Lecture 3A:
Components of the Immune System
Innate Immune Response Pathways

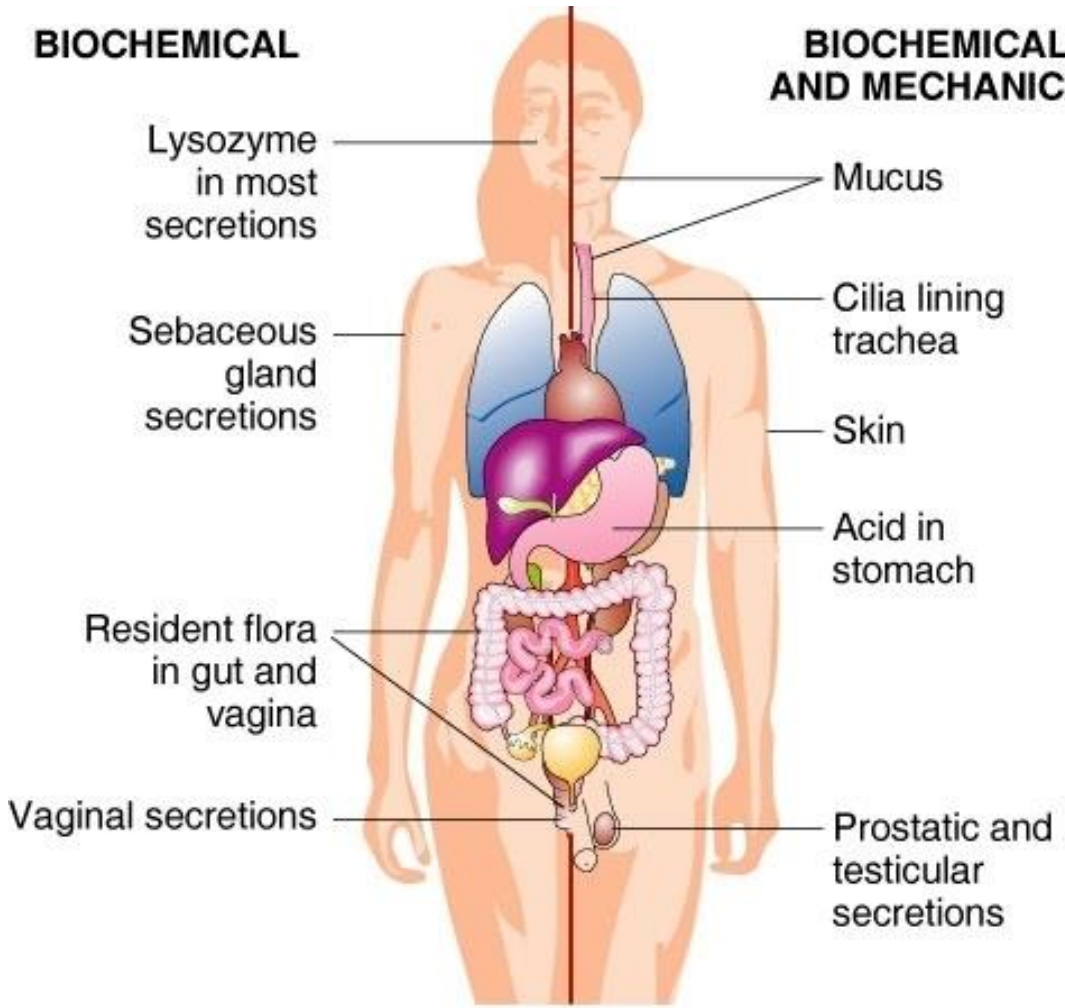
Components of the Immune System

- With such a wide variety of potential pathogens the human body must have a wide variety of defense mechanisms!
- The components of the human immune systems are the:
  - **Skin and Mucous Membrane Barriers**
  - **Red Bone Marrow**
  - **Phagocyte Systems**
  - **Lymphoid System**
- There are two general types of immune responses: innate and adaptive. The two types of responses usually occur simultaneously when an infection occurs.
  - **Innate immune responses** do not require a **specific** antigen trigger.
  - **Adaptive immune responses** do require a **specific** antigen trigger.

Components of the Immune System
Skin and Mucous Membranes

- The **skin and mucous membranes** are referred to as the **"first line of defense"** because they are the site of the first encounter with pathogens and the site of many injuries.
- These surface membranes create not only a **physical barrier** to pathogens, but they also secrete **antimicrobial chemicals**. And resident bacteria make the body surface inhospitable for pathogens. See the image on the next slide.
- If a surface epithelial membrane is penetrated by a pathogen, immune cells in the area will be alerted.
  - Recall that the epidermis of the skin is a thick **avascular** epithelial tissue while the dermis is **vascularized** alveolar connective tissue and **vascularized** dense irregular connective tissue.
  - Similarly, mucous membranes contain **avascular** luminal epithelial tissue with underlying **vascularized** alveolar connective tissue. Mucous membranes line all body tracts.

Components of the Immune System Skin and Mucous Membranes 1



Components of the Immune System: Skin and Mucous Membranes 2

- With very few exceptions, vascularized tissue including the tissue underlying the body surface contains both **blood capillaries and lymphatic capillaries**. Lymphatic capillaries regularly direct excess interstitial fluid (called **lymph** once inside the lymphatic system) through the lymphatic system of vessels and **lymph nodes** back to the blood. The largest lymphatic vessels empty into large veins in the upper chest.
- The vascularized tissue underlying the body surface contains **resident white blood cells such as macrophages** and **dendritic cells**. These cells are usually the first leukocytes to respond to infection or injury. They have membrane receptors for detecting molecules generally associated with pathogens and also for the molecules that are produced by other forms of cellular injury.

Components of the Immune System: Red Bone Marrow

- All blood cells (red and white) as well as platelets are produced in the **red bone marrow** (located in the pelvic bones, ribs, cranium and epiphyses of long bones) by the process of **hematopoiesis.**
  - The **red blood cells** are released immediately into the circulatory system and they stay there. They function to carry oxygen and carbon dioxide between the lungs and the tissues.
  - The **white blood cells** may be released from the bone marrow or remain in the bone marrow until needed. In general they leave the circulatory system and function in the tissues.
- There is just one type of red blood cell, but there are multiple types of leukocytes. White blood cells fall into one of two morphologic categories based on common staining techniques.
  - **Granulocytes**-cells have granules in the cytoplasm.
  - **Agranulocytes**-do not have granules in the cytoplasm.

Components of the Immune System: Red Bone Marrow 1

- **Types of Granulocytes:**
  - **Neutrophils**-most numerous WBCs, phagocytic (especially for bacteria), release inflammatory chemicals, have both red and blue granules.
  - **Eosinophils**-act against parasitic worms and participate in allergic responses, phagocytic, release inflammatory chemicals, have red granules
  - **Basophils**-least numerous WBCs, release inflammatory chemicals, histamine in particular; phagocytic, have blue granules
- Granulocytes participate in **innate immune responses, the "second line of defense"**. These innate immune responses are generalized responses **(NOT antigen-specific**) to infection or cell injury.
- Some granulocytes enter the circulation immediately, but most reside in the bone marrow until they receive a chemical signal to enter the blood. They are chemotactic. They can follow a chemical trail to the site of infection or injury.

Components of the Immune System: Red Bone Marrow 2

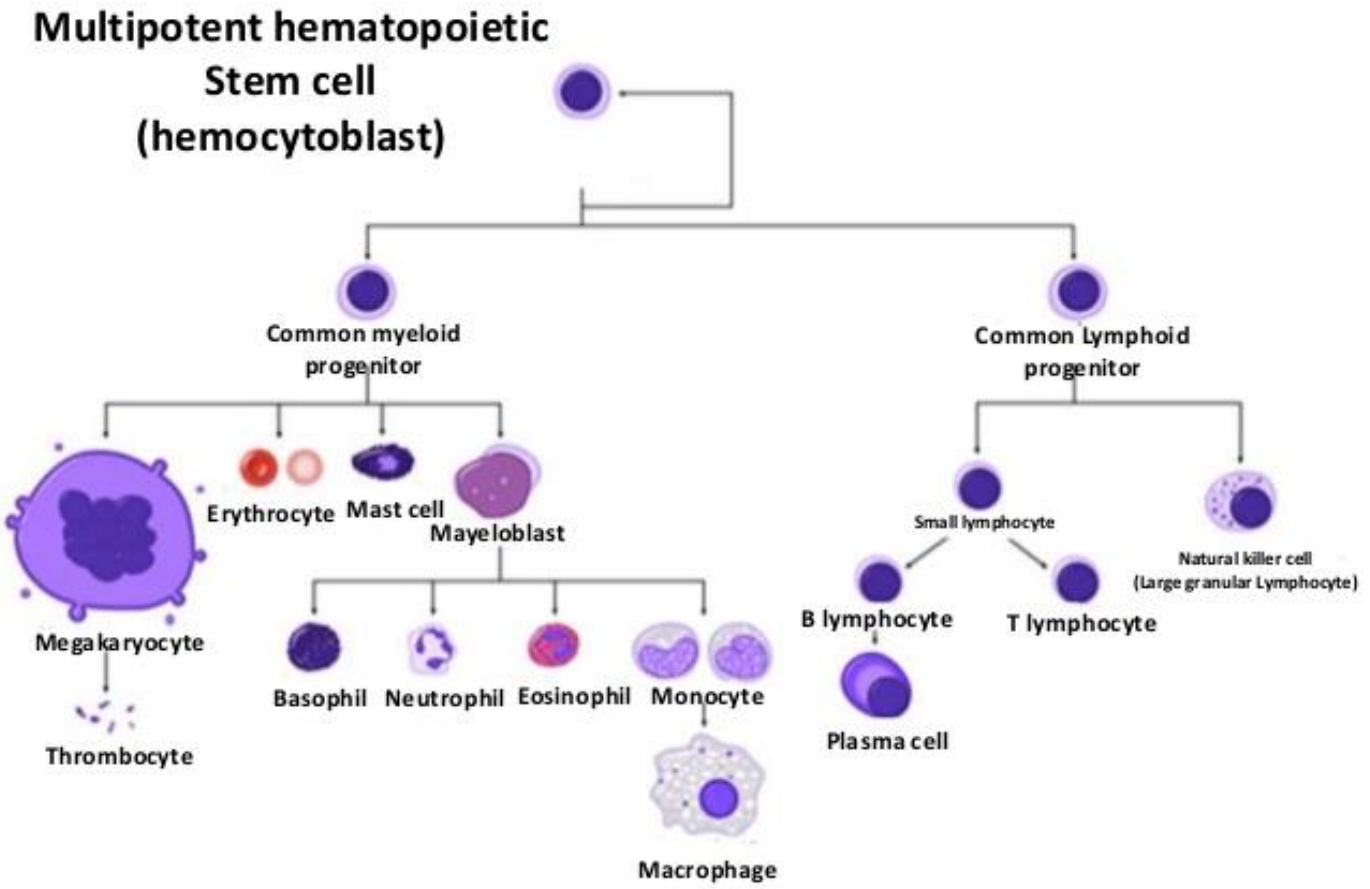
- **Types of Agranulocytes**
  - **Monocytes**-the largest WBCs, act as phagocytes in the blood and enter the tissues to become macrophages or dendritic cells
  - **Lymphocytes-**the smallest WBCs act in adaptive immunity
    - **T cells**-participate in **adaptive immune responses (antigen specific responses),** against infected or malignant cells.
    - **B cells**-participate in **adaptive immune responses (antigen specific responses) against humoral (body fluid) antigens.** B cells differentiate into antibody-secreting cells called **plasma cells**
    - **NK (natural killer) cells**-participate in **innate immune responses (NOT antigen-specific)**, they destroy infected or malignant human cells.

Components of the Immune System: Red Bone Marrow 3

- **Hematopoiesis**
  - The pluripotent stem cell that gives rise to all formed elements in the the red bone marrow is called a **hemocytoblast.**
  - **Two cell line develop from the hemocytoblast:**
    - **Myeloid Cell Line**-gives rise to
      - Erythrocytes
      - Granulocytes
        - Neutrophils
        - Eosinophils
        - Basophils
      - Monocytes
      - Megakaryocytes (produce fragments=platelets)
      - Mast cells
    - **Lymphoid Cell Line**-gives rise to
      - Lymphocytes
      - NK cells

Components of the Immune System: Red Bone Marrow 4

Note the two stem cell lines: myeloid and lymphoid.



Components of the Immune System: Phagocyte Systems

- **Monocytes, Macrophages and Dendritic Cells (Mononuclear Phagocytes. They have spherical nuclei.)**
  - Recognize, via **toll-like receptors (TLRs)** on their surfaces, common (not specific) molecules on pathogen surfaces or in pathogen secretions.
  - **Monocytes** phagocytize pathogens and debris in the blood.
  - **Macrophages** are derived from monocytes. They are present in many body tissues where they major roles in immunity.
    - When resident macrophages first encounter pathogens they not only act to phagocytize the pathogens, they also release **cytokines**. Cytokines are small protein signaling molecules that are secreted by cells with the purpose of **communicating with other cells**. Cytokines are important in **both** the innate and adaptive immune responses.

Components of the Immune System: Phagocyte Systems 1

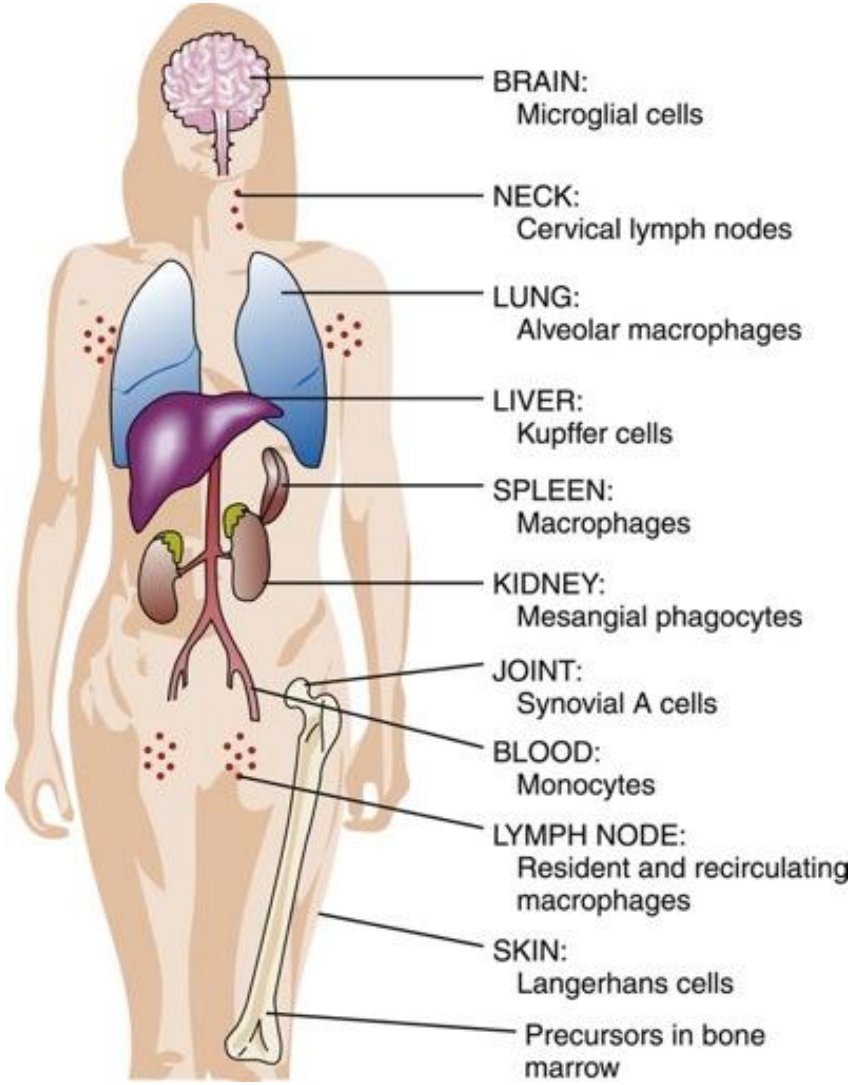
- **Macrophages act as APCs (antigen presenting cells).** They engulf pathogens in the tissues and then display pathogen-derived antigens on their surfaces.
  - They enter lymphatic capillaries and travel in the lymphatic circulation to **lymphoid tissues** where they present the displayed antigens to **lymphocytes**. Antigen presentation is required for the **acquired immune responses.**
- Macrophages have cell surface proteins and secretions that are important in **activating lymphocytes** in the acquired immune response.
- After the immune responses have subsided, macrophages clean up by phagocytizing dead cells and cell debris.

