**Cell Pathology**

**Cell injury** is depicted by structural and/or biochemical changes of a cell. Such changes cause deviation from the cell’s homeostatic conditions. Cell injury may either be reversible or irreversible.

**Reversible cell injury** is an injurious state of a cell whereby its function may be altered, but such alterations do not render the cell incapable of returning to its normal status. These changes are typically mild and pose no dire threat to the cell’s existence. Contrastingly, **irreversible cell injury** is seen when such changes are drastic enough to deter any recovery from the said adverse influences.

Causes of cell injury include, but are not limited to: *overabundance of normal stimuli* (in either amount or duration); *toxic effects* from either **endogenous** or **exogenous** substances; *hypoxia* or *anoxia* and lack of vital nutrients for cell survival.

* **Exogenous** **causes** of cell injury include: physical abuse, chemical or biological factors; toxins and drugs, and microbes.
* **Endogenous** **causes** of cell injury include: genetic anomalies, metabolic deficiencies and metabolites, hormones, cytokines, and other substances rendered by the cell.

**Hypoxia** is a when a cell is not receiving the appropriate amount of oxygen. Total oxygen starvation is termed, **anoxia**. *Hypoxic conditions may lead to anoxia*. Nevertheless, when hypoxic conditions are present it is either based on the lack of enough oxygen or there is an increased demand for oxygen that cannot be met by the cell’s innate processes.

*Causes of* hypoxia or anoxia are as follows: lessened amount of air-oxygen concentration, obstruction to airways, decreased oxygenation of lung capillaries, decreased transport of oxygen, lessened perfusion to tissues, and inhibition of oxidative respiration due to blockage of oxygen usage.

Hypoxia can cause cell injury by lessening the amount of oxygen received for oxidative respiration. If there is such a deficit in oxygen, then there will be a decreased amount of ATP production. This loss of cellular energy will result in the stagnation of Na+/K+ ATPase pumps which maintain cellular osmotic homeostasis. Once the concentrations of Na+ and K+ are no longer maintained, there will be an influx of Na+ which will in turn attract water to follow. Essentially, with hypoxia, there will be cellular swelling. Cellular swelling is termed, **hydropic** or **vacuolar** **change** and is usually reversible.

With hydropic change, water will accumulate within the cell membrane and cytosol; these areas of water retention are termed, **hypoxic vacuoles**. Water can also accumulate in mitochondria and the cisterns of RER (swollen mitochondria have decreased energy output and RER that swell lose their ribosomes resulting in cessation of protein synthesis). Thus, hydropic change influences malfunction of many vital structures of the cell.

Another component of cell injury is the increase in **cytoplasmic Ca2+.** Ca2+ will influx from the extracellular fluid and be leaked from mitochondrial compartments and the cisterns of RER. Ca2+ has a distinct role in enzyme activity; therefore, many enzymes are activated with increases in cytoplasmic Ca2+. Enzymes activated include: **lytic-ATPase** (degrades ATP molecules); **phospholipases** (deconstruct membranes); **proteases** (degrade membrane and cytoskeletal proteins); and **endonucleases** (degrade nuclear material, RNA and DNA). Note: all of these changes are initially reversible, but sustained conditions such as these will result in irreversible cell injury.

Compensation to loss of aerobic respiration and ATP production is made by initiating **anaerobic glycolysis**. As one could imagine, since anaerobic glycolysis uses glycogen stores, this form of stored energy will be depleted. Also a drop in pH will be observed because of the production of lactic acid and release of phosphates from ATP and phospholipids. This decreased pH will also contribute to lysosomal activity which may further endanger the cell. Lysosomal enzymes contain NaOH and other acids. **NaOH** *denatures* proteins and **acids** *precipitate* proteins.

There is no definitive answer for when reversible injury becomes irreversible cell injury. However, this transition is gradual and occurs until the cell’s adaptive mechanisms have been exhausted. Fortunately there are observable differences that mark irreversible damage from reversible. The differences are quantitative and are based on the size and number of specific changes that occur. When the *cell membrane ruptures* and *nuclear material dissociates*, the observer can be certain that *irreversible cell injury* has been established.

**Mitochondrial irreversible changes** include: rupture of the double membrane, fragmentation, myelin figure formation, and calcification. Damaged mitochondria are digested by **autophagosomes**.

