CHAPTER 1
The Interactions between Biomaterials and Tissues
Cell Injury & Regeneration

• Cells constantly adjust structure and function to accommodate alterations in environment, particularly responding to chemical and mechanical stressors.

• Cells attempt to maintain intracellular milieu (normal homeostasis).

• As encountering physiologic stresses or pathologic stimuli, cells can adapt, achieving a new steady state and preserving viability.
The Principal Adaptive Response

- **Hypertrophy**: an increase in size of individual cells
- **Hyperplasia**: an increase in cell number
- **Atrophy**: a decrease in size
- **Metaplasia**: transformation from one mature cell type to another

- There may also be more subtle changes in expression of selected genes that are functionally beneficial but are not necessarily reflected in alteration of morphology.
Injury

• Usually if extracellular stressors recede, cells will revert to prestressed state.
• If stressors persist and cell’s adaptive capability is exceeded, cell injury develops.
• Up to a point, cell injury is reversible, and with normalization of stimulus, cell returns to baseline state, usually no worse for wear.
• With severe and persistent stress, cell suffers irreversible injury and dies.
ex. When heart muscle cells are subjected to persistent increased load (high blood pressure), cells adapts by undergoing hypertrophy to compensate for the higher pressures must pump against.

In periods of prolonged starvation (illness or malignant tumor), all myocytes will undergo atrophy.

The myocytes, subjected to an imbalance between blood supply and energy demand due to an occluded coronary artery (ischemia), may be reversibly injured if the occlusion is incomplete or sufficiently brief; Alternatively, they may undergo irreversible injury (cell death, as in myocardial infarction) following complete or prolonged occlusion.
Cancer

- Certain genetic abnormalities and/or environmental stimuli may trigger abnormal tissue growth that is uncoordinated relative to normal tissues, has lost its responsiveness to normal growth controls, and persists after cessation of the stimuli that initiated it.
- This condition is called neoplasia; in its malignant form, this is more commonly called cancer (chap. 4.7)
Causes of Cell Injury

• **Hypoxia and ischemia:**
  
  – Hypoxia is decreased O₂ supply relative to the needs of a particular tissue.
  
  – Anoxia is the complete absence of oxygen.
  
  – Hypoxia due to diminished blood flow is called **ischemia**.
  
  – Irreversible tissue injury (necrosis) due to ischemia is called **infarction** (경색 as Korean)
  
  – Note that although diminished blood flow will invariably lead to hypoxia, oxygen deficiency can occur in the setting of inadequate tissue perfusion.
• **Chemical injury:**
  – Including of food, toxins, hormones, neurotransmitters, synthetic drugs, environmental pollutants, poisons, ethanol, tobacco, even toxic biomaterials
  – Inducing by one of two general mechanisms:
    • **Combining directly** with a critical molecular component or cellular organelle and thereby inhibiting its normal activity. (chemotherapeutic drugs)
    • Chemicals that are not intrinsically biologically active may be converted to **toxic metabolites** during normal physiologic breakdown. Such modification is usually accomplished by **P-450 mixed function oxidases** in SER of liver, and the most important mechanism is by formation of **free radicals**. (acetaminophen)
• **Biologic agents:**
  - From virus, bacteria to fungi, protozoans, helminths
  - Generally a preferred cells of invasion (tropism)
  - Viruses multiply intracellularly by appropriating host biosynthetic machinery.
  - Cell lysis may occur directly or as a result of the immune system’s recognition and destruction of infected cells.
  - Viruses may compromise the ability of cell to perform its normal functions; worse, transformation to malignant neoplasms.
  - Bacteria have toxic cell wall constituents (endotoxin) and can release variety of exotoxins.
  - Process of eradicating infections can also cause injury.
• **Physical injury:**
  – Result by direct mechanical force (trauma, pressure), temp. extremes (burn, frostbite), electric shock, or ionizing radiation.