Components of the Immune System Phagocyte Systems 2

- **Dendritic cells** are derived from monocytes during inflammation. They are the body's primary **antigen presenting cells (APCs)**. Like macrophages, they reside in body tissues and they travel to lymphoid tissues to present antigens they have phagocytized. **Langerhans cells** of the skin are examples of dendritic cells.
- **Granulocytes (polymorphonuclear, PMN, phagocytes)**
  - **Polymorphonuclear** refers to cells that don't have spherical nuclei. The nuclei have constrictions that produce lobes.
  - **Neutrophils** leave the bone marrow, enter the blood and follow the cytokine trail back to the infection site.
    - **Neutrophils** are especially active against bacteria. As they phagocytose pathogens, they release very potent **inflammatory chemicals including free radicals.**
- **Eosinophils and basophils** sometimes act as phagocytes.

Components of the Immune System: Phagocyte Systems 3

Body locations of mononuclear phagocytes: monocytes, macrophages and dendritic cells



Components of the Immune System Lymphoid Tissues

- **Primary Lymphoid Tissues**
  - The **primary lymphoid tissues** are the **red bone marrow and the thymus gland**. Those are the locations where mature lymphocytes are released.
  - A key component of B cell and T cell maturation is the development of **"self tolerance"**=lymphocytes must be able to **recognize (bind to)** "self antigens", but must **NOT react (bind too tightly)** to them.
  - B cells mature in the red bone marrow. They migrate from the periphery toward the center of the bone marrow as they mature. Only about 25% of B cells survive the "self tolerance" test. The rest die by apoptosis. **Mature, but naïve B cells** (Naïve because they haven't encounter their specific antigen yet.) leave the bone marrow to reside in the lymph nodes and other lymphoid tissues

Components of the Immune System Lymphoid Tissues 1

- Immature T cells leave the bone marrow and travel to the **thymus gland** where they mature. The thymus gland is located anterior to the heart and is much more prominent in children than adults. It contains no B cells or macrophages, **only T cells**.
- T cells migrate inward from the cortex to the medulla of the thymus gland as they mature. Only about **5%** of them survive the "self-tolerance" selection process! The rest die by apoptosis.
- **Mature, but naïve T cells** leave the thymus gland and reside in the lymph nodes and other lymphoid tissues.

- **Secondary Lymphoid Tissues**
  - Secondary lymphoid tissues: lymph nodes, spleen, tonsils, Peyer's patches. Lymphocytes and macrophages reside in the secondary lymphoid tissues. The lymphocytes in these organs and tissues are mature and self-tolerant, but most are naïve (They have not be exposed to their specific antigen yet.) The secondary lymphoid tissues are the major sites of **antigen presentation** to mature, self-tolerant lymphocytes.

Components of the Immune System Lymphoid Tissues 2

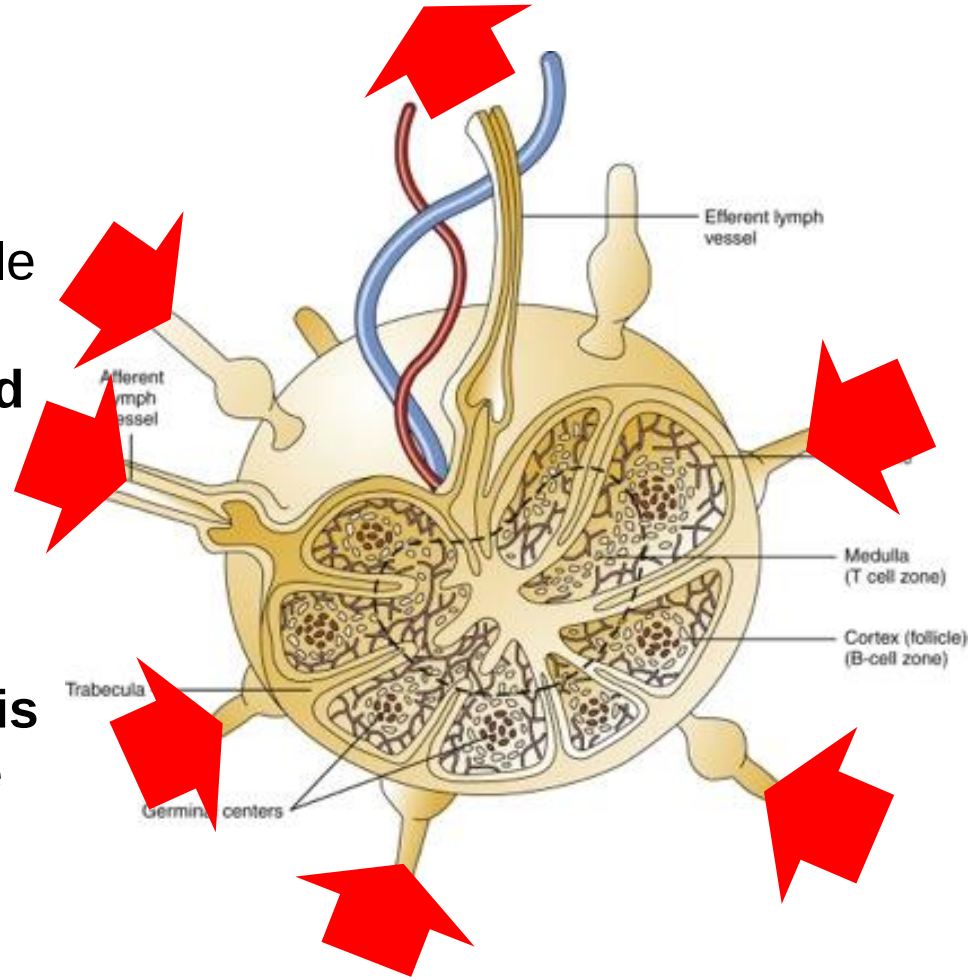
- **Lymph nodes** are located in superficial clusters in the neck, armpit, and groin area. They are also located in the thoracic and abdominal cavities. They contain resident macrophages, T cells and B cells. Lymphatic vessels constantly circulate lymph (Interstitial fluid is called **lymph** while inside the lymphatic system.) through the lymph nodes so that lymph is exposed to immune cells before it is returned to the blood.
- The **spleen**, the largest lymphatic organ, is located just behind the stomach. It is a very vascular organ that serves to expose blood to immune cells. It's function is similar to that of the lymph nodes, except that lymph nodes expose lymph to immune cells, and the spleen exposes blood to immune cells. The spleen is full of **red pulp** porous blood vessels lined with macrophages. The **white pulp** of the spleen consists of small islands of lymphocytes within the red pulp.

Components of the Immune System
Lymphoid Tissues 3

Lymph Node Structure

B cell follicles are located
In the cortex (outer portion) of the node. The germinal center of a follicle is where B cell proliferation **occurs.**
T cells and macrophages are located in the medulla (inner portion) of the node.

There are always more afferent lymphatic vessels than efferent. This slows the flow of lymph through the node.

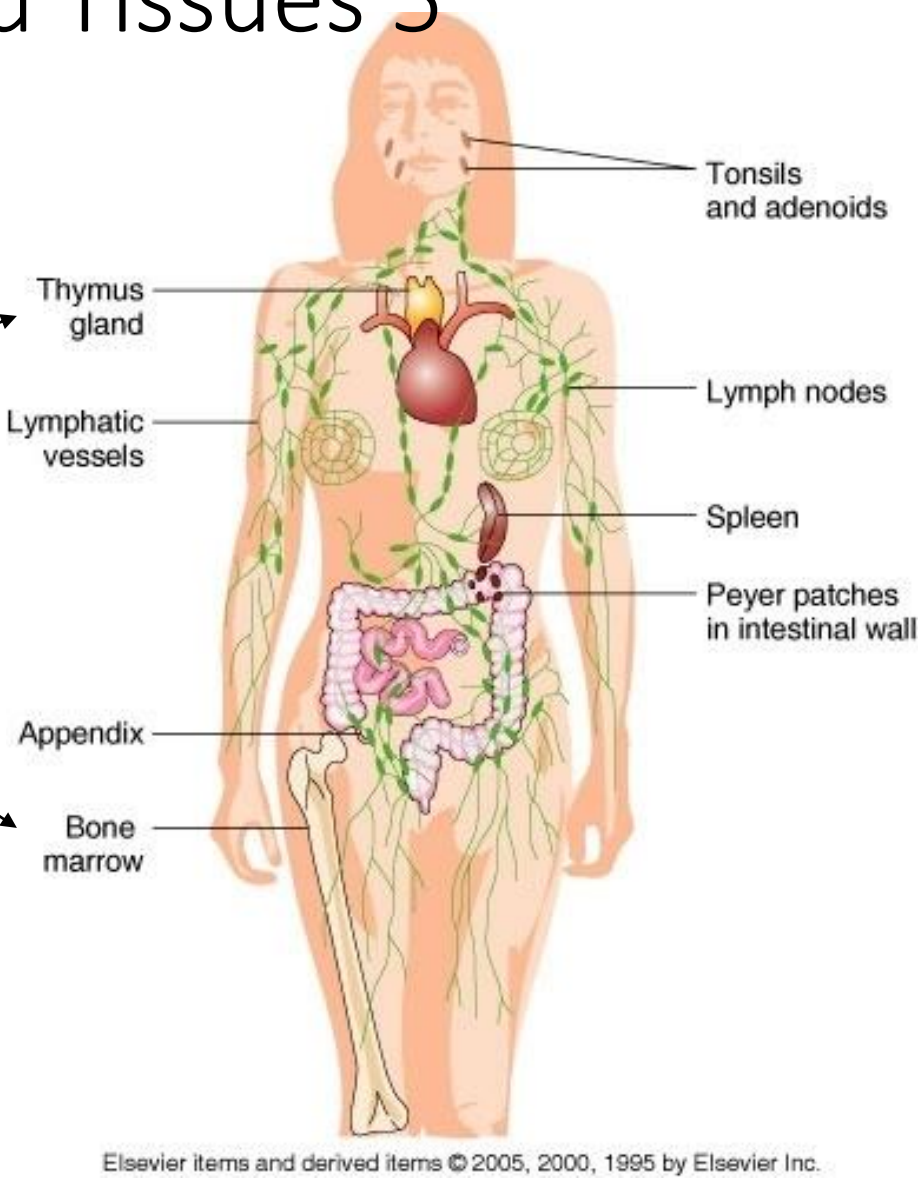


Components of the Immune System Lymphoid Tissues 4

- The **tonsils** are similar in size and structure to the the lymph nodes. They are located in a ring within the mucous membrane lining of the upper pharynx (throat). Being so close to the body surface, they have no lymphatic vessels attached to them. The tonsils have deep **crypts** in their surfaces to allow their leukocytes better access to antigens and pathogens.
- **Peyer's Patches** are patches of lymphoid tissue located in the mucous membrane of the intestines. They are similar to tonsils, and have no lymphatic vessels. Unlike the tonsils they have no crypts.

Components of the Immune System
Lymphoid Tissues 5

Primary Lymphoid Organs



Components of the Immune System Lymphoid Tissues 6

- Most T cells circulate through the lymph and blood and back to the lymphoid tissue once or twice each day in order to obtain maximum exposure to APCs.
- When naïve B cells and/or T cells encounter their specific antigens, they become **activated and undergo clonal expansion** (They proliferate at a high rate by mitosis.). The lymph nodes become swollen and tender. **Activated T cells** leave the lymph nodes and enter the circulation to seek out more of their specific antigen. **Activated B cells** tend to stay in the lymph nodes where they differentiate into **plasma cells** that produce antibodies (protein molecules) against the specific antigen. The antibodies are released into the circulation.
- Recall that **differentiation** involves turning genes on and off to support cell function. A B cell and the plasma cell it becomes are genetically identical, but each cell type expresses a different set of genes.

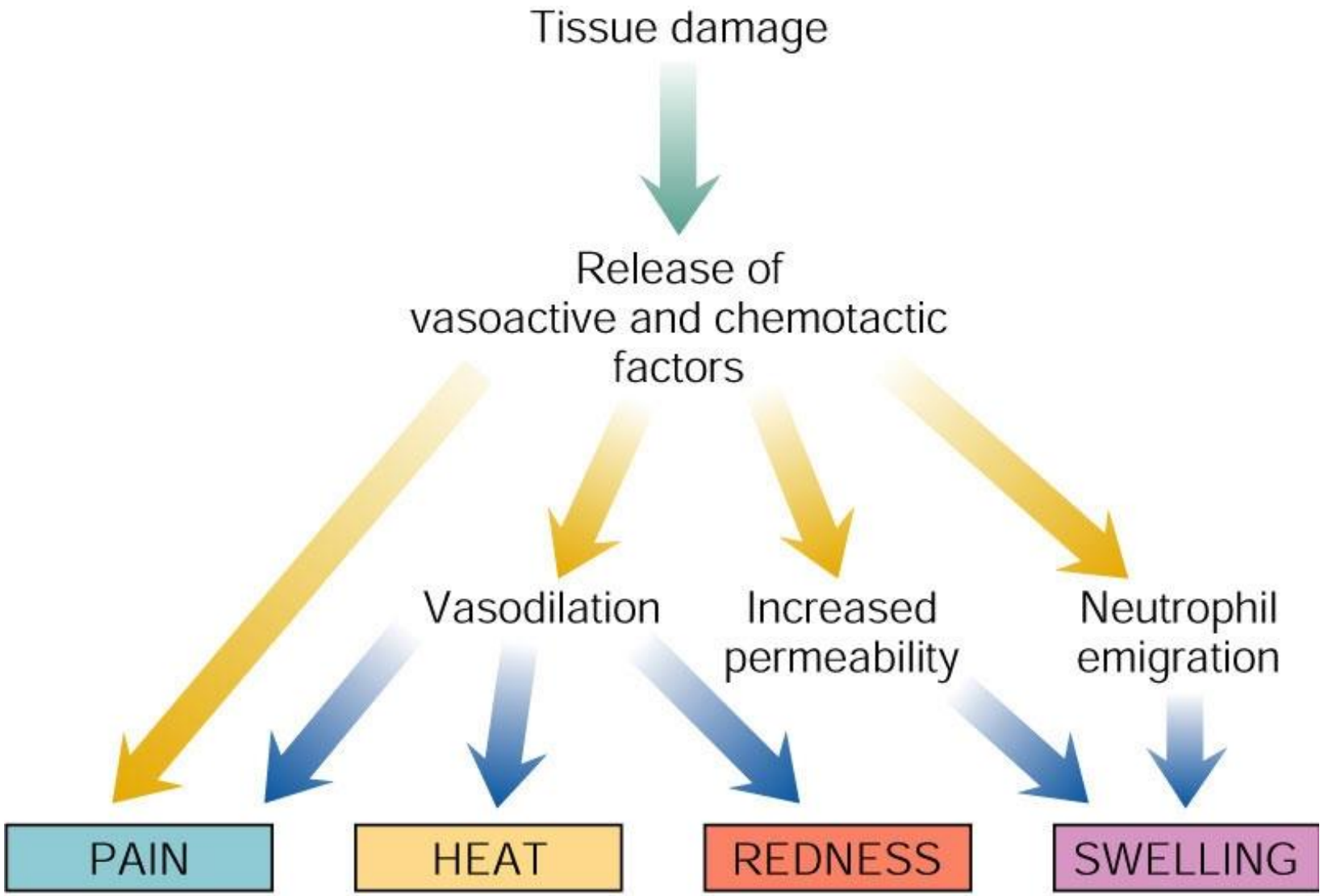
The Innate Immune Response

- **Inflammation (the inflammatory response)** is triggered when body tissues are injured by trauma or infection. It is **an innate immune response**. It does not require exposure to a specific antigen. It does not involve lymphocytes. It has several beneficial effects.
  - It prevents the spread of damaging agents to nearby tissues.
  - It disposes of cell debris and pathogens.
  - It alerts cells of the adaptive immune response.
  - It sets the stage for repair.
- BUT inflammation causes discomfort. The **Four Cardinal Signs of Inflammation** are:
  - **Redness**
  - **Heat**
  - **Swelling**
  - **Pain**