**Myelin figures** are simply whorls of membranes from damaged cytoplasmic organelles. Myelin figures are seen in neurons in **Tay-Sachs disease** and other inborn errors of metabolism causing membrane damage. Autophagosomes digest myelin figures as they accumulate.

Nuclear changes in cell death come in three stages: pyknosis, karyolysis, and karyorrhexis. **Pyknosis** is the first step in nuclear dissolution whereby chromatin condenses into an abnormal mass. This stage is followed by **karyolysis**, which is described by the lysis of chromatin by endonucleases. The third stage, **karyorrhexis**, is the final stage of nuclear dissolution whereby chromatin fragments and becomes “nuclear dust.”

Clinical Signs of Irreversible Cell Injury

These irreversible changes ultimately cease the cell’s normal functionality. Myocardial cell injury results in loss of myocardium contraction. Motor neuron cell injury show signs of motor paralysis. And cell injury to the Islets of Langerhans results in diabetes.

Diagnostic tools and procedures have been developed to clinically identify such irreversible cell injury. In severe cellular injury, cytoplasmic enzymes are typically released into the blood. Creatine kinase in plasma may point to myocyte or myofiber damage. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are released into the blood when hepatocytes are damaged. Lactate dehydrogenase (LDH) is released from ruptured erythrocytes and many other cells.

Revival of hypoxic cells is possible if adequate oxygen is administered in the appropriate amounts. Sometimes reoxygenation of hypoxic cells can be deleterious if oxygen administration is excessive. This type of injury is termed, **reperfusion injury**. Usually during reperfusion of tissues, oxygen-derived free radicals are formed, and thus will negatively affect healthy cells found in the marginal regions of the hypoxic territory (infarct). Remember too that ischemic injury involves *inflammation*, thus reperfusion causes an increased number of present leukocytes which hasten the inflammatory effects. Essentially, directing leukocytes to an area that does not need leukocytic activity receives it and suffers. Also, antibodies tend to be deposited in ischemic tissue, so when reperfusion occurs, **complement proteins** bind to the deposited antibodies, are activated and hasten injury and inflammation. An **infarct** (irreversible cell injury) is the area that suffered lack of oxygen (hypoxia) due to **ischemia** (decreased perfusion).

Basic causes of cell injury that may lead to irreversible cell injury are as follows: oxygen deprivation, physical, chemical and infectious agents, immunologic reactions, genetic defects, nutritional imbalances, and aging.

Free Radicals

**Free radicals** are atomic or molecular species that have an unpaired valence electron. These entities are highly reactive and can influence adverse changes to cellular components. Once formed, free radicals can propagate and form new free radical species in an autocatalytic sequence of reactions. Some free radicals include: **superoxide**, **hydrogen peroxide**, and **hydroxyl radical**. *Superoxide dismutase* inactivates superoxide. *Catalase* inactivates hydrogen peroxide and *glutathione peroxidase* inactivates hydroxyl radicals. Iron may also be considered a free radical and is associated with the famous **Fenton Reaction**: Fe3+ 🡪 Fe2+ + H2O2 🡪 OH. + OH- + Fe3+

Any of the mentioned free radicals can form during normal cell processes. There are mechanisms that can regulate free radical accumulations, such as natural antioxidants (vitamins E and C) and other protective enzymes. Overwhelming amounts of free radicals will cause deleterious effects (toxic effects) to the cell. Leukocytes produce oxygen radicals and use these reactive molecules as a part of their mechanism for destroying bacteria.

Free radicals can induce damage to vital cellular components. Cell membranes can be destroyed through **lipid peroxidation** caused by free radicals. Thiol bonds in the protein ion pumps are dismantled by free radicals. Enzymes can be deactivated due to free radicals which cause the proteins of the enzyme to be cross-linked or folded irregularly. Lastly, DNA can be broken apart by free radical exposure, which can lead to loss of DNA transcription, and mutations.

**Necrosis** is localized cell death. Definitive cell death (necrosis) is verified by cell membrane rupture and nuclear changes, such as pyknosis, karyolysis, and karyorrhexis. Necrosis is histologically identifiable by eosinophilia, which means the cell is no longer viable.

