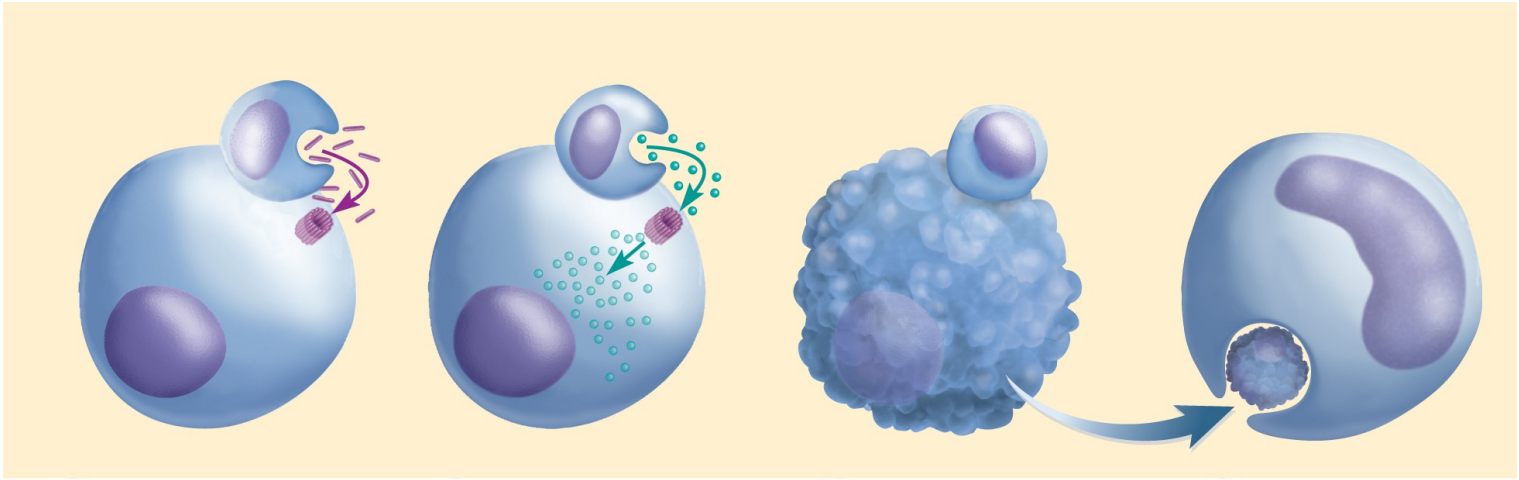
Chapter 21.4

The Immune System's Three Lines of Defense

The Features of the First Line of Defense

The Features of the Second Lines of Defense

Cellular Response VS Humoral Response to Pathogens



The Immune System Defends Us Against Pathogens

- What is a pathogen? (*microorganism capable of producing disease in a healthy person)*
- What may damaged our tissues? *infections (by bacteria, virus, parasites), toxins, chemicals, physical trauma, and radiation*
- *Human host cellss must defend themselves against internal and external threats that may cause them damage.*
- *Immune system use three separate "lines of defenses"*
  - First Line – physical barriers
  - Second Line – non specific resistance
  - Third Line – adaptive immunity

These two are innate

Not innate

The three defenses against pathogens.

- First line of defense = external barriers = Skin + mucous membranes // innate defense – because present at birth

- Second line of defense – provides non-specific resistance to pathogens

    - Innate- because present at birth

    - *Uses different methods /// leukocytes and macrophages, antimicrobial proteins, immune surveillance cells, inflammation, and fever*

    - effective against a broad range of pathogens / but not any one specific pathogen!

- Third line of defense (acquired immunity – requires activation // not innate -because this defense developes afer birth)
    - defeats “specific” pathogens // key idea is specificity!
    - leaves body with ‘memory cells’ – allows for more rapid secondary response to pathogen
    - cellular and humoral response (able to attack pathogen inside or outside of our cells!)

First Line of Defense - “Different Factors”

Physical Factors	Chemical Factors
Epidermis of skin	Sebum
Mucous membranes	Lysozyme
Mucus	Gastric juice
Hairs	Vaginal secretions
Cilia	
Lacrimal apparatus	
Saliva	
Urine	
Defecation and vomiting	

The External Barriers

- **Skin**
  - makes it mechanically difficult for microorganisms to enter the body
  - toughness of keratin
  - too dry and nutrient-poor to support microbial growth
  - **defensins** – peptides that kill microbes by creating holes in their membranes
  - **acid mantle** – thin film of lactic acid from sweat which inhibits bacterial growth

The External Barriers

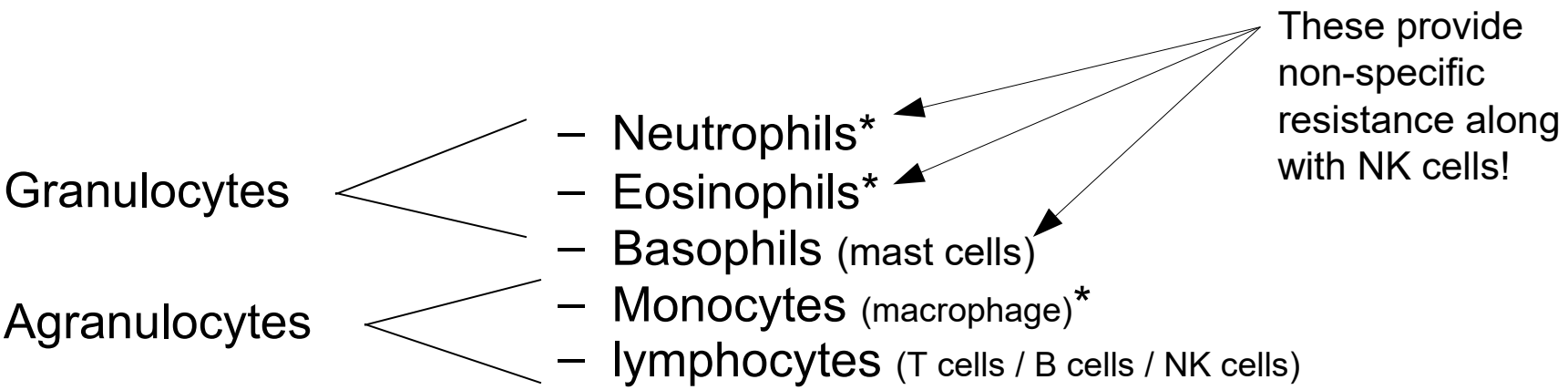
- Mucous membranes
  - digestive, respiratory, urinary, and reproductive tracts are open to the exterior and protected by mucous membranes
  - mucus physically traps microbes
  - lysozyme - enzyme destroys bacterial cell walls
- Sub-epithelial areolar tissue
  - viscous barrier of hyaluronic acid
  - hyaluronidase - enzyme used by pathogens to make hyaluronic acid less viscous

Second Line of Defense - Different Factors

Antimicrobial Substances	Cellular
Interferon	Natural killer cells
Complement system	Phagocytes
Iron-binding proteins	Physiologic
Antimicrobial proteins	Inflammation
	Fever
	TOLL Like Receptors / PAMP

Second Line of Defense / Cellular Contributions

(Review Function of Formed Elements)



- Phagocytes provide the cellular component
  - Able to engulf bacteria, endo-cytocytosis
  - Dead infected cells, and fragments of cells
  - internalized as phagosome / fuse with lysosomes
- Neutrophils kill bacteria and eosinophils kill parasites with respiratory bursts