The Innate Immune Response 1

- The suffix **–itis** is used to describe inflammation of a specific tissue or organ: tendonitis (tendon), appendicitis (appendix), myositis (muscle).
- The Four Cardinal Signs of Inflammation are due to **inflammatory chemicals.** These chemicals:
  - Cause **leukocytosis**, an increased number of WBCs in the bloodstream.
  - Recruit more WBCs to the site by **chemotaxis**. The extra cells cause some of the **swelling.**
  - Cause local **arterioles to dilate** resulting in **hyperemia** (more blood enters local capillary beds). This causes local **redness and heat.**
  - Cause local **capillary walls to become more permeable** (leaky). Protein-rich fluid (exudate) leaks out of capillaries into the interstitial spaces causing local **swelling and pain**.

The Innate Immune Response 2



The Innate Immune Response: Pathways

- The events of inflammation are remarkably the same regardless of the cause.
- **Events of Inflammation**
  - Inflammation begins with the activation of certain **biochemical pathways and certain WBCs.**
  - The whole process starts when chemicals produced by injured cells OR chemicals produced by infective agents (bacteria, viruses, fungi, etc.) bind to very important proteins called **toll-like receptors (TLRs)** on or in host cells.
    - TLRs play a pivotal role in both **innate and adaptive** immune responses as well as the **repair** mechanisms that are associated with tissue injury.
    - Recent research indicates that TLRs also play a role in the development of **cancer**!

The Innate Immune Response: Pathways 1

- The TLRs that are important in initiation of the innate inflammatory response are located on the surface of **endothelial cells** and on the surface of **WBCs** that normally reside in tissues (macrophages, dendritic cells, mast cells).
- When chemicals released by injured cells or invading microbes bind to their TLRs on endothelial cells or resident WBCs, those cells begin producing new secretions, and those secretions activate **biochemical pathways and other WBCs.**

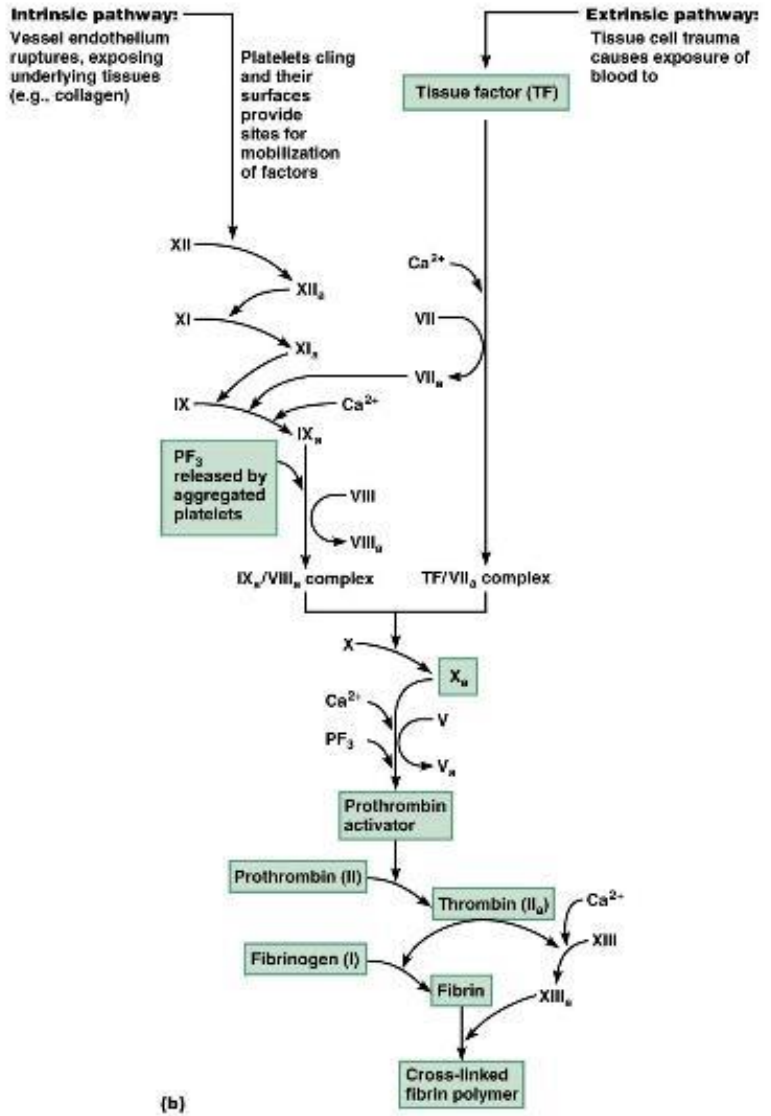
- Immediately after injury, there is a brief period of local **vasoconstriction (vascular spasm)** induced by chemicals released from endothelial cells to assist in reducing any blood loss that may be occurring. This is followed by an extended period of **vasodilation**, bringing increased blood flow to the site of injury.

The Innate Immune Response: Pathways 2

- The **blood coagulation pathway** is initiated either by chemicals released from platelets (**intrinsic pathway**) or from injured tissue cells (**extrinsic pathway**).
  - The insoluble **fibrin protein molecules** produced by the coagulation pathway will be used to form a **blood clot** and also to **block the local lymphatic vessels**. This will prevent spread of injurious chemicals and pathogens.
- The **kinin pathway** and the intrinsic blood coagulation pathway share a common factor (Factor XII). The kinin pathway is activated along with the intrinsic pathway.
  - The end product of the kinin pathway, **bradykinin**, is a vasodilator and potent pain producing chemical.

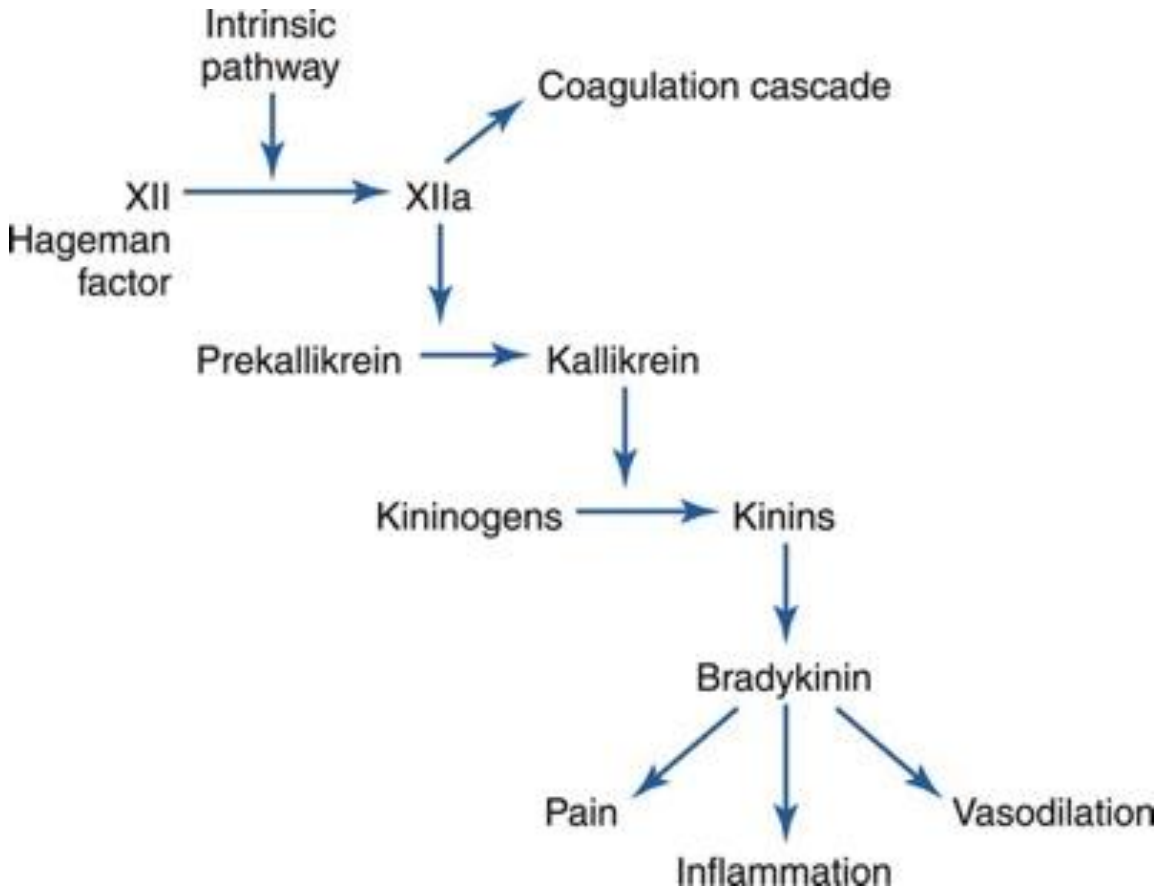
The Innate Immune Response: Pathways 3

The Coagulation Pathway



The Innate Immune Response: Pathways 4

The Kinin Pathway



The Innate Immune Response: Pathways 5

- The **arachidonic acid pathway** is initiated when **phospholipase A2**, an enzyme that releases arachidonic acid (a membrane phospholipid) from cell membranes, is activated in injured cells and also in mast cells at the site. Activation occurs due to **increased intracellular calcium** ions in injured cells. It also occurs as part of platelet activation.
- In general the products of the arachidonic acid pathway effect three properties:
  - Vasodilation/vasoconstriction
  - Chemotaxis stimulation/inhibition
  - Platelet aggregation stimulation/inhibition

The Innate Immune Response: Pathways 6

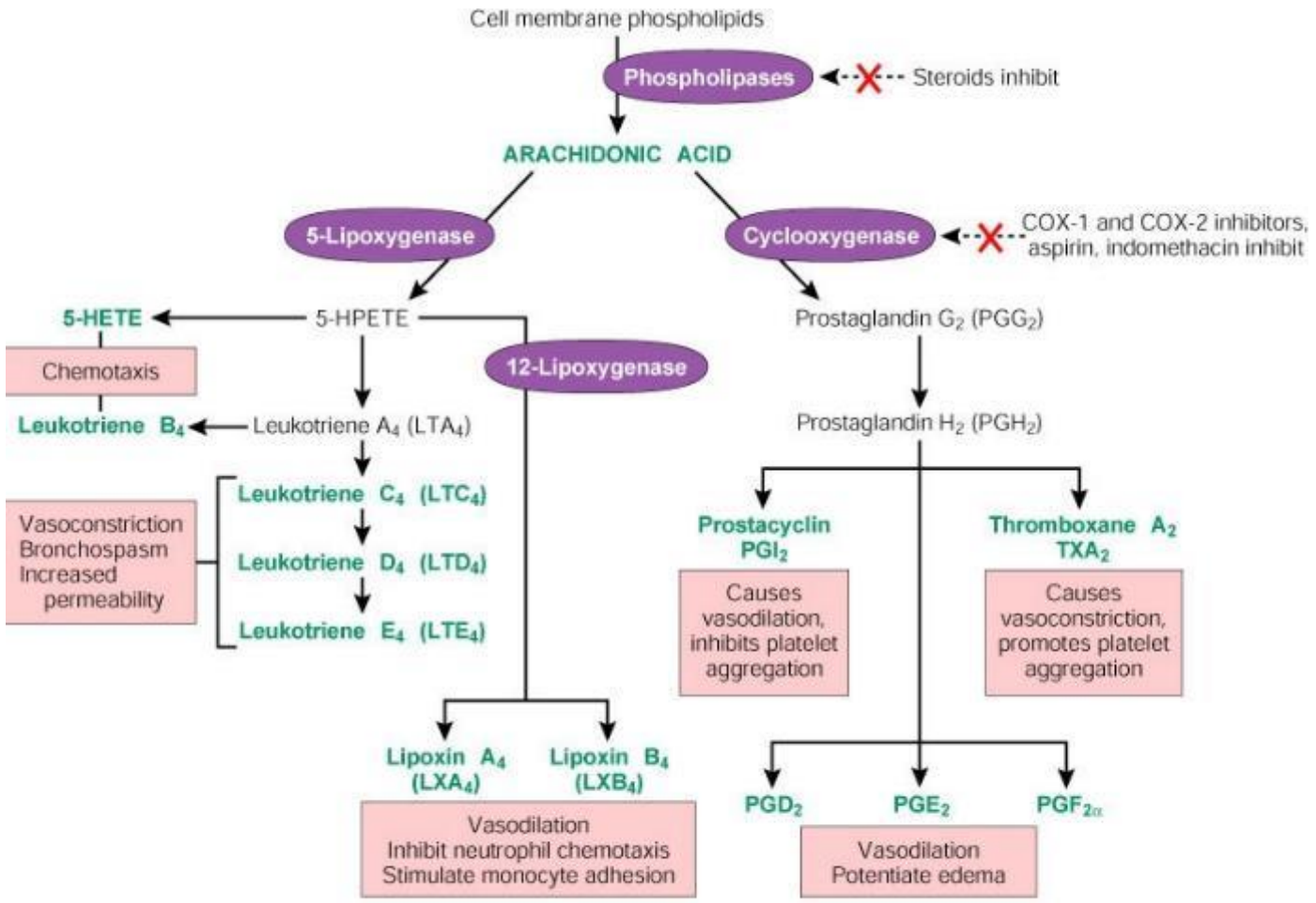
- **Cyclooxygenase enzymes (COX-1 and COX-2) convert arachidonic acid to prostacylin, thromboxane A2 and prostaglandins.**
  - Both prostacyclin and thromboxane A2 have vasoactive properties and mediate the aggregation of platelets.
  - Prostaglandins have many properties.
- Many **anti-inflammatory medications** are aimed at reducing the activity of arachidonic acid pathway enzymes.
  - **Corticosteroids** inhibit **phospholipase.**
  - Aspirin, indomethacin and ibuprofen (and other NSAIDS), inhibit **both COX-1 and COX-2.**

The Innate Immune Response: Pathways 7

- **COX-1** produces body-friendly prostaglandins and thromboxane A2. In particular, the **prostaglandins protect the lining of the digestive tract** by stimulating mucus production and facilitating platelet aggregation. Thus **inhibiting COX-1 causes digestive discomfort** and reduces the ability to form blood clots. Inhibiting unwanted blood clots is a big plus for many people, but digestive discomfort is unpleasant to say the least!
- **COX-2** produces prostaglandins that stimulate inflammation.
- NASIDS that **selectively inhibit only COX-2** have been developed. **Celecoxib** (Celebrex) is an example. A recent study suggests that acetaminophen (Tylenol), previously thought to have a mysterious mechanism of action, is actually a selective inhibitor of COX-2.