**Coagulative necrosis** is the most common form of cell death. This form of necrosis is seen in myocardial infarctions and also in kidney and spleen infarcts. Even infracted tumors experience coagulative necrosis. A cell that undergoes this type of necrosis experiences an inactivation of intracellular proteins, including hydrolytic cytoplasmic enzymes. Thus, the outline of the tissue structure is preserved. The necrotic tissue resembles boiled meat and is paler than normal.

**Liquefactive necrosis** is cell death that results in a cellular “soup.” Liquid like tissue is resultant because hydrolytic cytoplasmic enzymes are not affected and are released into the dead cells (seen in **brain infarcts**); therefore, the cell’s contents are degraded into a liquid material. Liquefactive necrosis can also be caused by enzymatic activity from arriving leukocytes via their lysosomal enzymes (seen in **abscesses**). Note: *Aerobic* bacteria cause liquefaction of tissues.

 Examples of Liquefactive Necrosis

* *Brain infarct*: **encephalomalacia** (softening of brain tissue) occurs, and the necrotic tissue is phagocytosed by macrophages. The cavity produced by the phagocytic processes fills with interstitial fluid via diffusion. Thus, the cavity filled with fluid is termed, a **pseudocyst**.
* *Abscess*: usually this is a cavity filled with **pus** – a liquefied tissue filled with dead and dying neutrophils.
* *Wet gangrene*: this condition is basically coagulative necrosis with a superimposed bacterial infection. The tissue becomes liquefied due to the bacterial lytic enzymes.

**Caseous necrosis** is a form of cell death by which the tissues appear to look like cottage cheese. A histological specimen expressing caseous necrosis will show cells that are amorphous, proteinaceous, and finely granular. This type of necrosis is seen in cases of tuberculosis (**tubercle** is formed with calcification centers that can be seen on radiographs) and fungal granulomas.

**Fat necrosis** occurs in fat cells in and surrounding the pancreas, the omentum, and in the wall of the abdominal cavity. If pancreatic cells (seen in **acute pancreatitis**) are damaged this will result in the release of pancreatic enzymes, including pancreatic lipase. The lipase will cause lipolysis of the fat cells. Initially, the fat cells appear gelatinous and soft, but after time they transform into chalky white patches possessing calcium soaps. Histological specimens expressing fat necrosis will show cells with indistinct outlines and will take on a bluish color due to calcium deposition.

**Fibrinoid necrosis** is a type of cell death that involves the histologically represented accumulation of fibrin and other plasma proteins in the walls of blood vessels and glomeruli. These cells appear red in routine H&E stains. This form of necrosis is typically limited to small blood vessels, arterioles, and glomeruli that have been affected by autoimmune diseases (seen in **systemic lupus erythematosus**) or malignant hypertension.

Mechanism of Fibrinoid Necrosis

Damaged blood vessel 🡪 vasoconstriction 🡪 **thrombocytes** to damaged area and become sticky 🡪 formation of “platelet plug” (acts as a screen) 🡪 fibrin protein seals opening 🡪 **plasmin** break down fibrin patch 🡪 endothelial cells reconstitute \*\*\*constant trauma overtime (hypertension) will result in fibrous scarring = evidence of blood vessel injury

**Gummatous necrosis** is the hallmark of syphilis-induced necrosis by *Treponema pallidum* infection.

**Hemorrhagic necrosis** occurs when dead tissues are suffused with *extravasated* erythrocytes. Blood is trapped within a tissue causing infusing of the erythrocytes.

Outcomes of Necrosis

**Complete restitution** is the process by which cells regenerate; dead cells are replaced by *almost* parenchymal cells. Regeneration can only occur in cells that contain **facultative mitotic** capability. Liver and kidney cells are cells with such capabilities.

**Repair** of tissue is sometimes accomplished by replacing dead cells with fibrous tissue or scar (**fibrosis**). Fibrosis commonly occurs after myocardial infarctions whereby dead myocytes are phagocytosed and replaced by a fibrous scar.

**Calcification** can occur in some tissue. Calcium salt deposits are seen in necrotic tissue (seen in **dystrophic calcification**).

**Resorption of necrotic tissue** is seen in brain infarcts. Macrophages remove necrotic tissue debris which causes a cavity formation. Fluid diffuses into this cavity forming a *pseudocyst*.