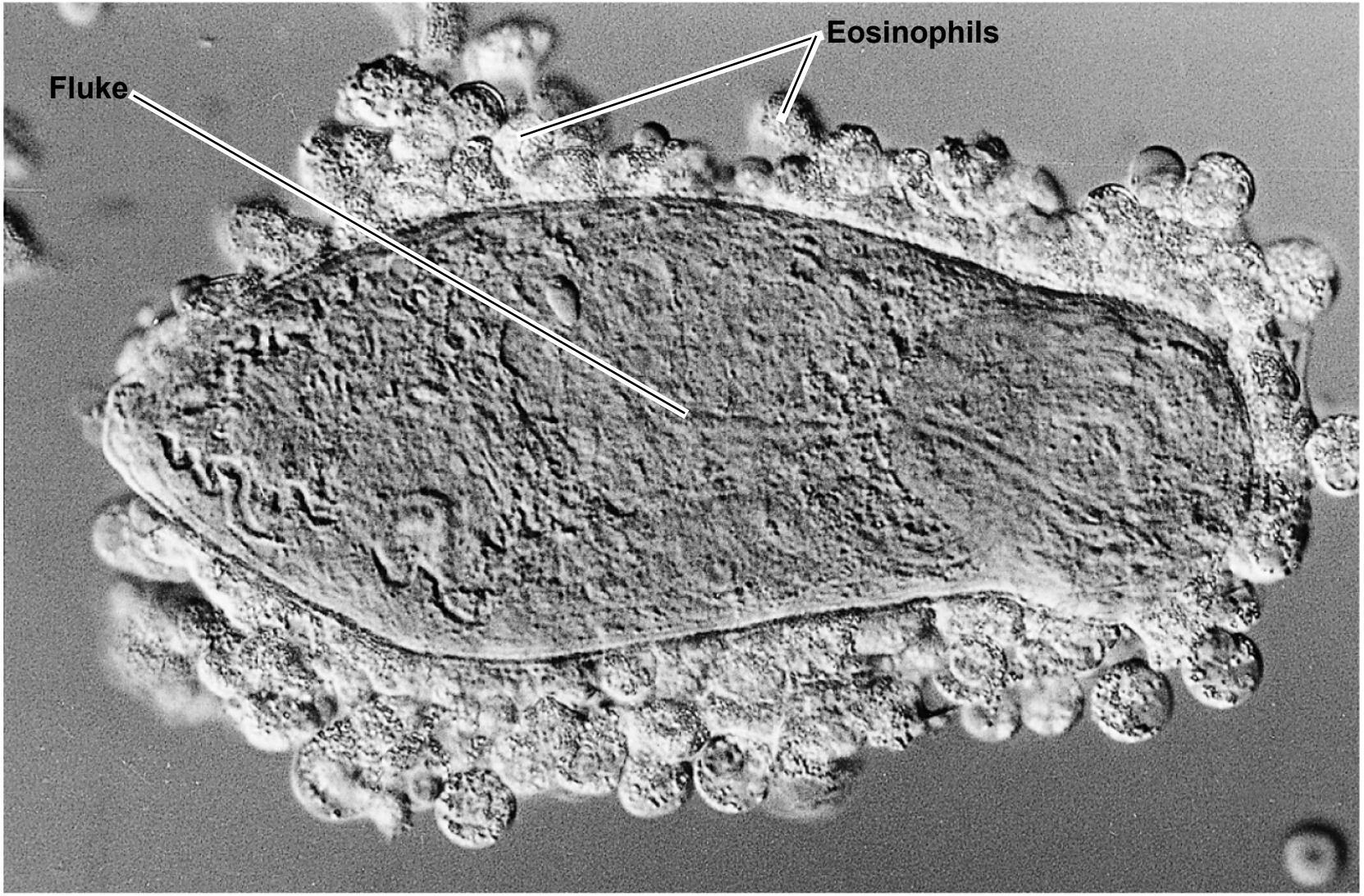
Neutrophils

- Wander about within the fluids and through the connective tissues of the body to seek and kill bacteria
  - Able to use phagocytosis, kill, and digestion the bacteria
  - Also able to create a killing zone when they identify pathogen in tissue
    - produces a cloud of anti-bacterial chemicals
    - Degranulation // lysosomes discharge into tissue fluid
    - respiratory burst – neutrophils rapidly absorb oxygen
    - toxic chemicals are created ($O_2^{\cdot -}$, H_2O_2, HClO) /// Free radical of oxygen, hydrogen peroxide, hypochlorite
    - kill more bacteria with these toxic chemicals than by phagocytosis

Eosinophils

- High concentration in the mucous membranes
- Stand guard against parasites, allergens (allergy causing agents), and other pathogens
- Kill tapeworms and roundworms by producing superoxide, hydrogen peroxide, and toxic proteins
- Promote action of basophils and mast cells
- Use phagocytosis to capture antigen-antibody complexes
- Limit action of histamine and other inflammatory chemicals
- Create a “respiratory burst” similar to kill parasites

Antibody-dependent cell-mediated cytotoxicity (ADCC).

