mitochondria around the canaliculi - the active transport of hydrogen into the canalicular lumen is very energy intensive.
40	Gastric chief cells produce pepsinogen. Note parallel arrays of abundant rER, zymogen granules, and lots of golgi. This is common appearance for all zymogenic cells.
41	Mucous neck cells (before and after acetylcholine treatment).
42	Apical surface of enterocyte from rat after fatty meal. Many fat droplets in Cisternae of sER in transport to lacteals. Note terminal web and plush microvilli of Even length (contrast to renal tubular epithelium).
43	Mouse peyer's patch. Note almost squamous M cell with few microvilli overlying lymphocytes and a macrophage.
44	Cecal epithelium from a chicken. Shorter microvilli with pronounced actin rootlets and terminal web. The colon of most species has a pronounced terminal web.
45	Pancreatic acinus. Note stubby microvilli along canaliculi and zymogenic cells (lots of granules and golgi.).
46	Question 1
47	The Alaska Range, AK
48	**LIVER**
49	Sinusoidal arrangement of hepatocytes. Hepatocyte have a round, central nucleus, rER, abundant SER, and numerous mitochondria.
50	Closeup of space of Disse. Hepatocytes have long, arborizing microvilli which extend into this space. The endothelial cell is attached to a reticulin sheet (like a basal lamina).
51	Hepatocytes and bile canaliculus. Note tight junction and stubby microvilli. Bile canaliculi are the most reliable identifying feature of hepatocytes on EM.
52	Bile duct epithelium with stubby microvilli and scant ER and mitochondria.
53	Triangular Kupffer cell.

54	Question 2
55	**URINARY SYSTEM**
56	SEM of glomerular capillary loops with podocytes and their processes.
57	Glomerular capillary loop with RBC'c, fenestrated endothelium, basal lamina, podocyte foot processes. Note mesangial cell.
58	High mag with endothelial cell on basal lamina and foot processes with filtration pores.
59	Glomerular deposits from 8th edition of Robbins and Cotran
60	EM of membranous glomerulonephropathy.
61	Canine Lyme disease. Fusion of foot processes, widening of glomerular basal lamina by flocculent material, plexiform splitting of basal lamina, subendothelial dense deposits, and endothelial cytoplasmic vacuoles.
62	Cheetah with uniform thickening of basal lamina by fibrillar material and fusion of foot processes.
63	Note mesangial processes between endothelial cells and basal lamina.
64	Proximal convoluted tubules. Note tall columnar cells with plush, uneven microvilli (as opposed to intestine, which has even microvilli), and basal oriented mitochondria. The microvilli provide 20x surface area for resorption of protein and glucose.
65	High mag of proximal tubular epithelium. Note elongated, basal mitochondria (required to run the NA/K pump) apical pinocytotic vesicles and lysosomes (for protein resorbtion and breakdown).
66	The loop of Henle has nearly squamous epithelial cells and lack mitochondria. and well-developed brush border. The lack of mitochondria is predictable as the LH has no active transport going on (all it does is set up an osmotic gradient in the kidney).
67	Question 3
68	Rock Creek, MT
69	**RESPIRATORY SYSTEM**
70	Tracheal or bronchial epithelium. Membrane delimited axoneme with basal Body. Numerous mitochondria to power cilia. Note microvilli between cilia.
71	Alveolus with capillaries and endothelial cells, and type I and II pneumocytes. The squamous processes of Type 1 pneumocytes cover the vast majority of the air diffusion area of the alveolus and are extremely thin since gaseous exchange must occur through them.
72	Alveolar capillary lined by endothelial cell and processes of type I pneumocytes. There is a thin and thick side of the septum. On the thick side the pneumocyte and endothelial cell rest on separate basal laminae. This collagen is what gives the septum strength.
73	Type II pneumocytes are less numerous that type I cells and have few stubby