The Innate Immune Response: Pathways 8

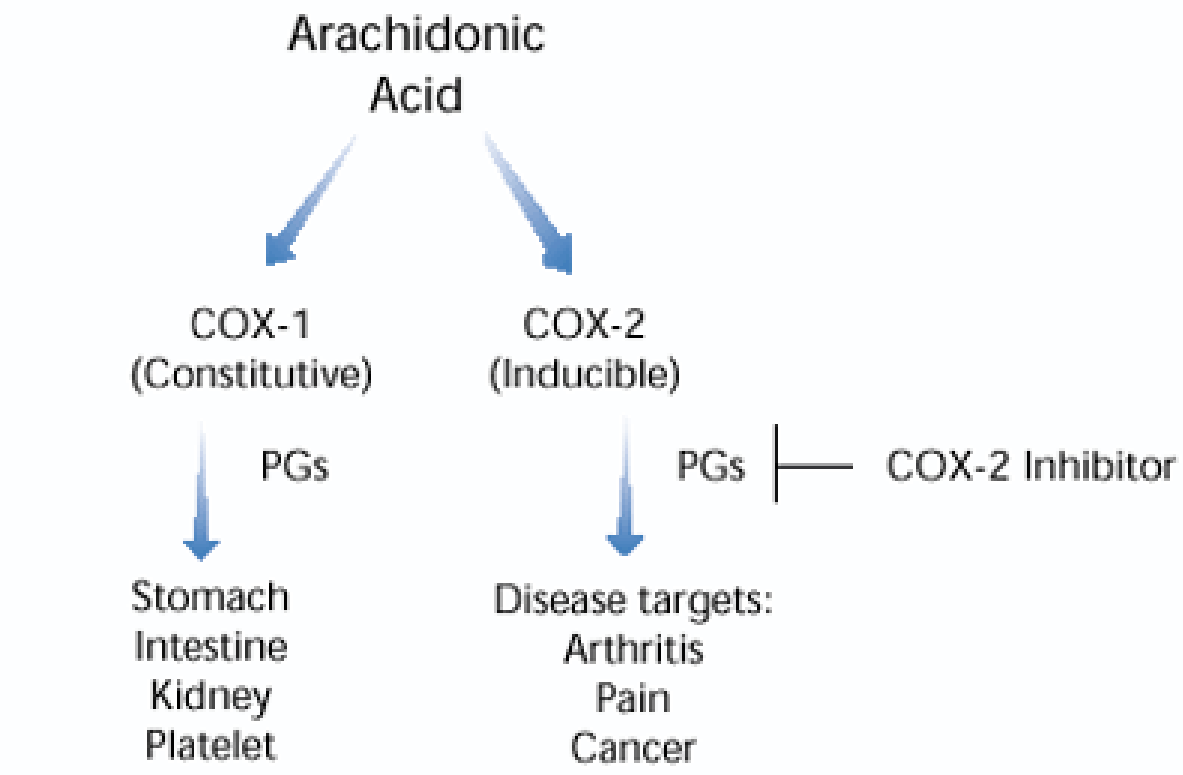
The Arachidonic Acid Pathway



From Kumar V, Cotran RS, Robbins SL: Robbins basic pathology, ed 7, Philadelphia, 2003, Saunders, p 281.

The Innate Immune Response: Pathways 9

Selective COX-2 inhibitors reduce inflammation while protecting the digestive tract.



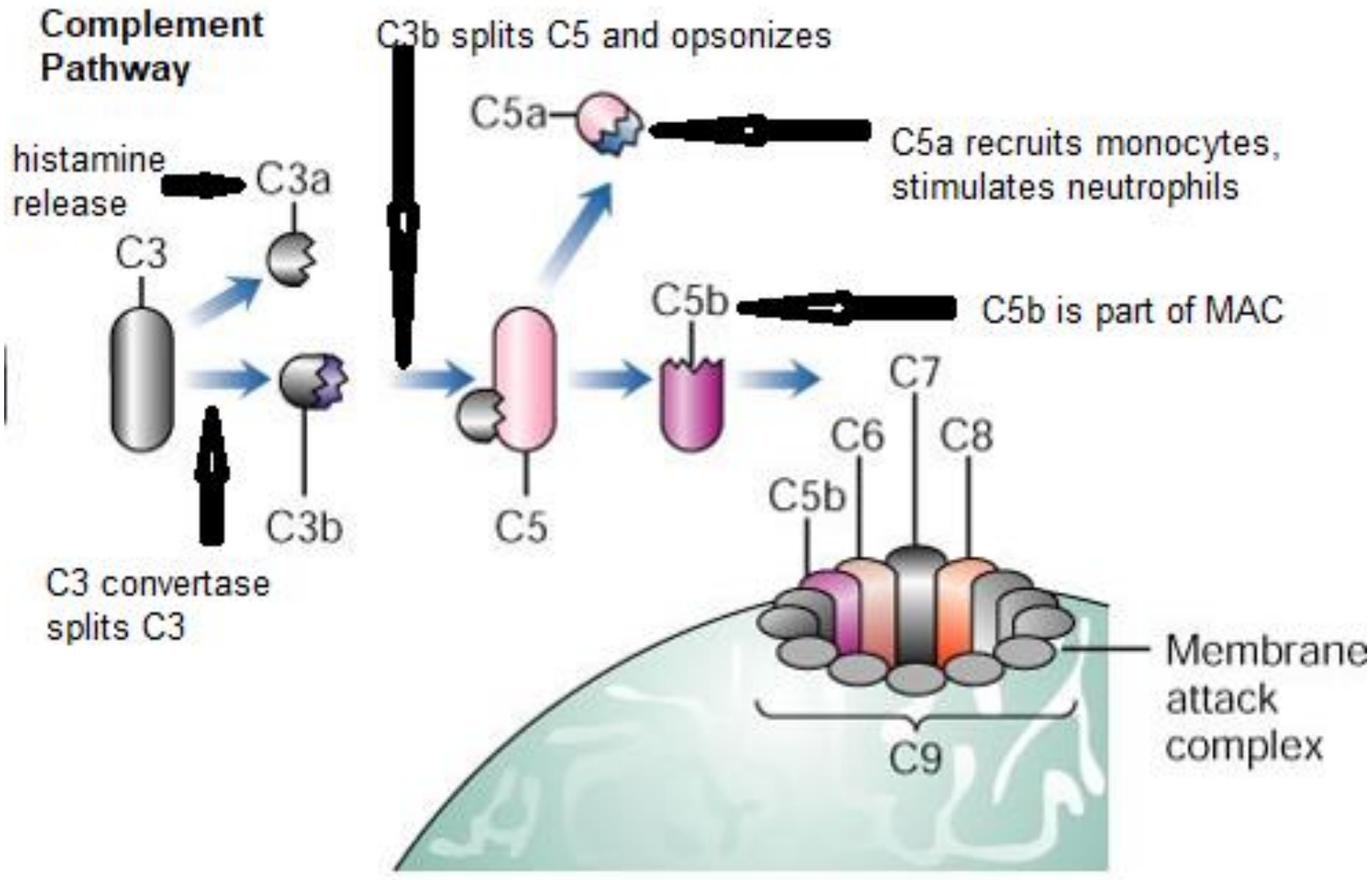
The Innate Immune Response: Pathways 10

- Another pathway that functions in the innate immune response (and the adaptive response too) is the **complement pathway**. Complement proteins are plasma proteins (synthesized by the liver) that normally circulate in the blood in an inactive form (like clotting factors).
- A pivotal enzyme in the complement pathway, **C3 convertase**, is activated in two possible ways.
  - The **classical pathway** requires the **adaptive** immune response and the presence of specific antibody-antigen complexes to activate C3 convertase.
  - The **alternative pathway** is part of the **innate** response and requires only the presence of a **pathogen** surface to activate C3 convertase. Molecules on pathogen surfaces that trigger the alternative pathway are lipopolysaccharide (LPS) and various bacterial exotoxins.

The Innate Immune Response: Pathways 11

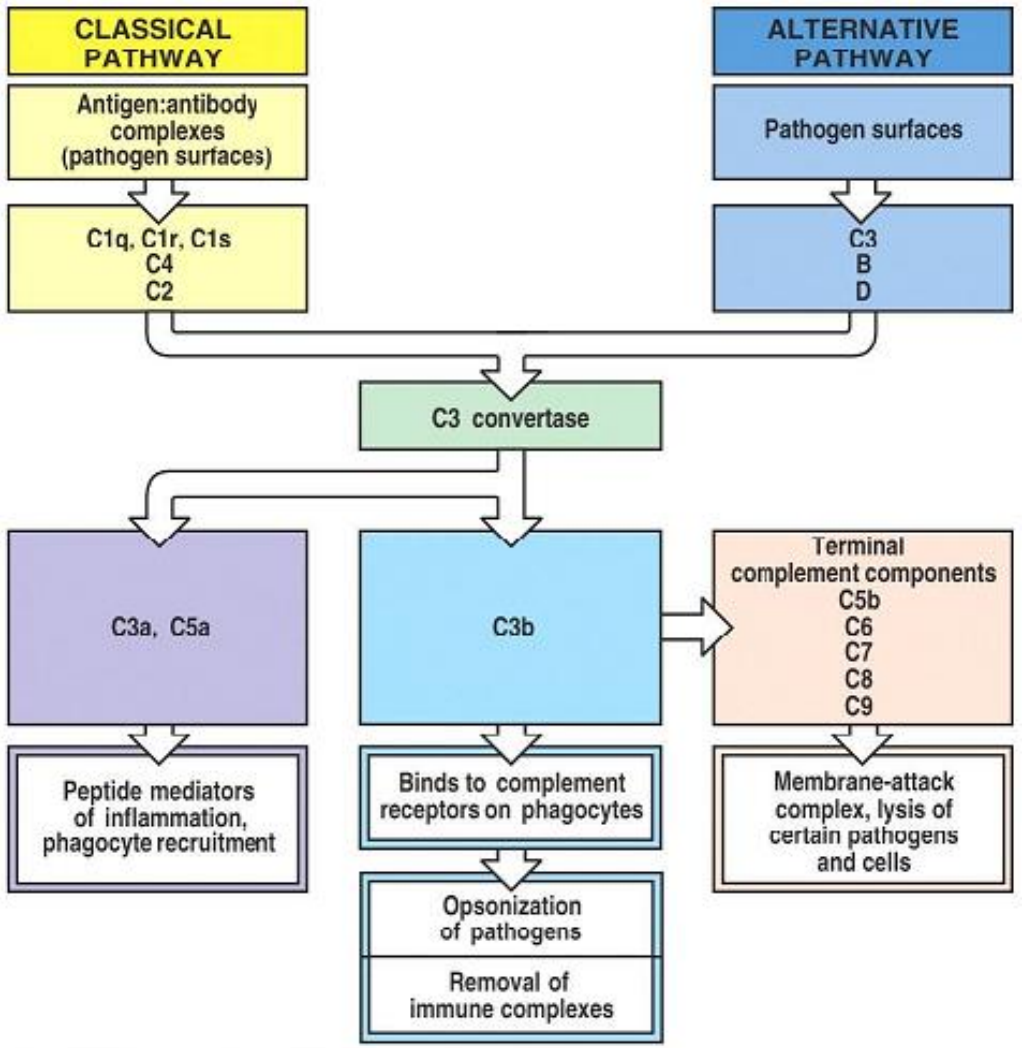
- Once C3 convertase is active, a group of several complement components are produced to act against the pathogen.
- **C3** is the most plentiful and most important complement protein. It is split by C3 convertase into C3a and C3b.
  - **C3a** stimulates the release of histamine from mast cells, increases capillary permeability, and stimulates smooth muscle contraction.
  - **C3b** cleaves C5 into C5a and C5b. It also acts to **opsonize** pathogens. Opsonization means to coat a structure with protein to make it more appetizing for phagocytes.
  - **C5a** is a chemotactic agent for neutrophils and monocytes, and it activates neutrophils by stimulating their production of oxygen free radicals and their uptake of glucose.
  - **C5b** combines with C6, C7, C8 and multiple units of C9 to form **"MACs", (membrane attack complexes)** donut shaped protein structures that will form a hole in the pathogen surface to kill it.