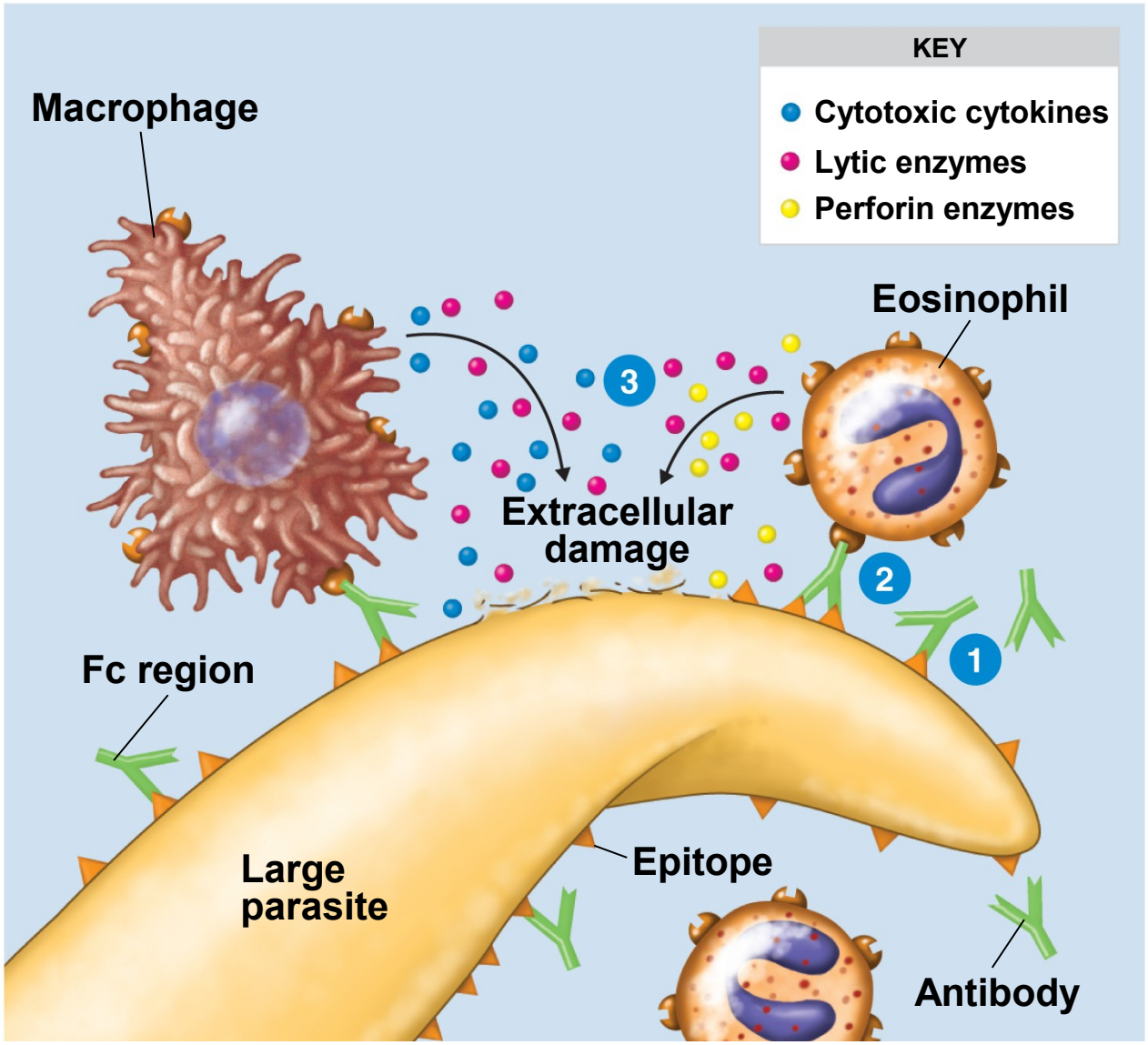


(b) **Eosinophils adhering to the larval stage of a parasitic fluke.**

SEM

20 μm

Antibody-dependent cell-mediated cytotoxicity (ADCC).



Organisms, like some parasites to large to be ingestion by phagocytic cells, must be attacked and eliminated by extracellular molecules.

Basophils Change into Mast Cells

- secrete chemicals that aid mobility and action of other leukocytes // initiates inflammation
  - leukotrienes – activate and attract neutrophils and eosinophils
  - histamine – a vasodilator which increases blood flow // peeds delivery of leukocytes to the area
  - heparin – inhibits the formation of clots // would impede leukocyte mobility
  - basophils become mast cells //// after basophil leave blood and lodge themselves into the CT throughout body
  - secrete similar substances
  - IgE become mast cell membrane receptors / antigen binding results in degranulation of mast cell.

Monocytes to Macrophage

- emigrate from the blood into the connective tissue
- change from monocyte to macrophage
- monocytes secrete many different types of cytokines which regulate inflammation and immunity while they are in blood
- macrophage system – include all the body's avidly phagocytic cells, include not only the "macrophage" (Note: neutrophils , B cells, and esinophils also phagocytic)
- wandering macrophages – actively seeking pathogens // widely distributed in loose connective tissue
- resident "fixed macrophages" = phagocytize only pathogens that come to them
    - microglia – in central nervous system
    - alveolar macrophages – in lungs
    - hepatic macrophages – in liver

What are the three lymphocytes?

- Three basic categories
- Circulating blood contains
  - 80% **T cells (cellular immunity)**
  - 15% **B cells (humoral immunity)**
  - 5% **NK cells (immune surveillance)**

Primary role 3rd line of defense

Primary role in 2nd line of defense

- Many diverse functions
- T and B lymphocytes play key role in Acquired Immunity
- NK Cells key role in 2nd line - because they perform general surveillance for cells infected with cancer or virus /// NK Cells recognize and attack infected cells – this is why we refer to NK cells as “immune surveillance”

NK Cells Provide Immune Surveillance

- Immune surveillance – a phenomenon in which natural (NK) killer cells continually patro our body looking for pathogens inside our cells (bacteria and virus).

- Natural killer (NK) cells attack and destroy infected cells then macrophage come to area and "clean up the mess"

  - Primary role to kill cells infected with virus and cancer cells

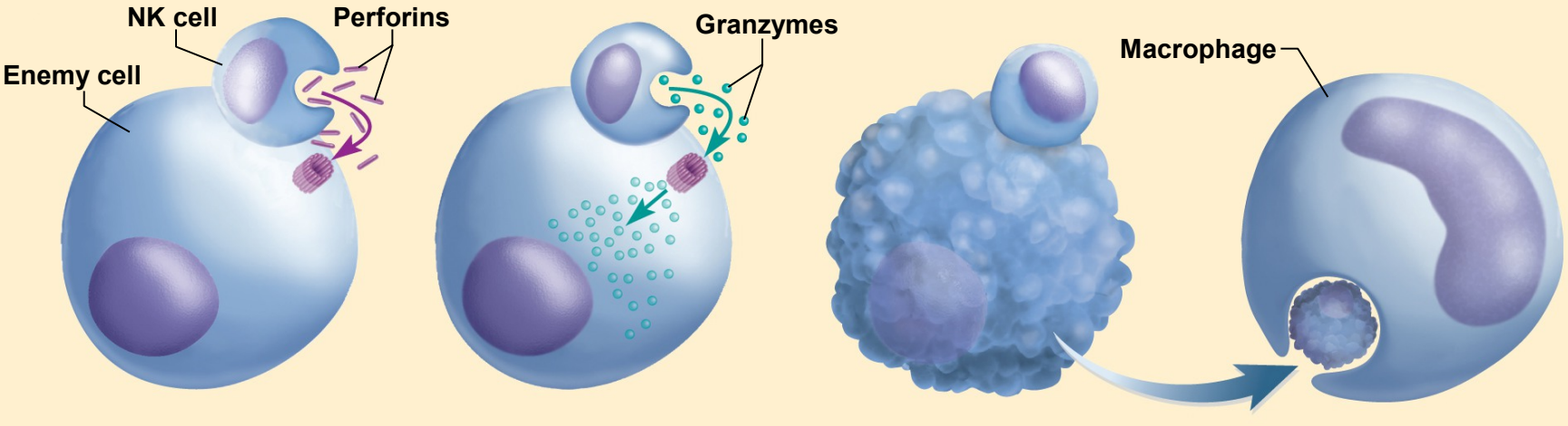
    - recognizes host cells infected with cancer or virus
    - NK cells bind to host cell
    - release proteins called perforins // cause the kiss of death!

      - polymerize a ring and create a hole in its plasma membrane

      - secrete a group of protein degrading enzymes – granzymes

      - enter through pore and degrade cellular enzymes and induce apoptosis

Action of NK cell



Note: same mechanism used by cytotoxic T cells to kill infected cells in aquired immunity!