	microvilli. The cytoplasm contains numerous multilamellar bodies composed of surfactant arranged in parallel stacks (The identifying feature of Type II pneumoncytes.). May be washed out in routine preps. Also contains multivesiculate bodies (which move the lipids around the cells.)
74	Question 4
75	Big Hole River, MT
76	**ENDOCRINE SYSTEM**
77	Pancreatic islet cells. Alpha cells have round, electron dense granules with a Variable halo then delimiting membrane. Beta cells may have granules like alpha Cells but characteristically have crystalloid, bar-shaped or pleomorphic dense cores.
78	Pancreatic cells from a vole. Delta cells have granules similar to alpha cells.
79	Thyroid follicular epithelium. Stubby apical microvilli, dense bodies (lysosomes Containing acid phosphatase which fuse with colloid droplets), and residual bodies (material left after hormone release).
80	Parathyroid chief cell with few, dense secretory granules, golgi, and rod-shaped mitochondria. Upper right shows oxyphil (nonsecretory) cell with central nucleus and abundant mitochondria.
81	Adrenal cortical cell with mitochondria containing vesicular cristae, lipid vacuoles, and abundant sER. Note lipofuscin.
82	Adrenal medulla. Adrenalin producing cells have garden-variety granules. Norepinephrine producing cells have granules with eccentrically located dense core.
83	East Fork of the Bitterroot River, MT
84	**NERVOUS SYSTEM**
85	Neuron with abundant mitochondria, golgi, and nissl substance composed or rER and free ribosomes.
86	Cross section through multiple unmyelinated nerves with a single Schwann cell nucleus Mitochondria, microtubules and neurofilaments are visible within the axoplasm of nerves.
87	Diagram of myelin acquisition through compaction of Schwann cell or oligodendroglial cell plasma membrane.
88	Oligodendrocytes perform myelination duties in the central nervous systems, servicing up to 50 axons.
89	Myelin sheath around axon. There is no Schwann cell cytoplasm within the myelin sheath, only the close apposition of cytoplasmic membranes.
90	Ependymal cell with occasional microvilli and cilia (use it to move CSF around).
91	Question 5
92	**CARDIOVASCULAR SYSTEM**
93	Sarcomere with Z band, I band, and A band.
94	Cardiac myocyte with sarcoplasmic reticulum surrounding myofibril. T tubules communicate with the extracellular space.
95	Note branching of myofibrils and large numbers of robust mitochondria aligned in

	columns between myofibrils. Intercalated discs bind cells, transmit forces of contraction, and provide areas of conduction. Tight junctions, desmosomes, and gap nexi are all part of the intercalated disk.
96	Pancreatic capillary
97	Smooth muscle cells have centralized nuclei, low numbers of mitochondria and other organelles. Dense bodies, which anchor microfilaments, are scattered throughout the cytoplasm. Flask shaped invaginations called caveolae at the cell membrane are likely analogous to the T-tubule system of skeletal muscle.
98	Question 6
99	
100	**MUSCULOSKELETAL SYSTEM**
101	Similar to cardiac muscle. Myosin filaments extend entire distance of A band and actin from Z band to edge of H band. Note well developed sarcoplasmic reticulum which is continuous with the sarcolemma.
102	Osteocyte. Bone has canaliculus into which a cell process extends. Lucent, granular material is osteoid. Cell contains a centriole, abundant ER, and golgi.
103	Chondrocyte. Note granular, moderately electron dense proteoglycan matrix. Abundant glycogen in cytoplasm due to low oxygen tension in cartilage.
104	Question 6
105	**Hematolymphatic**
106	Neutrophil from a sheep. Larger primary granules are more electron dense than secondary granules and comprise about 20% of total granules.
107	Migration of neutrophil between two endothelial cells.
108	Eosinophil from a rat. Bilobed nucleus with electron dense crystalloid material in some granules (thought to be major basic protein). Histaminase surrounds MBP in these granules.
109	Basophil from a sheep. Similar to mast cell with exception of segmented nucleus and fewer granules, which contain hepatin, histamine, SRS-A and ECF-A.
110	Macrophage from a rabbit. Abundant cytoplasm with long appendages, numerous lysosomes and reniform nucleus.
111	Lymphocyte from a sheep. Scant cytoplasm with few organelles.
112	Plasma cell from a sheep. Central round nucleus with marginated, clumped chromatin, abundant rER and golgi.
113	Russell body with crystal-like immunoglobins within ER.
114	Mast cells
115	Scroll-like granules in human mast cell (can be seen in some animal mast cells).
116	Megakaryocyte. Cleavage lines, dense granules, and nearly formed platelets.
117	Platelet from a mouse. Alpha granules (contain Platelet-Factor-4 and beta-thromboglobulin) and variety of normal organelles (ER in this case). In platelets, you usually see normal organelles, but no nuclei. Dense granules contain serotonin absorbed from the plasma. Will significantly deform and grow pseudopods during the release reaction.