**Apoptosis** is programmed cell death and is also energy-dependent. It is based on sequential activation of “death genes” and “suicide pathway enzymes.”

Apoptosis is initiated by two mainstream mechanisms. The **extrinsic pathway** is initiated by the activation of death receptors on the surface of the cell membrane. **Tumor necrosis factor** (TNF) and **Fas ligand** are two ligands for these receptors. The bound ligand activates initiator caspases. The **intrinsic pathway** involves the increase of mitochondrial membrane permeability to **proapoptotic molecules**, such as *cytochrome*. These molecules activate **initiator caspases** which directly influence the activation of **executor caspases**. The executor caspases act on enzymes, nucleic acids, and cytoskeletal proteins, thus causing fragmentation of cellular components and ultimately the formation of membrane-bound **apoptotic bodies**. Apoptotic bodies secrete *soluble factors* which attracts macrophages. These bodies are then phagocytosed by macrophages and some neighboring cells (**nonprofessional phagocytes**). Thus, no inflammatory response occurs during apoptosis.

Apoptosis usually affects single cells, whereas necrosis affects large groups of cells or tissues. Necrosis is not a result of genetic influence. Apoptosis is strictly regulated by activation of genes that either causes inhibition or activation of specific enzymes. During necrosis, **oncosis** (cellular swelling) is seen, whereas with apoptosis the cellular components are fragmented and packaged into membrane-bound apoptotic bodies.

If apoptosis does not occur, especially in the development of many organs, malformations may arise. Apoptosis occurs so that **syndactyly** (webbed hands/feet)is not seen in the hands and feet of neonates.

Viruses can induce apoptosis, and is seen commonly in **viral hepatitis**. These apoptotic hepatocytes appear as *anuclear*, round *eosinophilic* bodies. Similar bodies, **Councilman bodies**, are seen in apoptotic liver tissue of patients with yellow fever. Also, human immunodeficiency virus (HIV) kills **CD4+ helper T lymphoctyes** by apoptosis.

Mechanisms of Direct and Indirect Cytopathic Effects

**Directly cytopathic**: enterovirus latches onto cell surface 🡪 viral instructions induce formation of membrane channels 🡪 lysis of cell

* Heavy metals such as lead (Pb) and mercury (Hg) can cause direct cytopathic effects. Hg binds to the sulfhydryl bonds in the membrane proteins and cause degradation of the cell membrane.

**Indirectly cytopathic**: internalization of virus 🡪 phagosome made 🡪 phagolysosome made 🡪 all protein broken down save viral DNA 🡪 excess proteins made left over from virion replication and assembly are embedded into the membrane creating a pore 🡪 injury due to alteration in osmotic homeostasis 🡪 imminent cell death

* **Hepatitis B** has a similar viral mechanism to this. The major cause for indigestion of viral proteins and components is due to the hepatocyte lacking *endonucleases* within its lysosomes. *Neo-antigens* will remain due to the enzyme absence and be inserted into the cell membrane. Read the miscellaneous pathology factoids for complete mechanism of *neo-antigens*.

In **sickle cell anemia**, the hemoglobin molecule is abnormal. Thus, the conformation of the biconvex shape of the erythrocyte is aberrant and sickle shaped. The abnormality of the hemoglobin molecule is due to valine replacing alanine in the 6th position = *hemoglobin-S*. Sickle cells clog movement of erythrocytes, thus the amount of oxygen transported decreases which results in microinfarcts in various tissues.

**Chediak-Higashi syndrome** is a condition where there is a mutation in the *lysosomal trafficking regulator gene*, LYST. This mutation indirectly disallows cellular **bacteriolysis**, an impairment caused by microtubule polymerization, a direct correlate to the mutated LYST gene. **Microtubule polymerization** is a defect in which results in disallowed fusion of lysosomes to phagosomes in leukocytes; therefore, bacteriolysis is impaired.

The Role of Bcl-2 in B-cell Lymphoma

**Bcl-2** is a protein that normally suppresses apoptosis. In the case of **B-cell lymphoma**, the gene for Bcl-2 is overtly expressed in B-cell lymphoma cells which give them “*immortality*.” Such cells accumulate in lymph nodes causing the host to die due to an overwhelming amount of immortal tumor cells.