Toll Like Receptors Are on WBC and Pathogen Associated Molecular Patterns Are on Bacteria

(This is part of the 2nd line of defense // non specific resistance)

- Toll Like Receptors (TLR) are on plasma membranes of macrophage, neutrophils, and epithelial cells lining mucous membranes (e.g. respirtaory and GI tracts) and are docking stations for "pathogen associated molecular patterns" (PAMP) located on bacteria and virus

- 11 different types of human TLR

  - Each one recognizes a different "class" of microbe (e.g. gram negative bacterial like salmonella but same receptor would also bind to all gram negative bacteria)

  - Once bound to a PAMP the TLR activates the release of inflammatory chemicals called "cytokines" from monocyte

- Macrophage, dendritic cells, endothelial cells, and lymphocytes have TLR matched to PAMP associated with bacteria and viruses

Non-Cellular Antimicrobial Proteins Also Part of the Second Line of Defense

- Two families of antimicrobial proteins inside internal tissues of body

  - **interferons** // *proteins that inhibit viral reproduction – secreted by cells infected by virus // an "alarm signal" for other cells in area to initiate anti viral mechanisms within their cells*

  - **complement system** // proteins made by liver and circulate in blood // when activated these proteins kill pathogens by forming tubes that make holes in pathogen's cell membranes // different activation methods // *provide short-term, nonspecific resistance to pathogenic bacteria and viruses /// complement functions in both in the 2nd and 3rd line of defense*

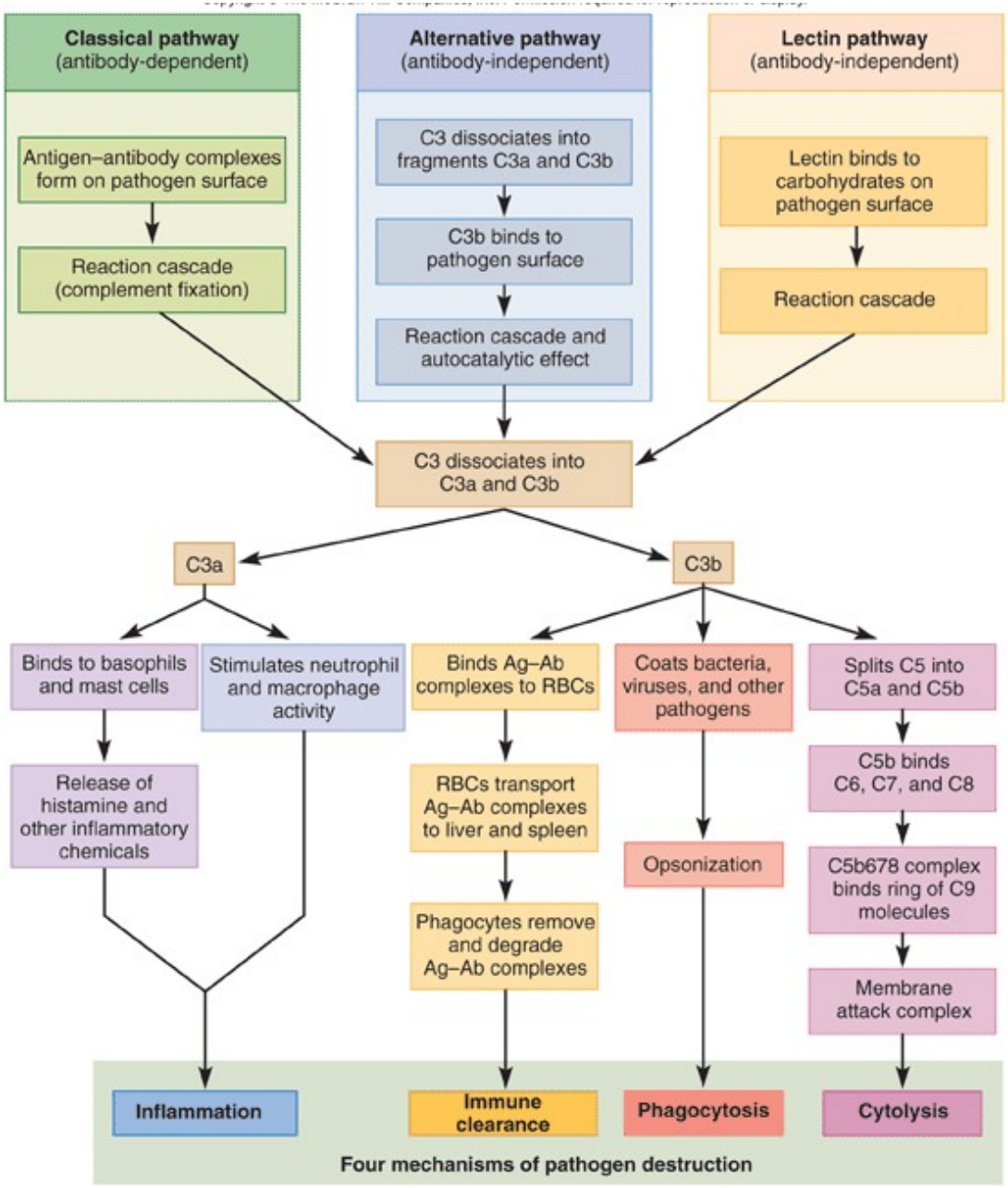
Interferon

- Secreted by certain cells infected by viruses
  - no benefit to the cell that secretes them // various cell types produce interferons (alpha, beta, gama) // including lymphocytes, macrophage, fibroblasts
  - alert neighboring cells and protect them from becoming infected
  - bind to surface receptors on neighboring cells /// activate second-messenger systems within
- Mechanism of actions:
  - alerted host cell metabolism to synthesizes various proteins that defend it from infection
    - breaks down viral genes
    - helps prevent replication of virus by “host cell”
  - signal NK cells and macrophages to migrate to area of infection /// goal is to destroy infected cell before they can liberate a swarm of newly replicated viruses /// activated NK cells to destroy malignant cells

Complement System

- A group of 30 or more globular proteins that make powerful contributions to both the second and third line of defense. / nonspecific resistance and specific immunity
- synthesized mainly by the liver
- circulate in the blood inactive
- activated by presence of the pathogen
- **C3 is the "key" starter protein in the complement system**
  - C3 must be split into C3a and C3b to activate system
  - C3a and C3b then activate separate "mechanisms"
  - three different "mechanisms" may activate complement by splitting C3 into C3a and C3b.