• **Genetic defects:**
  – Mutations in cellular proteins can lead to dysfunction and eventually irreparable injury.
  – Congenital defects generally manifest as progressive disorder: including lysosomal storage diseases where progressive accumulation of nondegradable metabolites, eventually causes cell rupture, disorders of muscle (myopathies) due to defective energy synthesis by mitochondria, and sickle cell anemia caused by a mutated hemoglobin that results in stiff deformable red blood cells.
Pathogenesis of Cell Injury

- 2 basic mechanisms of cell injury due to any cause:
- **Oxygen and oxygen-derived free radicals.**
  - Activated oxygen and free radicals are mediators of cell injury and death.
  - The most important one in biological systems are oxygen-derived and include **hydroxyl radicals** (OH\(^*\), from hydrolysis of water, by ionizing radiation), **superoxide radicals** (O\(_2\)^{*-}\)), and **nitric oxide radicals** (NO\(^*\)).
  - Free radicals initiate **autocatalytic** reactions: molecules that react with free radicals are in turn converted into free radicals, further propagating the chain of damage.
• When generated in cell, radicals cause single-strand breaks in DNA, fragment lipids in membranes via lipid peroxidation, and fragment or crosslink proteins leading to accelerated degradation or loss of enzymatic activity.

• Free-radical damage is a pathogenic mechanism in such varied processes as chemical and radiation injury, oxygen and other gaseous toxicity, cellular aging, microbial killing by phagocytic cells, inflammatory damage, and tumor destruction by macrophages.

• Free-radical generation is a normal part of respiration and other routine cellular activities including microbial defense.
• Cells have developed mechanisms to degrade free radicals, which are inherently unstable and generally decay spontaneously.

• Superoxide rapidly breaks down in the presence of water into oxygen and hydrogen peroxide. The rate of decay is significantly increased by the action of superoxide dismutases (SODs).

• Glutathione peroxidase also protect against injury by catalyzing free radical breakdown.

• Catalase directs the degradation of hydrogen peroxide.

• Antioxidant vitamin E may either block free radical formation or scavenge them once they have formed.
Failure of intracellular ion homeostatic mechanisms:

- Cytosolic free calcium is normally maintained by ATP-dependent calcium transporters at extremely low conc. (<0.1 uM); this is in the face of sequestered mitochondria and ER calcium stores.
- Extracellular calcium typically at 1.3mM (about $10^4$ fold gradient)
- Ischemic or toxin-induced injuries allow a net influx of extracellular calcium across plasma membrane, followed by release of calcium from intracellular stores.
- Increased cytosolic calcium activates variety of phospholipases, proteases, ATPases, and endonucleases.
Responses to Cell Injury

• Whether a specific form of stress induces adaptation or causes reversible or irreversible injury depends not only on the nature and severity of the stress, but also on several other cell-specific variables including vulnerability, differentiation, blood supply, nutrition, and previous state of the cell.

  – Cellular response to injurious stimuli depends on the type of injury, its duration, and its severity.
  – Consequence of an injurious stimulus are also dependent on the type of cell being injured, its current state, and its adaptability.
• **ex.**, striated skeletal muscle in the leg can tolerate complete **ischemia** for 2~3 hours without suffering irreversible injury, whereas cardiac muscle will die after only 20~30 minutes, and CNS neurons are dead after 2~3 minutes.

• A **well-nourished** liver can withstand an ischemic or anaerobic challenge **far better** than a liver without any energy reserve.

• Most injury alters the ability of cells to generate energy to run various intracellular housekeeping chores.

• Hypoxia and ischemia are the most common ways that energy production is abated.
4 intracellular systems particularly vulnerable to injury

I. Aerobic respiration (important in generating ATP energy stores that maintain the intracellular ion gradients (by active pumping) and synthetic pathways)

II. Cell membrane integrity (critical to cellular ionic and osmotic homeostasis)

III. Protein synthesis

IV. Integrity of the genetic apparatus
Cell Response in Hypoxia

- As a consequence of **reduced oxygen tension**, the intracellular generation of **ATP** is markedly reduced.

1. Activity of plasma-membrane ATP-driven sodium pump ($\text{Na}^+/\text{K}^+$ ATPase) declines with **accumulation** of intracellular $\text{Na}^+$ and **diffusion** of $\text{K}^+$ out of cell. The net gain of $\text{Na}^+$ solute is accompanied by an **isosmotic gain of water**, producing acute cellular **swelling**.

2. This is further exacerbated by the **increased osmotic load** from the accumulation of **other metabolites**, including inorganic phosphates, lactic acid, and purine nucleosides, as the cell struggles **to generate ATP** via anaerobic pathways. The **cytoplasm** becomes **acidic**.
3. Ribosomes begin to detach from RER and polysomes dissociate into monosomes, with consequent reduction in protein synthesis.