The Innate Immune Response: Pathways 12



The Innate Immune Response: Pathways 13

Two Modes of Complement Activation:



Lecture 3B:
Innate Immune Response Cells
Healing
Systemic Effects
Adaptive Immunity:
Cellular Immune Responses

The Innate Immune Response: Cells

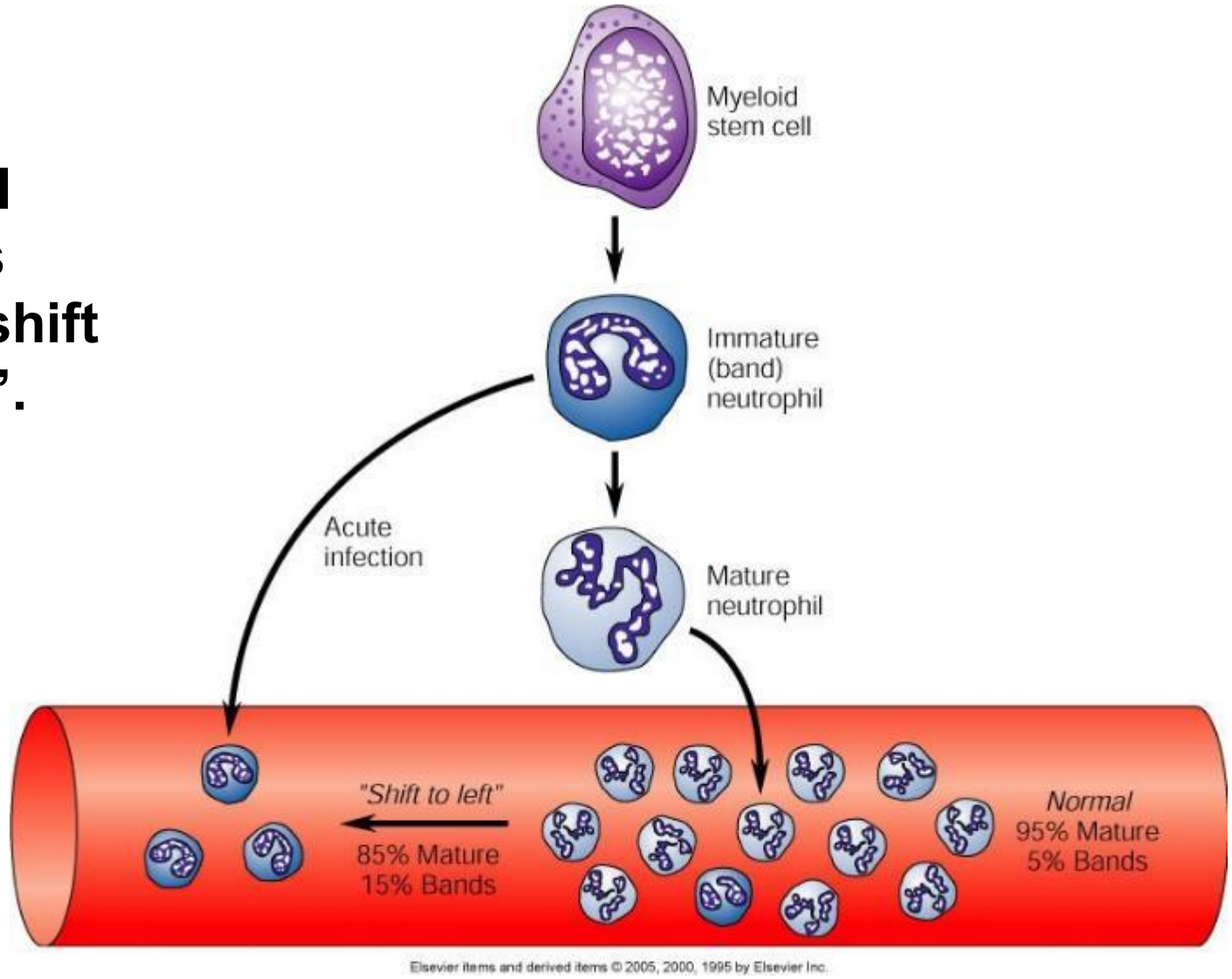
- WBCs that normally reside in the tissues, **macrophages, dendritic cells and mast cells**, are the first to be activated to participate in the inflammatory response.
  - The cells have **toll-like receptors (TLRs)** on their surfaces that recognize and bind molecules released by injured tissue cells and molecules produced by pathogens. TLR binding causes activation of the WBC.
  - Activated macrophages and mast cells secrete **cytokines** to stimulate **leukocytosis** (elevated production of WBCs by the red bone marrow) and recruit additional WBCs to the site by **chemotaxis**. Chemicals released by injured tissue, bacterial toxins and complement C5a also function as chemotactic agents for WBCs.
  - **Neutrophils** are the first and primary recruited responders. They mediate about 80% of the WBC innate response.
  - **Neutrophilia** (increase in circulating neutrophils) occurs.

The Innate Immune Response: Cells 1

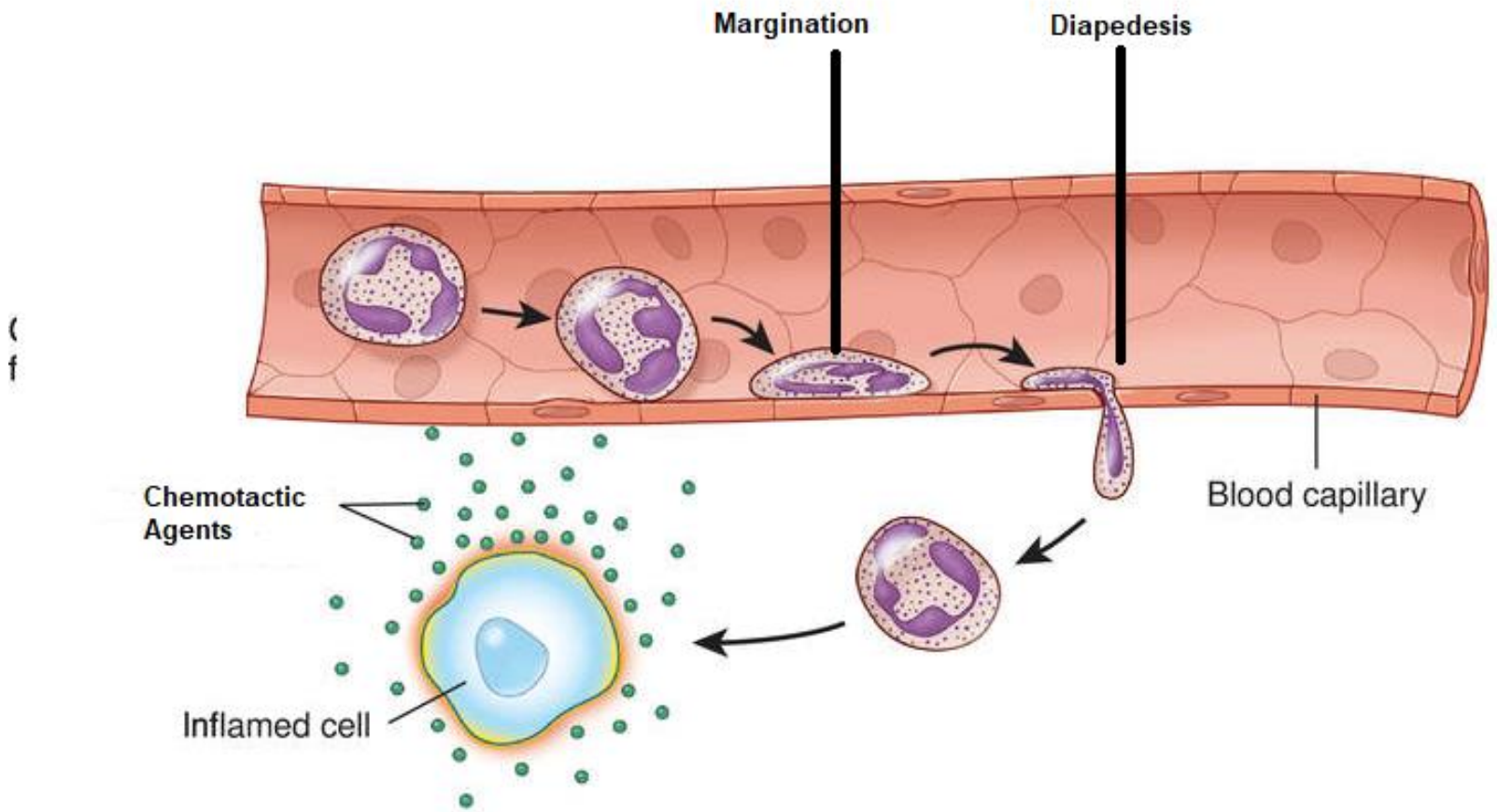
- If the injury is due to a bacterial infection, the demand for neutrophils is very high, so immature neutrophil **band cells** (They lack the nuclear constrictions of mature cells.) are released from the bone marrow into the bloodstream. This increase in band cell numbers is termed a **"shift to the left".**
- Chemicals released by injured tissue cells cause the expression of **adhesion molecules** on the endothelial cells that form the walls of the capillaries running through the site of injury.
- Receptors on the leukocyte surface bind to the adhesion molecules on the capillary walls.
- This causes **margination.** (Leukocytes coat the inside wall of the capillaries near the site of injury.)
- Neutrophils then undergo **diapedsis**. They squeeze through the gaps between endothelial cells and enter the tissue space.

The Innate Immune Response: Cells 2

Neutrophil band cells cause a "shift to the left".



The Innate Immune Response: Cells 3



The Innate Immune Response: Cells 4

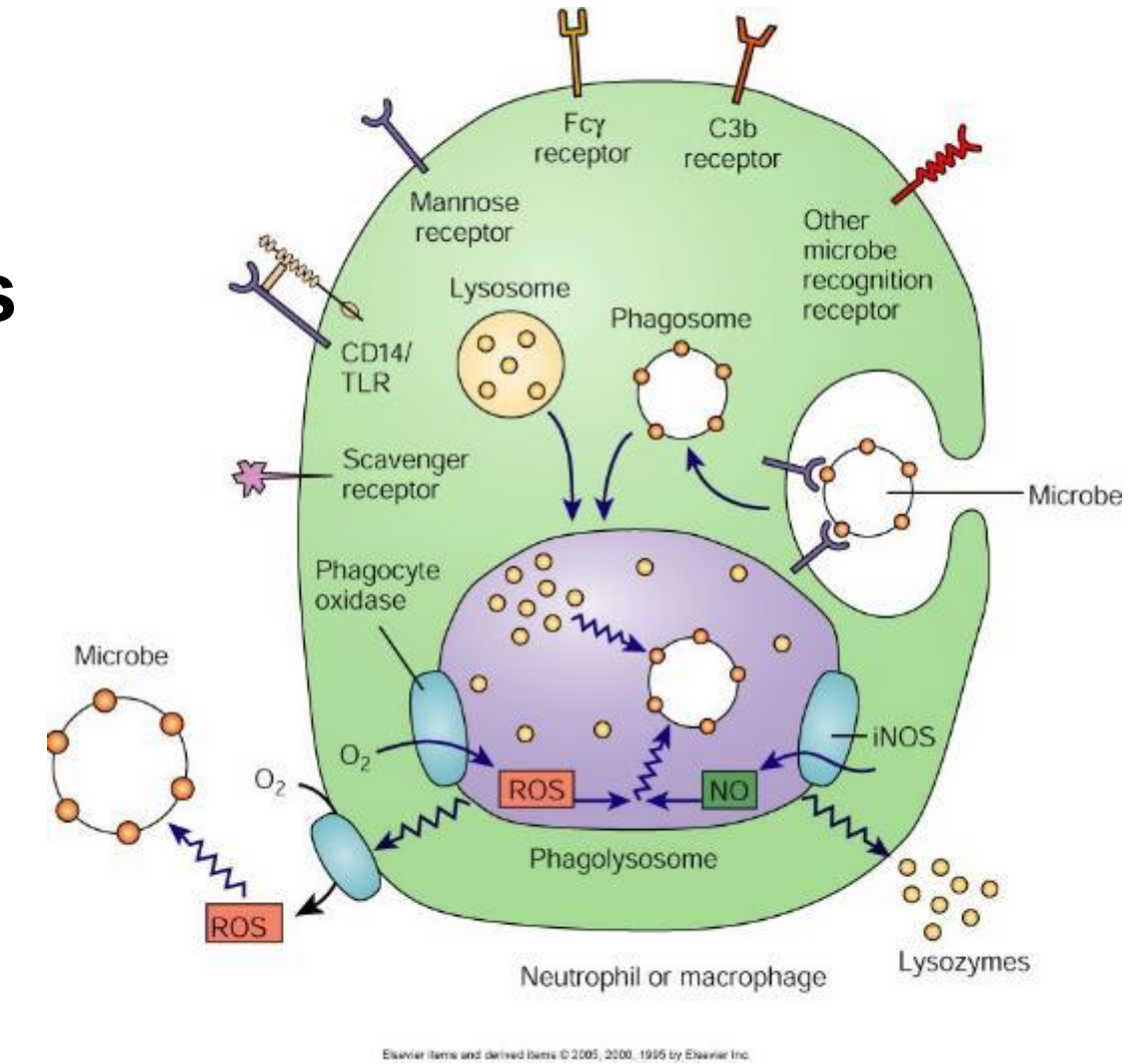
- **Monocytes** arrive next (becoming macrophages when they enter the tissue space) followed by **NK cells, eosinophils basophils and mast cells** (not necessarily in that order).
- **Eosinophil** numbers are highest if the cell injury is due to infection by parasitic worms. Worms are too large to be phagocytosed. Eosinophils release toxic chemicals onto the surface of the worms to kill them.
- Both **eosinophils and mast cells** are involved in allergic reactions, a later topic in this presentation.
- Leukocytes that are phagocytes have numerous types of receptors on their surfaces, not only for common microbe molecules, but also for the molecules that opsonize (coat) microbes such as **antibodies and complement C3b.** Antibody receptors on WBCs are called **Fc receptors**. Opsonizing antigens makes the antigens more appealing to phagocytes!

The Innate Immune Response: Cells 5

- Internalized **phagosomes** are fused with **lysosomes**. Some lysosomal enzymes are **proteases** others produce oxygen and nitrogen **free radicals**. If the microbe is too large to phagocytose the enzymes are secreted by the phagocyte onto the pathogen.
- **NK cells** directly kill virally infected human cells and cancerous human cells. Infected and cancerous human cells are altered so that they lack normal **MHC I self antigens** on their surfaces.
- It is the detection of the **"lack of self"** that causes NK cells to attack.

The Innate Immune Response: Cells 6

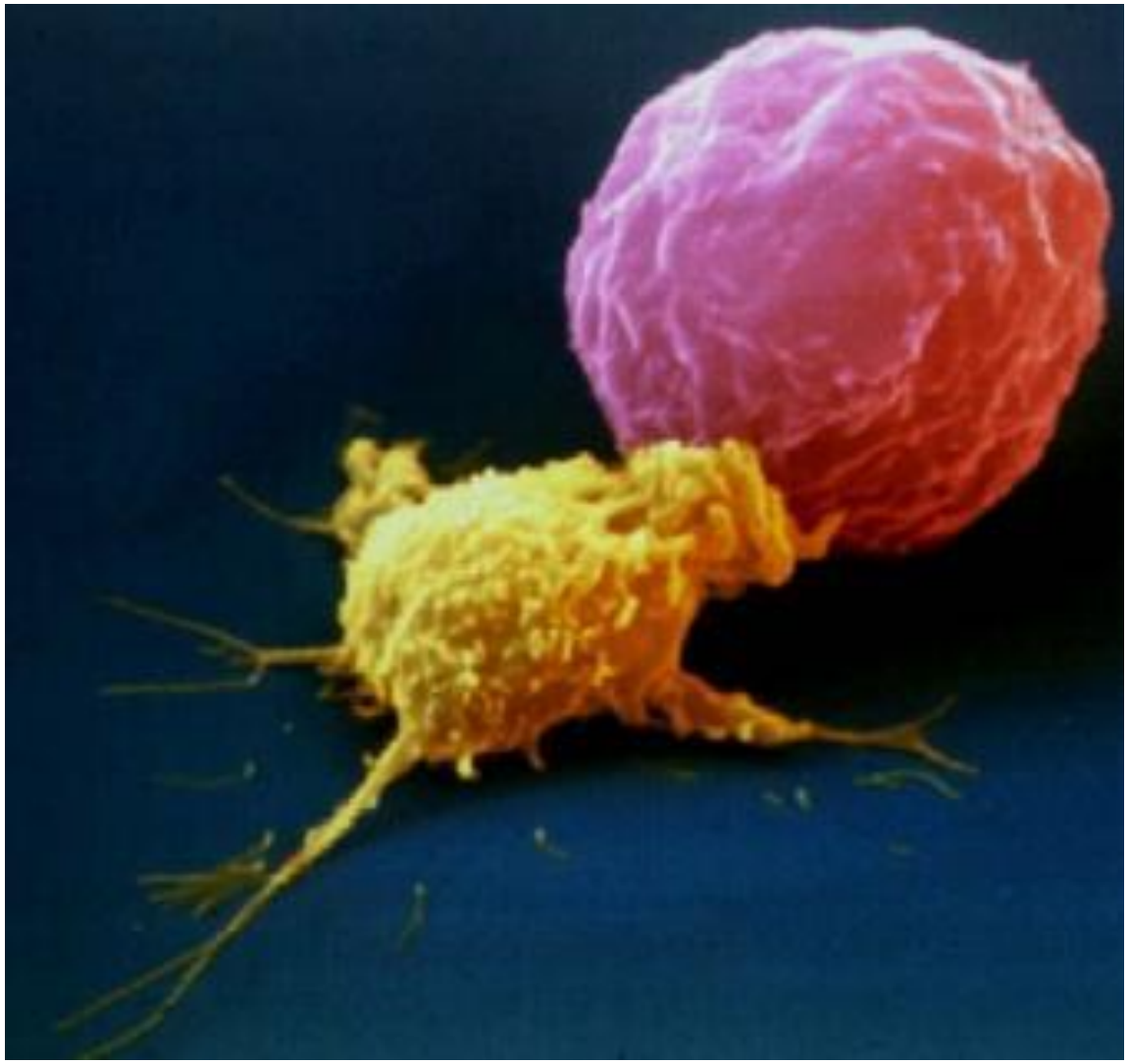
Active Phagocytes Release ROS and Enzymes



ROS=reactive oxygen species

The Innate Immune Response: Cells 7

NK cell (yellow) is activated by “lack of” self MHC I proteins on body cell surface (The pink cell is a tumor cell.).