**Adaptations** can either physiologic or pathologic. The word, adaptation, is used to describe the changes that cells and tissues undergo due to prolonged or increased stimulation, decreased amount of oxygen and nutrients, or chronic injury. Some adaptations may be irreversible, but *most* are reversible.

**Atrophy** is an adaptation and may be *physiologic* or *pathologic*. A decrease in cell size means the cells have atrophied. Sometimes a loss of cells will cause a subsequent decrease in cell size, a process known as **involution**. Involution is typically age-dependent and irreversible.

* **Physiologic atrophy** is demonstrated in atrophy of the uterus after pregnancy, and in atrophy (involution) of the thymus.
* **Pathologic atrophy** is demonstrated by several causes: *disuse*, *denervation*, *lack of trophic hormones*, *ischemia*, and *malnutrition*.

**Hypertrophy** is an increase in cell size. Pure hypertrophy, with no hyperplasia, can only occur in cardiac and skeletal muscle. Since the cells in cardiac muscle are *amitotic*, the only way for myocytes to compensate for increased demands is to increase in size. The factors that cause myocardial gains in size are as follows: mechanical stimuli (increased workload) and vasoactive substances (angiotensin II). **Cardiac dilation** may result, which is the repetitive increase in size that eventually reaches a limit to where the *elastic recoil* does not occur, and thus the pumping mechanism of the heart is lost. Calcium also plays a role, acting as a secondary messenger. Skeletal muscles contain cells that could theoretically divide (**satellite cells**), but usually do not. Thus, with increased demand in skeletal muscle hypertrophy of myofibers will result. During hypertrophy, cellular cytoplasm increases and nuclei enlarge. Cytoplasmic contractile proteins increase and nuclear material (DNA and RNA) also increases to the proportion of the increase in total cell mass. Note: Some genes are expressed in hypertrophic cells that are typically not expressed in normal cells. **Oncogenes** (*c-fos* and *c-jun*) and **fetal genes** (*myosin heavy chain* and *atrial natriuretic factor*) that were once expressed are a few of the activated genes seen with cellular hypertrophy.

Some hypertrophic changes are seen in some organelles. One organelle, the SER, is seen to undergo hypertrophy when it is bombarded with toxins. This bombardment causes an increase in centered **P-450 mixed-oxidase system**, which in turn causes the observed hypertrophy. The increase in SER size also signals that the organism will have an increased “tolerance” to the specific toxin and any other toxins that are similarly neutralized by the SER. The half-life of the toxin is reduced 2-fold when there is hypertrophy of the SER (tolerance).

Also seen in cells of hypertrophic tissue is hyperplasia of mitochondria. In some instances, such as vitamin A deficiencies or liver alcohol disease, the mitochondria will become hypertrophic. Enlarged mitochondria are termed, **megamitochondria**. Megamitochondria are not efficient, and thus they stain purple due to an increase in RNA. **Oncocytomas**, which usually arise as tumors in salivary gland tissue, are observed to have megamitochondria; eosinophilia is seen.

**Hyperplasia** is the increase in cell number which causes an increase in tissue or organ size. Hyperplasia can be both physiologic and pathologic. Examples of **physiologic hyperplasia** are the increased size in the uterus in pregnancy (due to hyperplasia and hypertrophy) and with erythroid bone marrow hyperplasia as seen in high altitudes. **Pathologic hyperplasia** may be the result of excessive hormonal or growth factor stimulation. An example of this would be uterine epithelial proliferation after menstrual cycles, which is excited by *estrogen* and *pituitary hormones*, and inhibited by *progesterone*. Hyperplasia, more often than not, is combined with hypertrophy. This is typically seen in the thickening of an obstructed urinary bladder and the uterus as mentioned.

**Grave’s disease** is a good example of *pathologic* hyperplasia. This disease is an *idiopathic* autoimmune disease. Normally, the *adenohypophysis* will be stimulated by **thyrotropin-releasing hormone** (TRH), which is produced and released from the *hypothalamus*, to release **thyroid-stimulating hormone** (TSH). TSH will reach the thyroid to stimulate its follicular cells to release **thyroid hormone** (TH). Once the action of TH has been rendered effective, the TH will increase to the point that it inhibits release of FSH from the *adenohypophysis*. But with the autoimmune disease, Grave’s disease, TH continues to be produced and released. The culprit in this pathogenesis is **long-acting thyroid stimulator** (LATS) which causes a continual release of TH. LATS is an antibody and is often referred to as **thyroid stimulating immunoglobin** (TSI). This antibody binds to the FSH receptors on follicular cells in the thyroid gland and causes increased TH production. As a result the increased TH causes thyroid hyperplasia (**hyperthyroidism**).