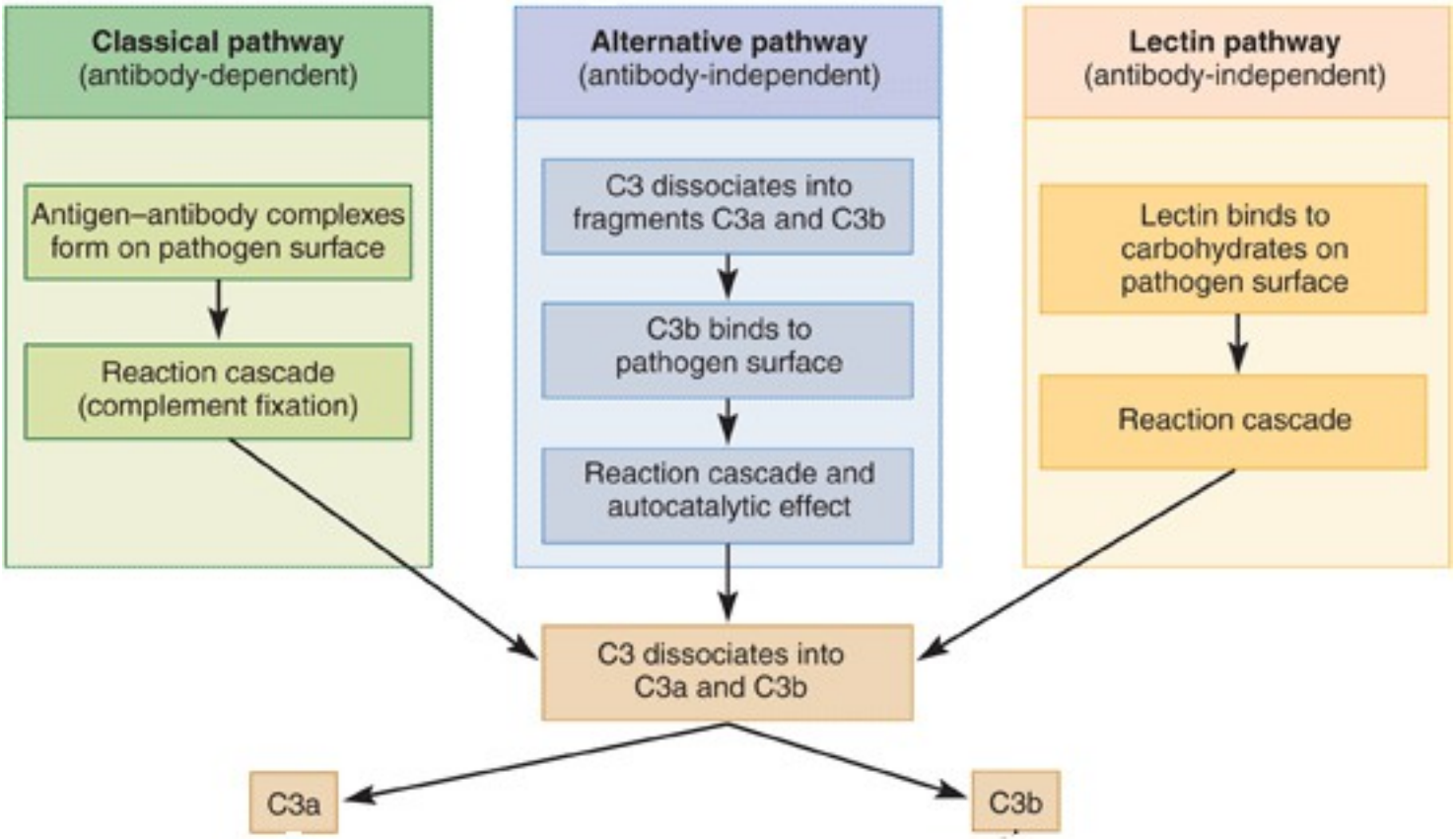
Complement Activation



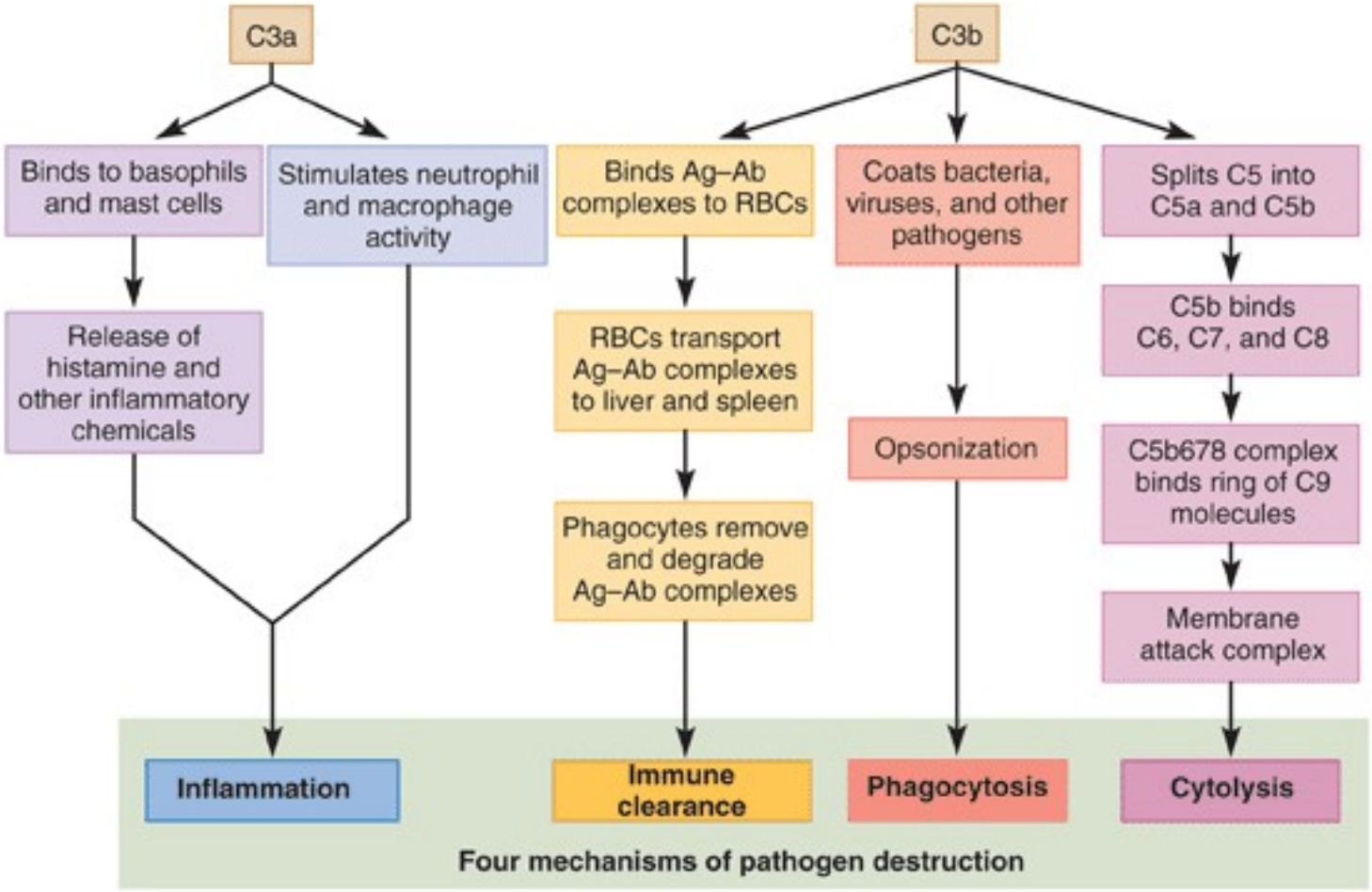
This is how complement is activated. (Three Options)

This pathway is part of specific immunity because it depends on the B Cells / plasma cells antibodies.

These pathways are part of the non specific resistance because they function independent of the B and T cells.



Complement's outcomes are a mixture of non-specific resistance and immunity:



What are the four outcomes of complement?