The Innate Immune Response: Cells 8

- NK cells kill target cells by one of two methods.
  - **Apoptosis** by delivery of a **Fas ligand** on the NK cell surface to a **Fas receptor** on the target cell. (A ligand is a general term for a molecule that binds to a receptor.)
  - **Apoptosis** of the target cell by NK cell exocytosis of toxins: **perforins** (chemicals that cause perforations in the cell membrane of the target cell) and **granzymes** (enzyme molecules that enter the cell through perforations to trigger apoptosis).
- **Interferon (INF)** proteins of various types are released by NK cells, macrophages and activated lymphocytes.
  - All types of interferons **inhibit viral replication** inside human cells. They are stimulatory to leukocytes in general, and some of them induce fever.

The Innate Immune Response: Cells 9

- **Summary: Innate Immune Response, Initiation and WBC Recruitment**

1. The TLRs of resident WBCs (macrophages, dendritic cells, mast cells) bind to extracellular molecules that indicate the presence of infection or injury.
2. Resident WBCs secrete cytokines in response. Cytokines enter the blood.
3. The red bone marrow releases WBCs leading to leukocytosis. WBCs follow the cytokine trail (chemotaxis) through the blood into capillaries near the site of tissue infection/injury.
4. Recruited WBCs coat (marginate) the inside of capillary walls near the site of infection/injury.
5. Recruited WBCs exit the blood via diapedesis. Neutrophils arrive first, then monocytes, then NK cells, basophils, mast cells and eosinophils.

The Innate Immune Response: Healing

- Because inflammation can severely damage tissue, there is a system of inactivators in place.
  - **Alpha-1 antitrypsin** is an enzyme produced by the liver. It circulates constantly to disarm the proteases released by phagocytes, particularly neutrophils.
  - **Neutrophils have a limited life span** once they leave the bone marrow. They will die at the site of injury and contribute to the formation of **pus**=dead microbes, dead neutrophils and tissue debris.
- As the inflammatory response subsides **macrophages** stay on the scene to assist in repair. They have multiple repair functions.

The Innate Immune Response: Healing 1

- Phagocytosis of dead microbes, dead neutrophils and dead tissue.
- Release of **tissue thromboplastin** to stimulate fibroblasts to lay down new connective tissue.
- Release of **angiogenesis factors** to stimulate the formation of new blood vessels.

- Regeneration of the damaged tissue to the original tissue type requires **survival of the basement membrane and stem cells**. Otherwise **scarring**, the replacement of injured tissue with highly fibrotic connective tissue, occurs.
- **Chronic inflammation**, may impair healing and cause the formation of a **granuloma**, an accumulation of modified macrophages, fibroblasts and collagen. Granuloma formation is due to the **failure of phagocytosis** to remove foreign materials.

The Innate Immune Response: Healing 2

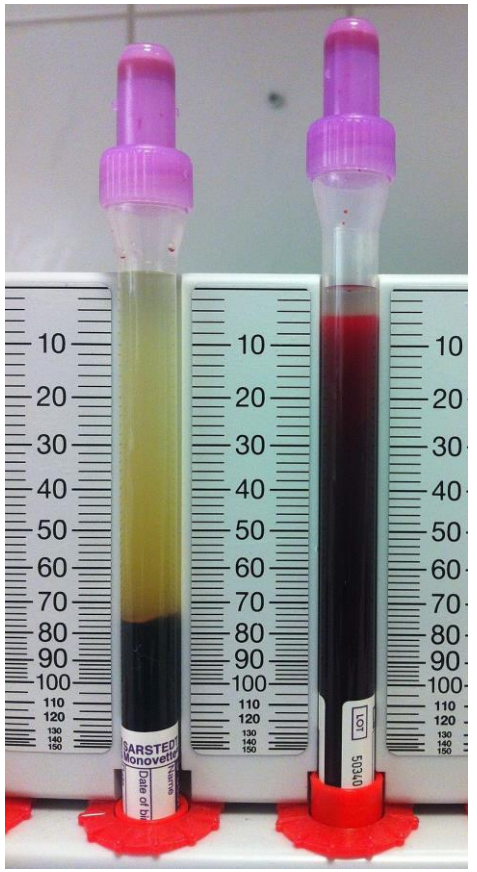
- **Inflammatory exudate** is the fluid that leaks out of capillaries combined with neutrophils and cell debris from phagocytosis. Exudate functions in three ways:
  - Transport of leukocytes and antibodies.
  - Dilution of toxins and irritating substances
  - Transport of nutrients required for repair
- The type of exudate indicates the severity of inflammation.
  - **Serous exudate** is watery with a low protein content. It is associated with mild inflammation.
  - **Fibrinous exudate** contains fibrin protein, so it is thick and sticky. It may have to be removed to promote healing.
  - **Purulent exudate** is **pus**. It forms most often due to a severe inflammatory response to bacterial infection. Large collections of pus are **abscesses**. They are usually drained to promote healing.
  - **Hemorrhagic exudate** contains a high number of red blood cells and is associated with the most severe inflammation and the leakiest capillaries.

The Innate Immune Response: Healing 3

- Thus far inflammation has been discussed as a local phenomenon, but it can have **systemic effects** depending on the severity of the injury and the immune capabilities of the individual. Common systemic effects are fever, lethargy and muscle catabolism.
- Three macrophage-derived cytokines are responsible for most systemic effects: **IL-1, IL-6 and TNFα.** (IL=interleukin; TNF=tumor necrosis factor)
  - **IL-1, IL-6 and TNFα** act on the liver, causing it to release **acute phase proteins**. For example, **C-reactive protein (CRP),** produced by the liver, is an opsonin. It binds to phospholipids on bacterial surfaces. Serum CRP is a **clinical marker** for inflammation.

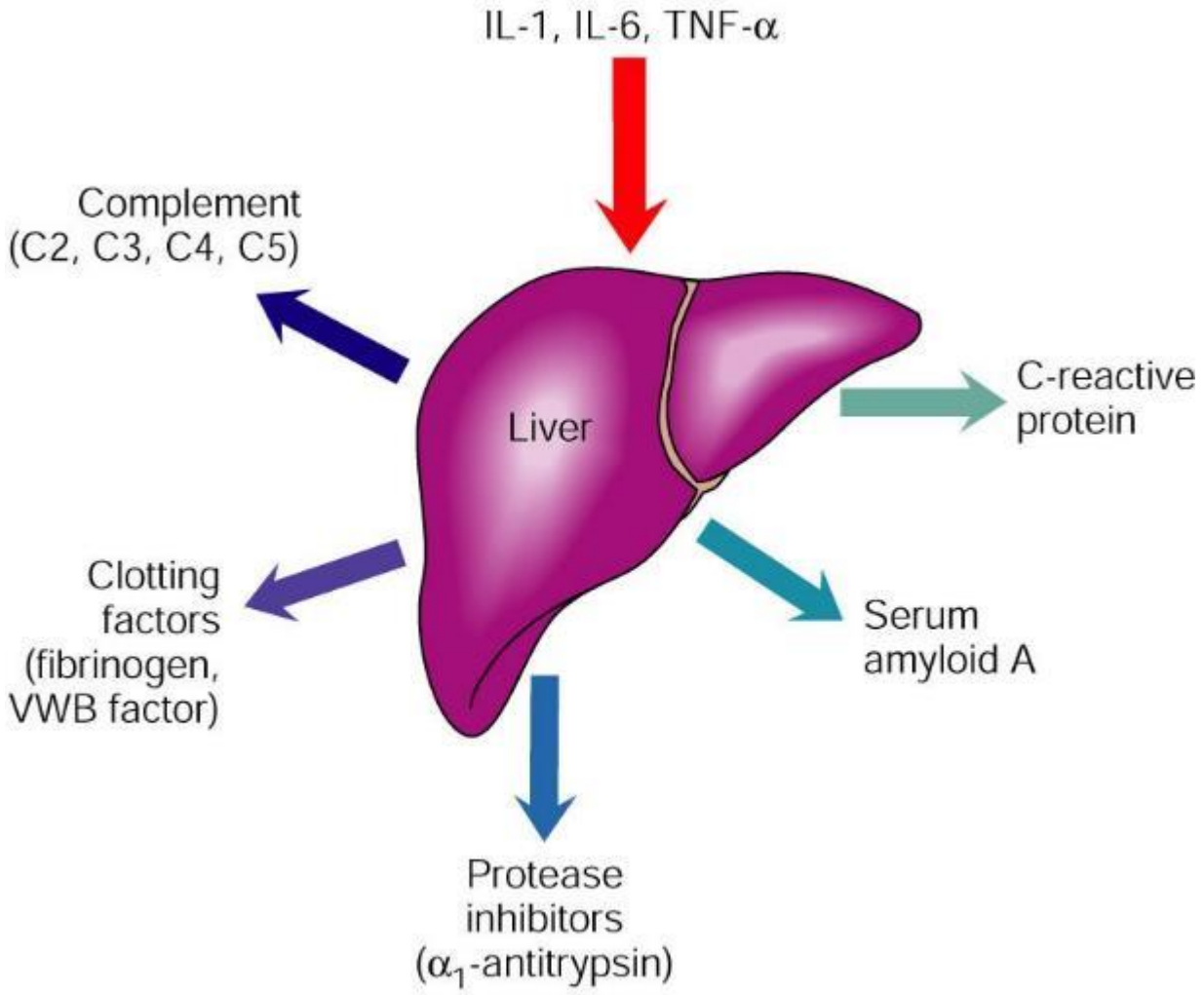
The Innate Immune Response: Systemic Effects

- A blood test called the **ESR (erythrocyte sedimentation rate or "sed rate")** is important clinically as an indicator of the extent of inflammation.
- The **fibrin** produced by the coagulation pathway (The liver synthesizes clotting factors.) adheres to red blood cells causing them to become heavier.
- The RBCs thus sink to the bottom of a tube of blood (sedimentation) at a faster rate. The higher the ESR, the more extensive the inflammation.
- In the photo the tube on the left contains blood from a patient suffering from pneumonia. The control is on the right.



The Innate Immune Response: Systemic Effects 1

In response to macrophage cytokines the liver synthesizes proteins that play several roles in innate immunity.



The Innate Immune Response: Systemic Effects 2

- **Fever**
  - **Fever** is an activity of the hypothalamus that causes an increase in body temperature. Fever speeds repair, increases cytokine secretion and impedes the activity of some microbes.
  - Fever is stimulated by **prostaglandin E-2 (PGE-2),** a derivative of arachidonic acid. **PGE-2** is an **internal pyrogen**.
  - **External pyrogens** also exist. The endotoxin, **lipopolysacharride (LPS),** of Gram- bacterial cell walls, is an example.

The Adaptive Immune Response

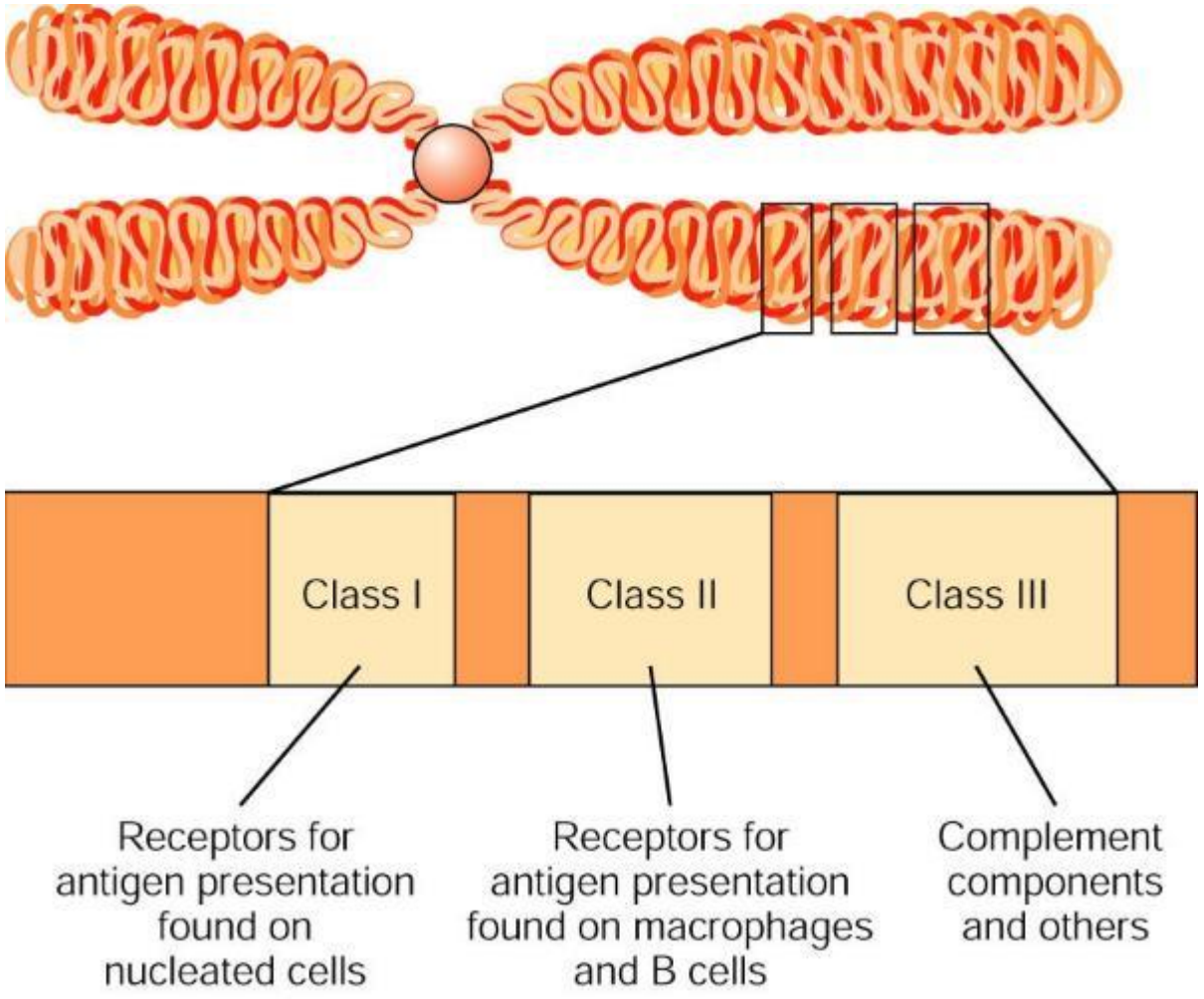
- The adaptive immune response is considered the **third line of defense** against tissue injury. It includes the **cellular immune response** provided by **T lymphocytes** and the **humoral immune response** provided by **B lymphocytes**. (Body fluids are also known as body humors.)
- The adaptive response takes longer to develop than the innate response.
- An amazing property of the adaptive immune response is **immunologic memory**. Having once reacted to a specific antigen, the adaptive immune system will provide a faster, more effective response upon a subsequent exposure to the same specific antigen. **Memory cells** are produced and persist.
- B cells and T cells must react to foreign antigens yet remain tolerant of self antigens.