**Metaplasia** is replacement of a mature cell type with another cell type. Basically, the reason for such an adaptation is due to an environmental change that renders a normal cell obsolete for such conditions. A new, better-suited cell type will take its place. Metaplasia is commonly seen in smokers, whereby the bronchial stratified columnar epithelium is replaced with squamous epithelium (**squamous metaplasia**). With *gastroesophageal reflux disease*, highly acidic gastric juices regurgitate into the lower esophagus. The cells undergo **columnar metaplasia** (of the esophagus) so that they are better suited for acidic conditions. These mucus-secreting columnar cells are metamorphosed from stratified squamous epithelium. Note: Metaplasia is usually reversible, but if adverse influences persist metaplasia will be followed by **dyplasia** (abnormal cell production), which will eventually lead to **neoplasia** (tumor formation).

**Cellular Inclusions** are substances that accumulate in the cell. These substances may be exogenous or endogenous. **Exogenous substances** include: carbon particles inhaled from air which accumulate in macrophages (**anthracosis**) or pigments used for tattoos. Endogenous substances include: *lipids* (triacylglycerides accumulate in hepatocytes leading to fatty liver or **hepatic steatosis; foam cells** – macrophages that have accumulated cholesterol due to phagocytosed membranes containing the non-degradable cholesterol); *proteins* (*α1-antitrypsin* accumulation in hepatocytes and plasma cells results in **cytoplasmic globules** and **Russell bodies** (eosinophilic immunoglobins accumulated the RER), respectively); *glycogen* (seen in **congenital glycogenosis**); *lipofuscin or lipochrome* (lipid-rich brown pigment seen in aging processes and chronic diseases; derived from peroxidation of polyunsaturated lipids of subcellular membranes); *hemosiderin* (iron-rich brown pigment that accumulates in the liver and other organs in **congenital hemochromatosis** or in patients who have received multiple transfusions). Causes of increased iron concentrations are as follows: hemorrhage, increased absorption of iron, impaired utilization of iron, and hemolytic anemias.

Bruise Process

Physical injury (bruise) 🡪 hemorrhage 🡪 lysis of erythrocytes 🡪 macrophages phagocytose erythrocytes 🡪 hemoglobin catabolism by lysosomes 🡪 accumulation of heme iron in ferritin micelles (hemosiderin) 🡪 red-blue color from *bilirubin* conversion 🡪 green-blue color from *biliverdin* conversion and iron ions accumulate as golden-yellow

**Melanin** is also a cellular accumulation and is formed in melanocytes. Melanin forms from the reaction: tyrosine + tyrosinase 🡪 dihydroxyphenylalanine. If this substance accumulates in adjacent *basal keratinocytes* and *dermal macrophages*, then the condition is called, **freckles**.

**Mallory bodies**, or alcoholic hyaline, are cellular masses of cytokeratin, **intermediate filament accumulation**, and *ubiquitin* found in hepatocytes and are eosinophilic. Alternatively, **Lewy bodies** are found in the CNS and are also eosinophilic.

**Cholesterolosis** is the accumulation of lipids within macrophages found within the lamina propria of the gallbladder.

**Hereditary hyperlipidemic diseases** produce **xanthomas** (aggregations of *foam cells*).

 *Hepatic steatosis*, or **fatty change**, does not decrease function of hepatocytes unless the accumulations are severe or the hepatocytes are poisoned by CCl4. The #1 cause for fatty liver is alcohol abuse and diabetes accompanied with obesity.

**Nonalcoholic steatohepatitis** is a condition that shows an enlarged yellow liver weighing 1.5-3 times its normal weight. In early stages, vacuoles are seen congregating around the nucleus; in later stages the vacuoles displace the nucleus. Fatty cysts finally accumulate and decrease the proportion of liver parenchyma.