First of Four Outcomes By Complement
Immune Clearance

- C3b binds together antigen-antibody complexes with red blood cells
- these RBCs (with attached antigen-antibody) circulate through the liver and spleen
- macrophages of those organs strip off and destroy the Ag-Ab complexes leaving RBCs unharmed
- principal means of clearing foreign antigens from the bloodstream

Second of Four Outcomes By Complement
Phagocytosis

- neutrophils and macrophages **cannot phagocytize "naked"** bacteria, viruses, or other pathogens

- C3b assist them by **opsonization**

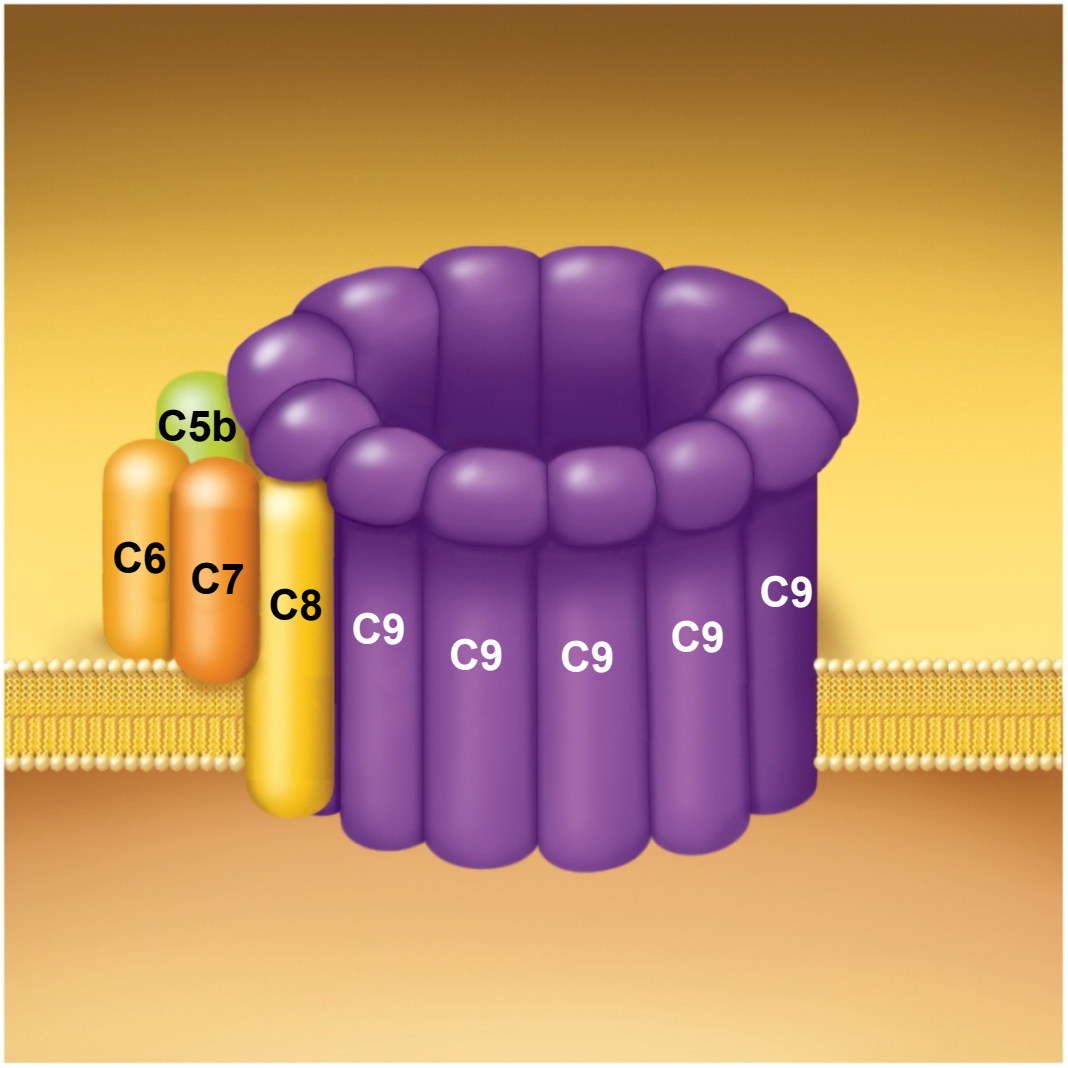
  – coats microbial cells and serves as binding sites for phagocyte attachment

  – makes the foreign cell more appetizing

Third of Four Outcomes By Complement
Cytolysis

- C3b splits other complement proteins
- bind to enemy cell
- attract more complement proteins (results in formation of the **membrane attack complex**)
  - forms a hole in the target cell
  - electrolytes leak out, water flows in rapidly, and **cell ruptures**

Membrane Attack Complex



Fourth Outcomes By Complement
Inflammation

- C3a stimulates mast cells and basophils to secrete histamine and other inflammatory chemicals / this "initiates" inflammation
- activates and attracts neutrophils and macrophages
- speed pathogen destruction in inflammation
- *Note: at the end of this presentation we will outline the individual steps of inflammation*

Inflammation

(This Is Also Part of the 2nd Line of Defense)

- Local defensive response to tissue injury of any kind, <u>including trauma and/or infection</u>

- General purposes of inflammation

  - limit spread of pathogens
  - destroy pathogens
  - remove debris from damaged tissue
  - initiate tissue repair (i.e. remember regeneration vs fibrosis)

- Four cardinal signs of inflammation

  - **redness**
  - **swelling**
  - **heat**
  - **pain**

More About the Four Cardinal Signs

- **heat** – results from hyperemia
- **redness** – due to hyperemia, and extravasated RBCs in the tissue
- **swelling** (edema) – due to increased fluid filtration from the capillaries
- **pain** – from direct injury to the nerves, pressure on the nerves from edema, stimulation of pain receptors by prostaglandins, bacterial toxins, and a kinin called **bradykinin**
- ***Note: immobilization of sore area like a joint is sometimes referred to as a "fifth event" but not a "cardinal sign"***

What are the four steps of Inflammation?