The Adaptive Immune Response 1

- **Major histocompatibility Complex (MHC) proteins** play a major role in lymphocyte reactivity and self tolerance.
- MHC refers to a cluster of genes on chromosome 6. These genes fall into three classes: I, II and III.
- **Class I and II genes** code for proteins that **present antigens** on the surface of APCs. Antigen presentation is the first step in many adaptive immune responses. T cells cannot recognize foreign antigens unless they are displayed along with MHC proteins. MHC proteins are also called **self antigens.**
- **Class III genes** code for proteins important in the inflammatory response, complement proteins, for example.
- Each individual has three pairs of Class I alleles and three pairs of Class II alleles. Multiple possible alleles exist for each MHC gene, so it is highly unlikely that any two individuals will have identical MHC genotypes, except for identical twins.

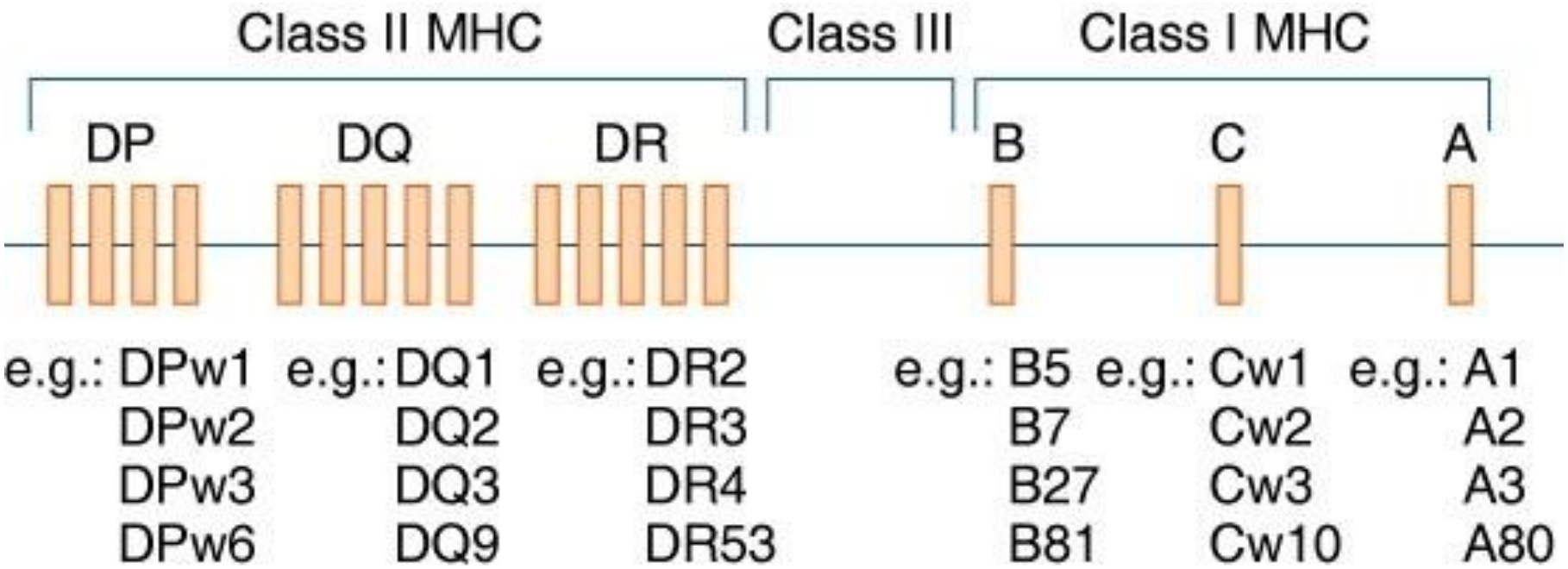
The Adaptive Immune Response 2

MHC Genes on Chromosome 6



The Adaptive Immune Response 3

MHC Genes Have Several Alleles



The Adaptive Immune Response 4

- Partial matching (75% is best) of MHC alleles is crucial in organ transplantation to avoid rejection. Genetically related individuals have the best chance of having some alleles in common. Organs donors are usually family members.
- **MHC I proteins** are expressed by and displayed on the surface of all **nucleated** body cells (**all body cells but mature RBCs**).
- **MHC II proteins** are expressed by and displayed only on the surface of leukocytes that function as **APCs** (antigen presenting cells): macrophages, dendritic cells and B cells. So APCs have both MHC I and MHC II proteins.
- MHC I and MHC II surface proteins bind and display bits of normal **INTRACELLULAR (synthesized in the cell displaying it)** self protein. Self-tolerant lymphocytes **notice (bind loosely), but do not react** to those MHC/self peptide complexes.

The Adaptive Immune Response: Cellular Immunity

- **Antigen Presentation by MHC I to Cytotoxic T cells**
  - If a body cell becomes infected or malignant the bit of **normal** self protein displayed by **MHC I** will be replaced by a bit of **abnormal** (non-self) protein produced inside the injured cell in response to the infection or cancer.
  - **NK cells and effector cytotoxic T cells** recognize this change and induce apoptosis in the cell bearing the **MHC I/non-self protein complex**. Cytotoxic T cells induce apoptosis by the same mechanism as NK cells.
  - NK cells respond to **"absence of normal MHC I complex"**. The bit of abnormal protein displayed could be from any virus or any form of malignancy. NK cells don't "know" the diff.
  - **Cytotoxic T cells (CD8 T cells)** respond to the **specific non-self antigen** displayed by MHC I. Each CD8 cell is genetically programmed to respond to the **"presence of a specific foreign (non-self) antigen"**, a piece of viral protein perhaps.

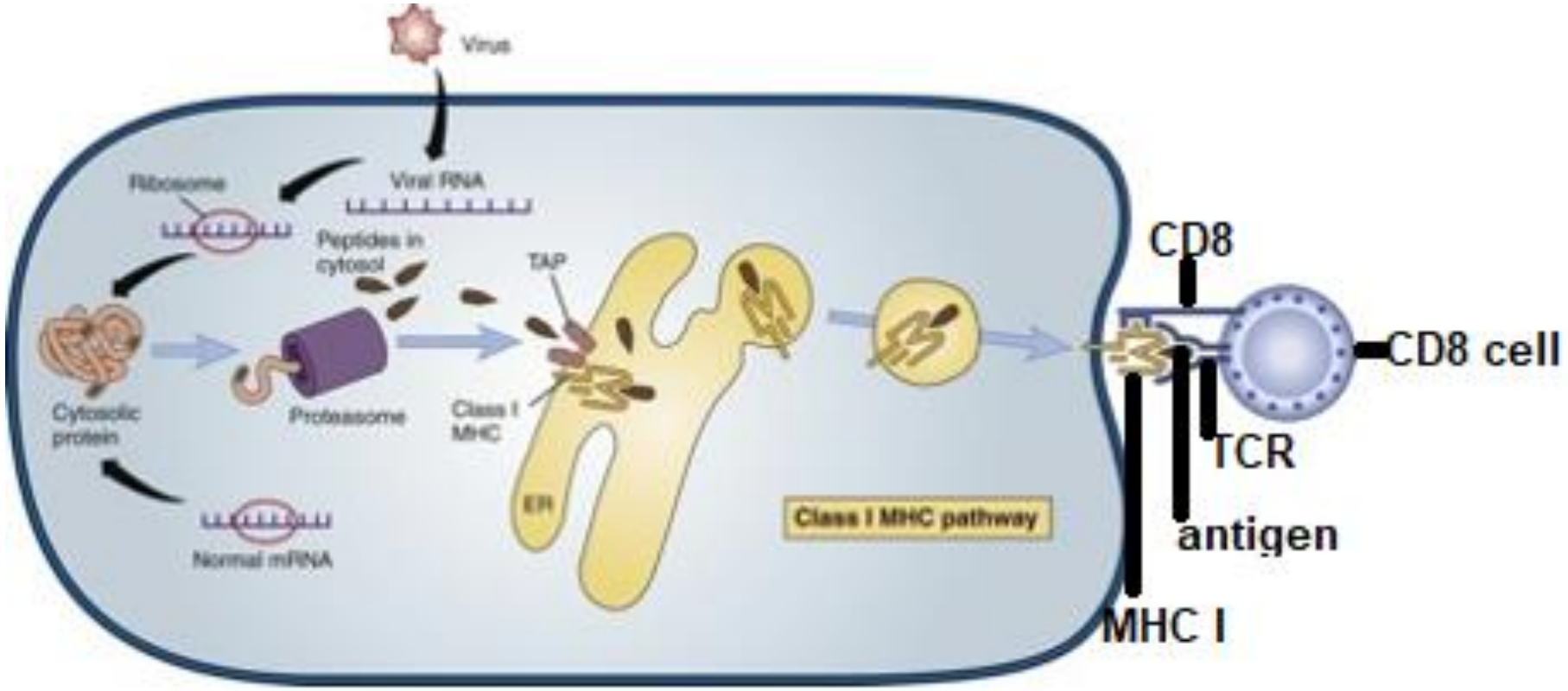
The Adaptive Immune Response: Cellular Immunity 1

- Multiple requirements for activating a **naïve** cytotoxic T cell:
- **T cell receptors (TCRs)** on the surface of the cytotoxic T cell must bind to the MHC I/abnormal peptide complex. This binding is **foreign antigen specific**. The 3D shape of the TCR will normally bind only to the 3D shape of one type of MHC I/peptide complex. Cytotoxic T cells are "MHC I restricted".
- A **CD8 protein** on the cytotoxic T cell surface (Each cytotoxic T cell has thousands of CD8 protein molecules). CD8 stabilizes the binding of the TCR to the MHC I/abnormal peptide complex**. CD3 protein** on the T cell acts in signal transduction into the cell.
- The cytotoxic T cell may require a **costimulatory signal** from the body cell. Most commonly a **CD28** protein on the T cell must bind to a **B7 (aka CD 80)** protein on the body cell.

The Adaptive Immune Response: Cellular Immunity 2

- Activation of naïve lymphocytes, both T cells and B cells, includes **clonal expansion and differentiation**.
  - **Clonal expansion** refers to rapid **mitosis** to form a clone of identical cells all having the same antigen specificity.
  - **Differentiation** refers to maturation by changes in **gene expression**. Some genes are turned off; others turned on.
    - Some members of the clone become **effector cytotoxic T cells** that seek out and attack body cells (using perforins and granzymes or by delivering the Fas ligand) bearing the same MHC I/abnormal peptide complex.
    - Other members of the clone become **memory cytotoxic T cells** that will wait until a subsequent exposure to the same foreign antigen to react. Memory cells are already activated, so they will respond immediately to the subsequent exposure.

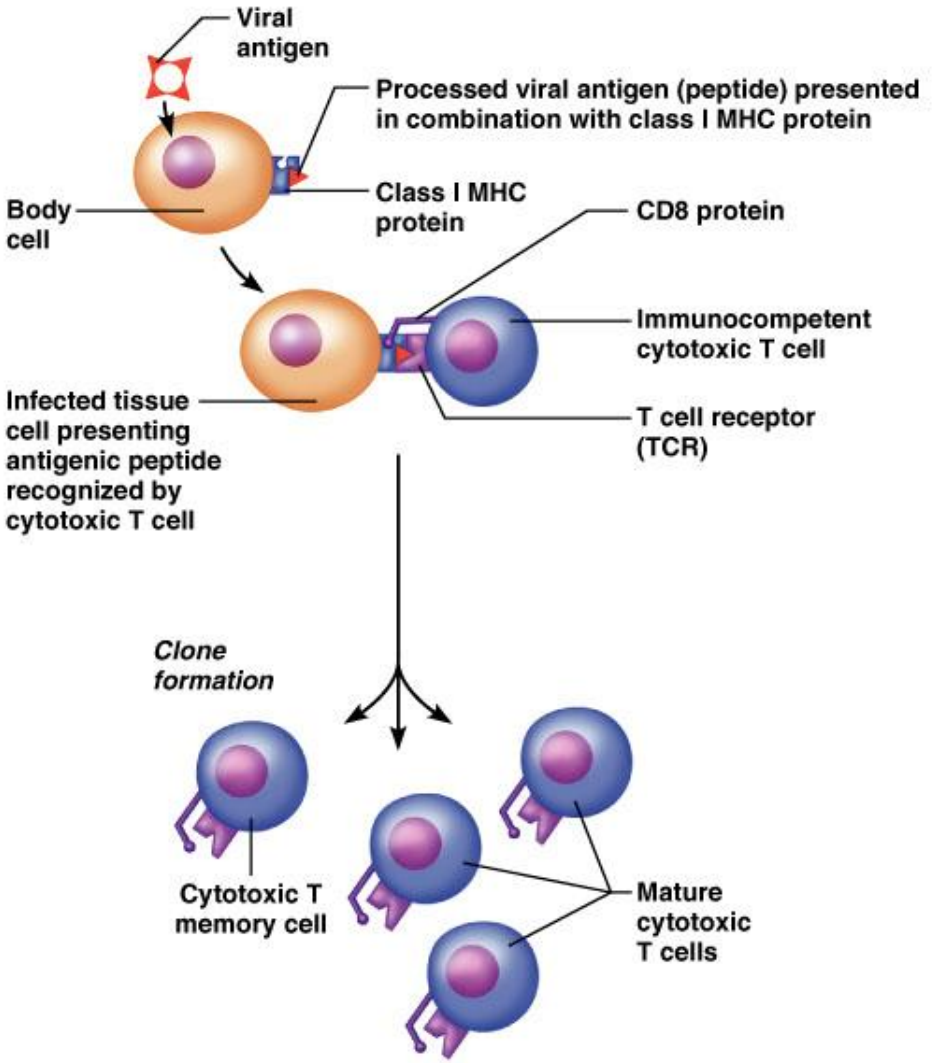
The Adaptive Immune Response: Cellular Immunity 3