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118	Montana with Rainie
119	**Infectious Agents**
120	**Viruses**
121	Eastern equine encephalitis (Togaviridae genus alphavirus), mouse brain. 20 nm virions stud the ER of neuron. Note larger virion in extracellular space. Togaviridae enter cells via coated pits, and the virus modifies lysosomes into cytoplasmic vacuoles where new viral proteins and nucleocapsids are made.
122	Corona virus in ER. Coronaviridae enter the cell by phagocytosis, and use lysosomes and ER for intracellular replication, forming large vacuoles in which they are characteristically seen.
123	Mandibular salivary gland, skunk. Microfilament aggregates and acinar lumen filled with virions.
124	Liver from chimpanzee. Adenovirus, 70 nm, in intranuclear paracrystalline array
125	Canine distemper virus – note the paracrystalline array.
126	Canine urinary epithelium. Cytoplasmic distemper (Morbillivirus) inclusion and free adenovirus in nuclei.
127	Feline herpesvirus, lung. Note immature virions in nucleus and intracellular mature virions with envelope. 150 nm.
128	Canine cutaneous papillomavirus. Intranuclear paracrystalline array, 45 nm virions. Note keratin filament inclusion.
129	Poxvirus in dermal fibroblast of a squirrel. 250x200 nm virions with cytoplasmic Inclusion.
130	**Bacteria**
131	Rabbit ileum. Note loss of microvilli, pedestal formation, swollen ER, and Karyorrhexis.
132	Turkey trachea, Bordetella avium. Bacteria among cilia.
133	Bronchial epithelial cell, mouse. CAR bacillus among cilia.
134	Helicobacter felis in stomach. Periplasmic fibrils located on gastric mucosa.
135	Pulmonary alveolar macrophage from African green monkey, Yersinia pestis. Immunogold labeled.
136	Intestinal chlamydiosis pig. Elementary and reticulate bodies with Occasional intermediate body. The elementary body is the hardy, invasive form; the reticulate bodies are the stage that synthesizes RNA and DNA.
137	Leptospirosis in proximal tubule, pig.
138	**PROTOZOAL DISEASES**
139	Diagram of apicomplexan zoite. Note Conoid, microtubules, rhoptry, and nucleus.
140	Micrograph of apicomplexan zoite. Note Conoid, microtubules, rhoptry, and nucleus. It is rare to see all of the structures in a single zoite due to sectioning.
141	Toxoplasma bradycyst in cardiac muscle of opossum. Note cyst wall.
142	Toxoplasma schizont in enterocyte of kitten.
143	Gametes of Toxoplasma in the enterocytes of a kitten. Note axonemes in flagella of developing microgametes. Macrogamete (right): Note central nucleus and numerous vacuoles containing variable amounts of medium electron dense material which will form the hard wall of the oocyst.

144	Sarcocystis zoites free in cytoplasm of host cell.
145	Hamster ileum with cryptosporidium and Lawsonia intracellulare in apical cytoplasm of enterocytes. Remember cell changes when you describe.
146	Cutaneous leishmaniasis in a horse. Pyogranulomatous inflammation with organisms in macrophages, many contain viable kinetoplasts. Kinetoplasts act as mitochondria and contain DNA.
147	Textbooks for EM