Mobilization of body defenses

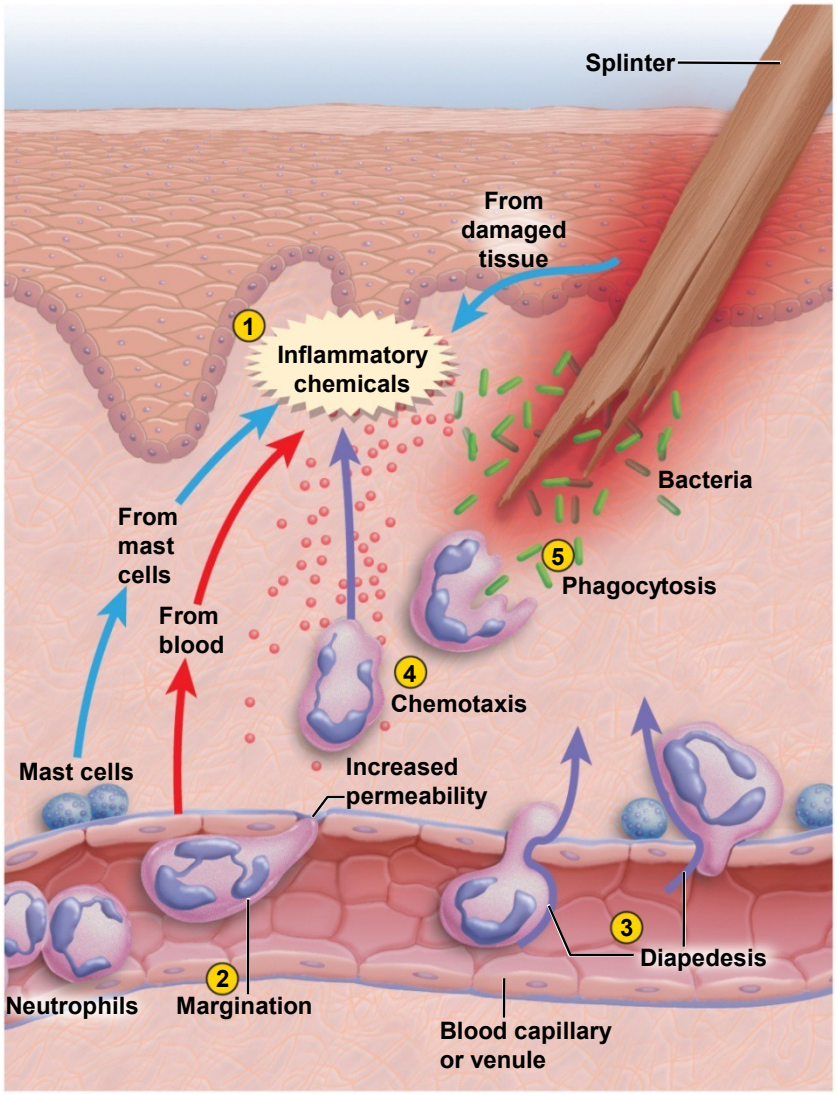
Containment and destruction of pathogen

Tissue cleanup

Tissue repair

Inflammation in a tissue causes fibroblast to migrate into the damaged area. Fibroblast secrete extracellular collagen fibers into the interstitial space (scar tissue). What is the clinical significance for someone with a chronic inflammatory response to asthma? Asthma becomes what?

Inflammation - Mobilization of Defenses



Leukocyte behavior

- **Margination**
  - **selectins** cause leukocytes to adhere to blood vessel walls
- **Diapedesis** (emigration)
  - leukocytes squeeze between endothelial cells into tissue space

Inflammation - Mobilization of Defenses

- selectins – cell-adhesion molecules made by endothelial cells that aid in the recruitment of leukocytes /// make membranes sticky and snag leukocytes

- Integrins – made by neurtrophils and interact with selectins to stop neutrophils near location of inflammation.

- margination – adhesion of the leukocytes to the vessel wall

- diapedesis (also called emigration) - leukocytes crawl through developing gaps between endothelial cells so they can enter tissue fluid

- extravasated – cells and chemicals that have left the bloodstream

Inflammation - Containment and Destruction of Pathogens

- a priority of inflammation is to prevent the pathogens from spreading throughout the body
  - fibrinogen that filters into tissue fluid clots /// forms a sticky mesh that walls off microbes
  - heparin prevents clotting at site of injury
    - pathogens are in a fluid pocket surrounded by clot
    - attacked by antibodies, phagocytes, and other defenses
- neutrophils, the chief enemy of bacteria, accumulate at the injury site within an hour /// after leaving the bloodstream, move to site of infection by chemotaxis

Inflammation - Containment and Destruction of Pathogens

- chemotaxis – attraction to chemicals such as bradykinin and leukotrienes that guide them to the injury site
- neutrophils are the “first responders” to arrive at site of infection
- kill bacteria by phagocytosis & respiratory burst (main killing force)
  - secrete cytokines for recruitment of macrophages, NK cells, and additional neutrophils
  - macrophages and T cells secrete colony-stimulating factor to stimulate leukopoiesis
    - **neutrophilia** – 5000 cells/μL to 25,000 cells/μL in bacterial infection
    - **eosinophilia** – elevated eosinophil count in allergy or parasitic infection

Inflammation
Macrophage: These are the tissue cleanup crew!

- Macrophage are the primary agents of tissue cleanup // note: fibroblast come into area to make new extracellular fibers.
  - arrive in 8 to 12 hours
  - as monocytes in blood emigrate into tissue spaces they become macrophage
    - engulf and destroy bacteria
    - engulf damaged host cells
    - engulf dead and dying neutrophils
    - Remember, macrophage are also APC

Inflammation
Why do we have edema during inflammation?

edema contributes to tissue cleanup mechanism

swelling compresses veins and reduces venous drainage

Interstitial fluid now directed into the lymphatic capillaries

- Lymphatic vesels collect and move lymh into lymph nodes where there is a high concentration of microphage /// in nodes macrophage remove bacteria, dead cells, tissue debris, and antigen presentation occur here

pus is the accumulation of dead neutrophils, bacteria, other cellular debris, and tissue fluid forms a pool of yellowish green fluid

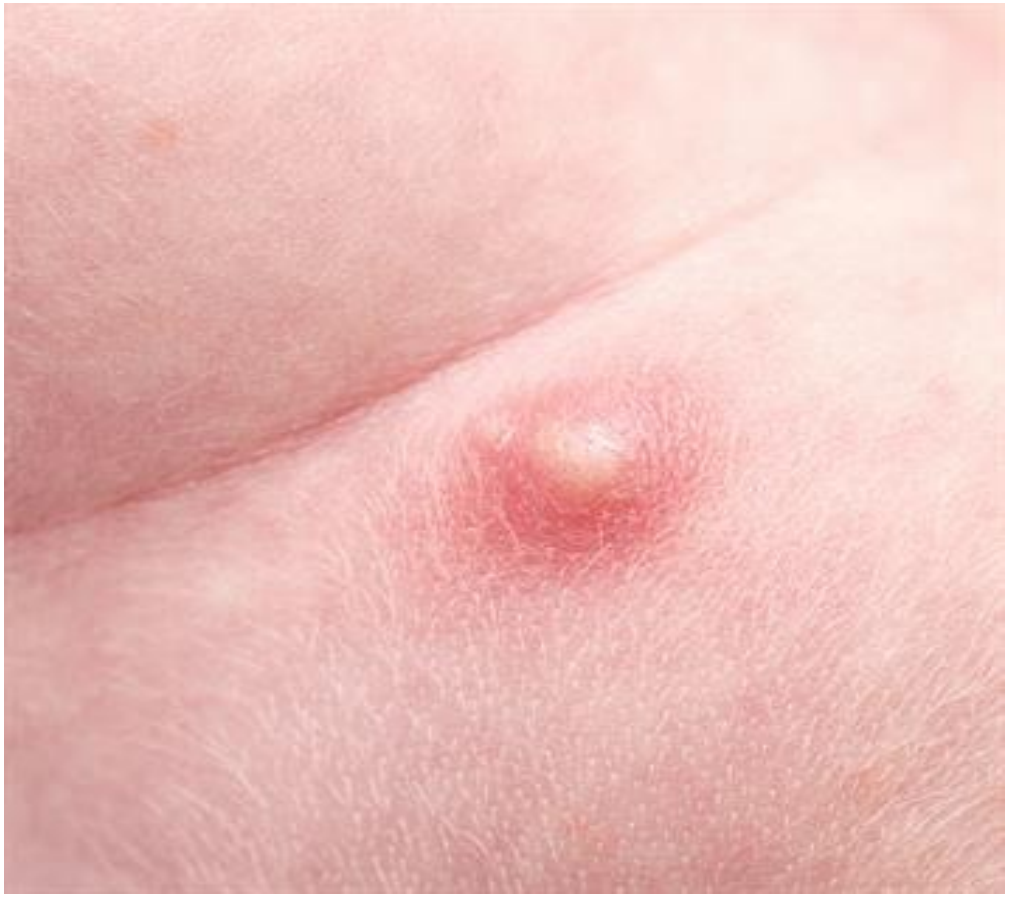
abscess – accumulation of pus in tissue surrounded by fibrin

Inflammation
Tissue Repair

- **platelet-derived growth factor** secreted by blood platelets and endothelial cells in injured area
  - stimulates fibroblasts to multiply
  - synthesize new collagen fibers
- **hyperemia** delivers oxygen, amino acids, and other necessities for protein synthesis
- increased **heat** increases metabolic rate, speeds mitosis, and tissue repair
- **fibrin clot** forms a scaffold for tissue reconstruction
- **pain** makes us limit the use of a body part so it has a chance to rest and heal.