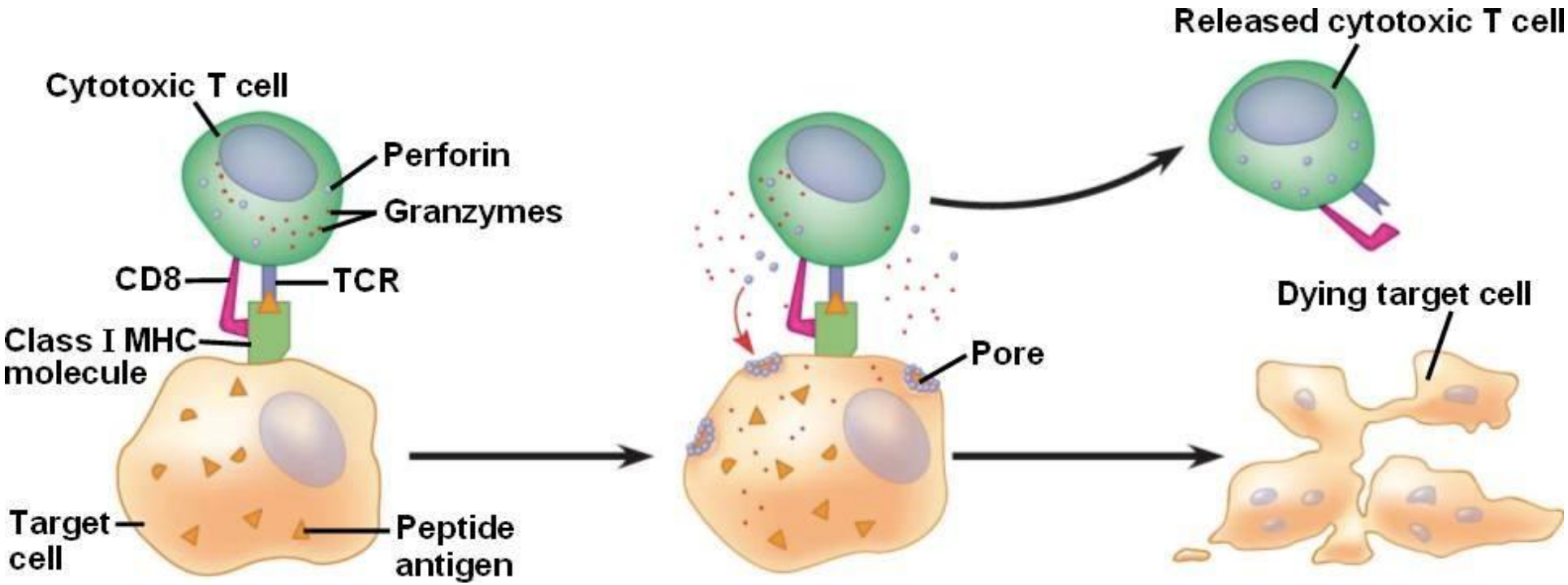
Naïve Cytotoxic T Cell Activation by a Virus-Infected Body Cell Through MHC I INTRACELLULAR Antigen Presentation

The Adaptive Immune Response: Cellular Immunity 4

Clonal Expansion and Differentiation by Activated Cytotoxic T cell

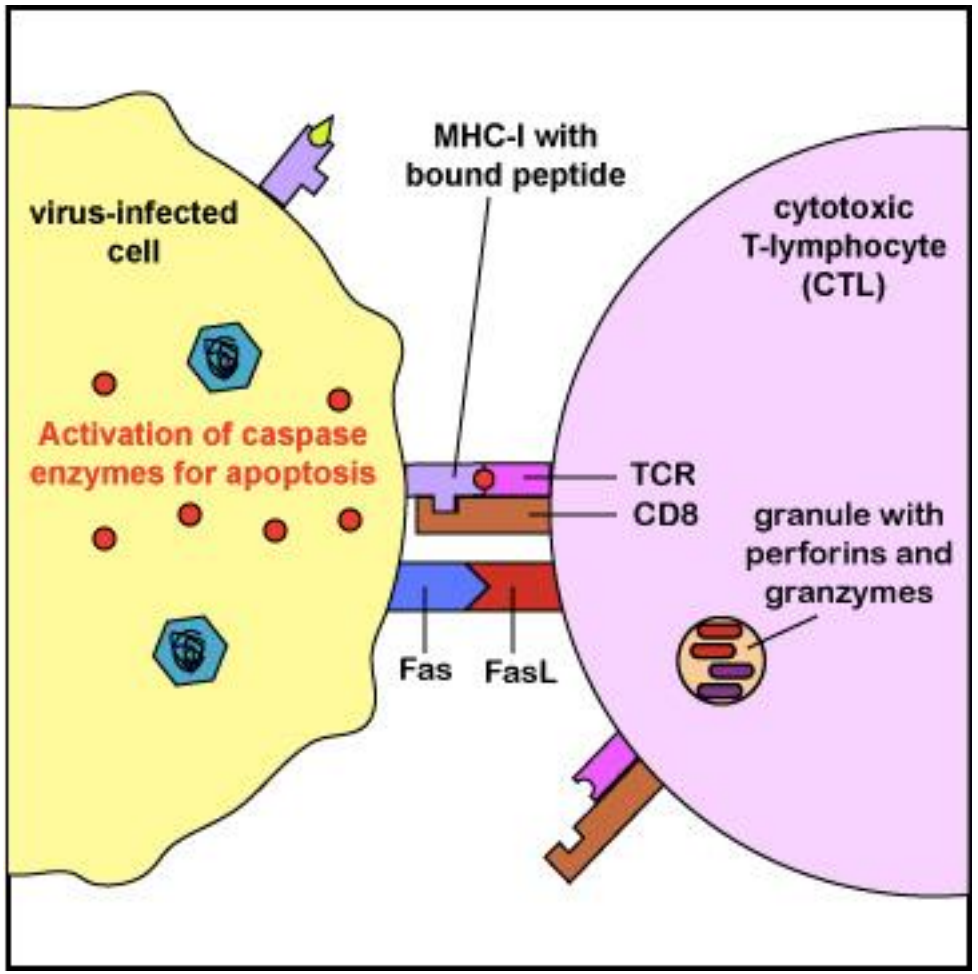


The Adaptive Immune Response: Cellular Immunity 5



Effector Cytotoxic T Cell Employing Perforins and Granzymes to Kill Virus Infected Cell

The Adaptive Immune Response: Cellular Immunity 6



Effector Cytotoxic T Cell Employing Fas Ligand (FasL) to Kill Virus Infected Cell by Apoptosis.

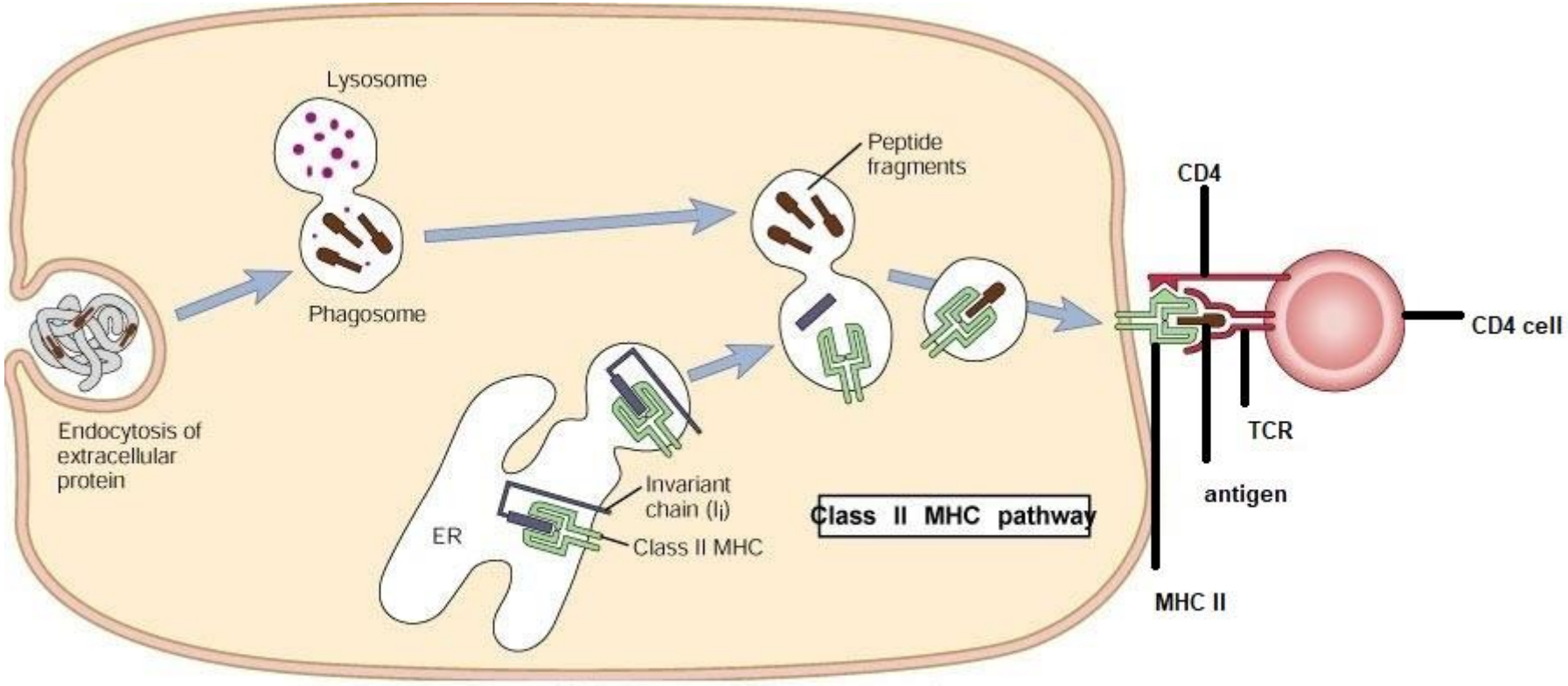
The Adaptive Immune Response: Cellular Immunity 7

- **Antigen presentation by MHC II Proteins to Helper T cells**
  - **Helper T cells (CD4 cells)** play a crucial role in BOTH cellular immunity and humoral immunity because they "help" other immune cells **by secreting substances** that stimulating immune cell activity.
  - A **naïve** helper T cell is activated to become an **effector** helper T cell when it interacts with an APC (usually a dendritic cell in a lymph node) that is presenting the specific foreign **EXTRACELLULAR** (not synthesized by the APC) antigen to which the naïve helper T cell is genetically programmed to respond.
  - The APC has **phagocytosed** and **processed** the protein antigen (broken it into peptides). A peptide from the antigen has been **bound to MHC II** protein. The MHC II/antigen complex has been **moved** to the surface of the APC.

The Adaptive Immune Response: Cellular Immunity 8

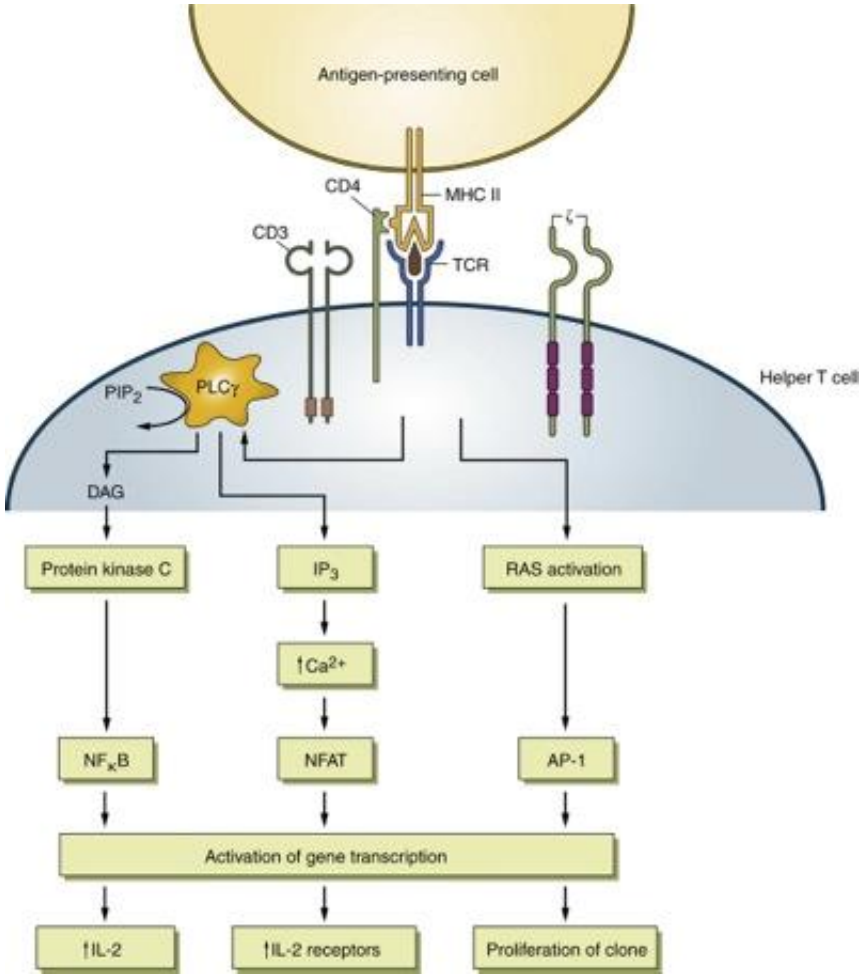
- **T cell receptors (TCRs)** on the surface of the naïve helper T cell bind to the MHC II/abnormal peptide complex. This binding is **foreign antigen specific**. The 3D shape of the TCR will normally bind only to the 3D shape of one type of MHC II/peptide complex. Helper T cells are "MHC II restricted".
- A **CD 4 protein** on the cytotoxic T cell surface (Each cytotoxic T cell has thousands of CD 4 protein molecules). CD4 stabilizes the binding of the TCR to the MHC II/abnormal peptide complex. **CD3 protein** transmits the signal into the T cell.
- The naïve helper T cell must receive a **costimulatory signal** from the APC (**B7** on the APC surface binds to **CD28** on the T cell surface is the most common costimulatory signal.).
- The helper T cell responds by synthesizing and displaying **IL-2 receptors** and by synthesizing and secreting **IL-2**. The IL-2 must bind to **IL-2 receptors** on the helper T cell surface.

The Adaptive Immune Response: Cellular Immunity 9



Helper T Cell Activation by a Dendritic Cell Through MHC II EXTRACELLULAR Antigen Presentation

The Adaptive Immune Response: Cellular Immunity 10



Naïve Helper T Cell Activation: Cytoplasmic Changes

The Adaptive Immune Response: Cellular Immunity 11

- Activation of a naïve CD4 cell involves clonal expansion and differentiation into memory Helper T cells and effector Helper T cells. There are two types of effector Helper T cells.
  - **TH1 cells**
    - stimulate **T cell, NK cell and macrophage** activity
    - are involved in **autoimmunity.**
    - secrete mostly **IL-2 and INFγ** (interferon gamma)
  - **TH2 cells**
    - stimulate **B cell** activity
    - are involved in **allergies.**
    - secrete mostly **IL-4, IL-5, IL-10 and IL-13**

The Adaptive Immune Response: Cellular Immunity 12

T cell Activation Summary Chart

	Cytotoxic T cell Activation	Helper T cell Activation
MHC Restriction	MHC I	MHC II
Antigenic Peptide Source	Intracellular	Extracellular
APC Provides	MHC I, antigen, B7	MHC II, antigen, B7
T cell Provides	TCR, CD8, CD3, CD28 (may require Helper T cell assistance)	TCR, CD4, CD3, CD28, IL-2, IL-2 receptor
Activation Result	Effector cells Perforins, granzymes, Fas ligand Memory cytotoxic T cells	Effector cells TH1: help T cells, NK cells, Macrophages TH2: help B cells Memory helper T cells

QUIZ 3AB

- COMPLETE QUIZ 3AB.
- THEN GO ON TO MODULE 3CD PPT.