4. Worsening mitochondrial function and increasing membrane permeability cause further morphological deterioration with dispersion of the cytoskeleton and formation of cell surface “blebs”. Organelles and indeed whole cells appear swollen because of loss of osmotic regulation. Precipitation of intracellular proteins and organelles in conjunction with the cellular edema leads to the microscopic appearance of “cloudy swelling”.

Necrosis (괴사)

• If the injurious **stimulus persists**, cell **death** may follow.
• If oxygen is **restored**, all of the above disturbances are potentially **reversible**. However if ischemia **persists**, irreversible injury follows; cells and tissues become necrotic.

• **Two phenomena consistently characterize irreversibility.**
  1) The **inability to reverse** mitochondrial **dysfunction** (lack of oxidative phosphorylation and ATP generation) even upon restoration of oxygen.
  2) The development of profound disturbances in **membrane function**.
• Massive calcium influx into the cell occurs, particularly even if ischemia tissue is reperfused after the point of irreversible injury, with broad activation of calcium-dependent catabolic enzymes.

• Proteins, essential coenzymes, and ribonucleic acids seep out through the newly permeable membranes, and the cells also lose metabolites vital for the reconstitution of ATP.

• Injury to the lysosomal membranes results in leakage of their enzymes into the cytoplasm; the acid hydrolases are activated in the reduced intracellular pH of the ischemic cell and will further degrade cytoplasmic and nuclear components.
Apoptosis (자멸)

• Apoptosis is responsible for the *programmed death* (or cellular *suicide*) in several important physiologic processes;
• The programmed destruction of cells during *embryogenesis*, including implantation, organogenesis, and developmental involution
• *Hormone-dependent physiologic involution*, such as the *endometrium* during the menstrual cycle, or the lactating *breast* after weaning; pathologic atrophy as in the *prostate* after castration.
• Cell deletion in proliferating populations such as *intestinal crypt epithelium*, or cell death in *tumors*. 
• Deletion of autoreactive T cells in the thymus, cell death of cytokine-starved lymphocytes, or cell death induced by cytotoxic T cells.
• Failure of cells to undergo physiologic apoptosis may result in unimpeded tumor proliferation, autoimmune diseases, or aberrant development.
• Apoptotic cells rapidly shrink, form cytoplasmic buds, and fragment into apoptotic bodies composed of membrane-bound vesicles of cytosol and organelles.
• These fragments are quickly extruded, phagocytosed, or degraded by neighboring cells and do not elicit an inflammatory response.
• The **nuclear** changes are due to fragmentation of DNA into **histone-sized pieces** through activation of endonucleases.

• The process may be triggered by **granzymes**, exogenous serine proteases, released by cytoplasmic granules within **cytotoxic T cells and natural killer cells** by activation of intrinsic pathways in embryogenesis or direct radiation injury, or by interaction of a number of related plasma membrane receptors, e.g., tumor necrosis factor (TNF) receptor.

• The **plasma membrane receptors** share an **intracellular death domain protein sequence** that when multimerized, leads to a cascade of enzyme activation culminating in cell death *(Ch.3.3: Fig.10,11).*
Current data suggest that various activators of the apoptosis pathway are ultimately funneled through the synthesis and/or activation of a number of cytosolic proteases. These proteases are termed caspases because they have an active-site cysteine and cleave after aspartic acid residues. Overexpression of any of the caspases will result in cellular apoptosis, suggesting that under normal circumstances, they must be tightly controlled. Increased cytosolic calcium can directly activate some intracellular proteases, and induces “permeability transition” in mitochondria, resulting in caspase-3 activation.
• Initial activation of one or more such enzymes with broad specificity leads to a cascade of activation of other proteases, inexorably culminating in cell suicide.

• **Cell volume and shape** changes are caused by breakdown of the cytoskeleton.

• The general **framework** of cell injury is summarized in *(Ch.3.3: Fig.12)*.

• In the **following chapter**, we will extend the concepts of **structure-function correlation** beyond cell to include the **ECM and complex tissues**, examine what happens following cell & tissue injury, and describe how normal and abnormal tissues are examined.