What are the four cardinal signs of inflammation?

(What often occurs with inflammation but is not considered a cardinal sign?)



Four Cardinal Signs = Redness, Edema, Pain, Heat

What is fever?

- An abnormally elevation of body temperature
  - also called pyrexia /// febrile refers to pyrexia
  - results from trauma, infections, drug reactions, brain tumors, and other causes // e.g. inflammation

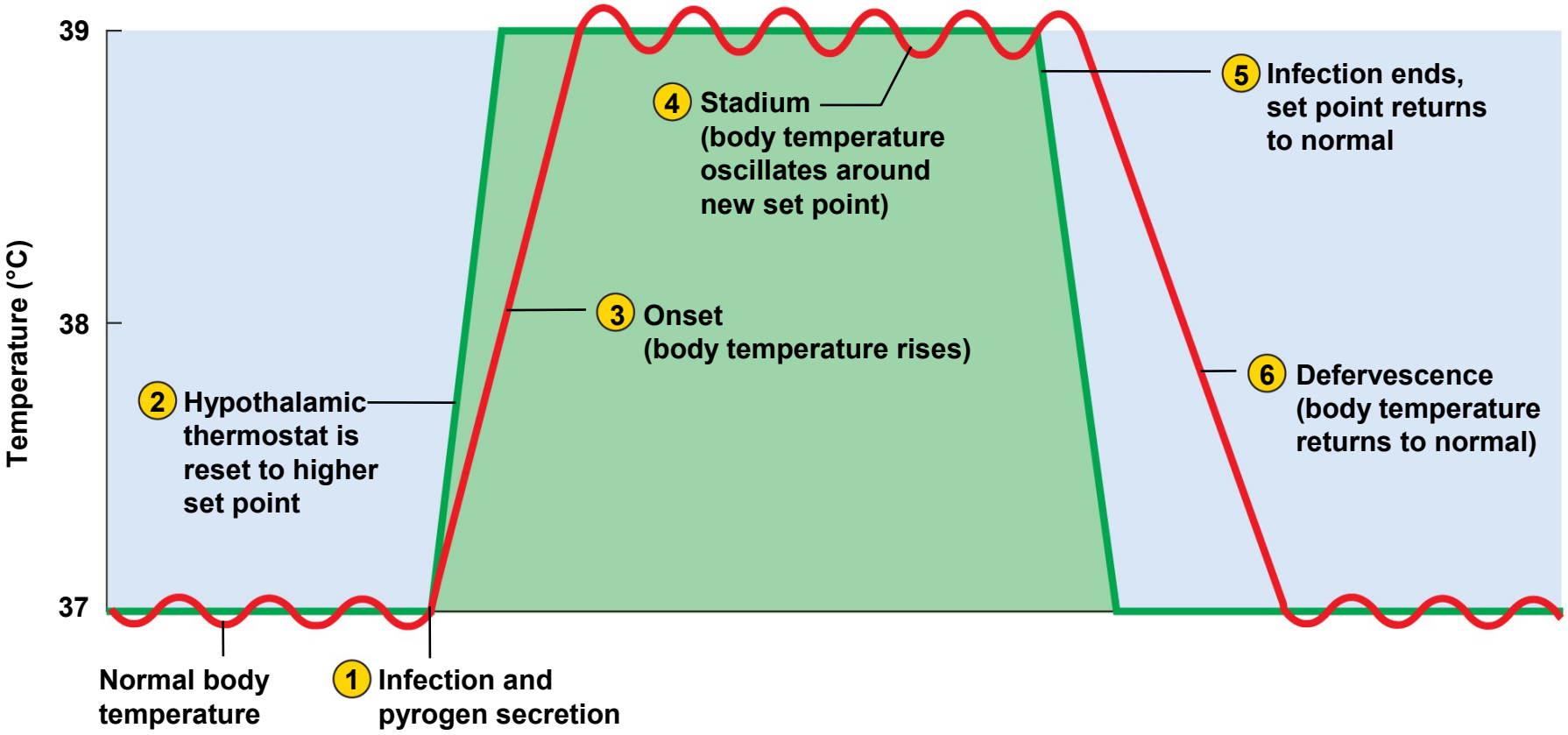
- Fever is an adaptive defense mechanism /// a moderate fever inhibits bacterial infection /// therefore, fever does more good than harm
  - promotes interferon activity
  - elevates metabolic rate and accelerates tissue repair
  - inhibits reproduction of bacteria and viruses by limiting access of iron to bacteria

- **Antipyretic** – fever-reducing medications by inhibiting PGE_2

What causes fever?

- initiation of fever by exogenous pyrogens
  - fever producing agents
  - glycolipids on bacterial and viral surfaces
- attacking neutrophils and macrophages secrete chemicals like interleukins, interferons, and other cytokines that act as endogenous pyrogens
  - stimulate neurons in the anterior hypothalamus to secrete prostaglandin E_2
  - PGE_2 raises hypothalamic set point for body temperature
- three stages of fever = onset, stadium, defervescence

Inflammation – Course of Fever



So what makes acquired immunity special?

- **The Third Line of Defense**

  - Specificity - able to recognize specific species of pathogen // recognize foreign antigen // after pathogen enters our body the immune system "captures then identifies the pathogen and activates B cells to neutralize pathogens outside of our cells and cytotoxic T cells to kill pathogen inside of our cells. (Note: the same pathogen may be in either location!) The response takes several days to a week.

  - Memory – stores record of the type of pathogen after the first exposure // if the same pathogen enters our body then there is a veru rapid response to eliminate the pathogen (minutes not days!)

What is the difference between a cellular and humoral responses?

- **Third Line of Defense is Adaptive (also called Acquired)**

  - This is a "Cellular and Humoral Response" by lymphocytes
    - Cytotoxic T cells and helper T cells provide the cellular response
    - B cells (with helper T cells) provide the humoral response

  - Activated (turned on) Cytotoxic T cells (Tc) will kill host cells infected with pathogen by giving these cells a "kiss of death". Host cell and pathogen inside cell die. After all infected host cells are killed then these specific Tc will die by apoptosis. Helper T cells required to complete cTc activation.

  - When cTc cells are activated, a memory T cell (Tm) is formed. These cells do not help with this infection but rest in lymph nodes . They will immediately recognize the same bacteria if it reenters the body at a later time and kill it before it can even form a fever upon the second exposure. Helper Tc also make memory cells /// (see next page)

What is the difference between a cellular and humoral responses?

- **Third Line of Defense continued:**

B cells are also activated (turned on) against the same pathogen and at the same time that the cTc were activated.

Activated B cells morph into plasma cells which make antibodies against the pathogen. Antibodies are in the fluids of the body and specifically bind to foreign antigen to render the pathogen harmless and tag the pathogen for destruction. Helper Tc are required to fully complete B cell activation.

When B cells are activated, memory B cells are formed.

After cTc defeat pathogen inside host cells and antibodies render pathogen harmless and tag them for destruction outside of our cells, these activated cells die but their memory cells rest in the lymph nodes for future use.

This is the immune system's cellular and humoral response to pathogens. Remember, helper T cells are required to activate both the cellular and humal responses.