TYPES AND GRADES OF IMMUNITY

TYPES OF IMMUNITY

- The physiologic function of the immune system is to confer protection against microbes and non-infectious macromolecules and there are two types of immunity.
  - Innate or Non-specific or Natural or Native immunity and
  - Adaptive or acquired or specific immunity.
- This can be further divided based on cellular and humoral responses. Both could be either natural or artificial.

INNATE IMMUNITY

- Innate immunity is the first line of defense against infectious organisms and do not depend on previous exposure i.e. the immunity existed before encountering the microbes.
- The principal components of innate immunity are
  - Physical and chemical barriers - e.g. skin , epithelial cells, tears etc.
  - Biological Barriers - Phagocytic cells (neutrophils, macrophages) and NK (natural killer) cells.
  - *Blood proteins* - complement and other mediators of inflammation (Humoral factors).
  - Cellular factors - cytokines
  - Genetic factors

PHYSICAL AND CHEMICAL BARRIERS

- Intact epithelial surfaces form physical barriers from microbial invasion. The skin, mucous membrane of the respiratory tract and gastrointestinal tract are the three main interfaces between the environment and the host.
- Skin
  - The intact skin is effective mechanical barrier.
  - Keratinized layer of skin is impermeable to microorganisms.
  - Microbial growth is less in dry environment of the skin.
  - Sweat contains lactic acid, salts that inhibit the growth of many bacteria and fungi.
  - Normal microorganisms present in the skin inhibit the growth of super infecting microorganisms.
  - Sebaceous glands liberate sebum, which protect the skin.
  - Epithelia produce some peptides, which have natural antibiotic function.
- Mucous membrane
  - Stratified squamous epithelium of mucous membrane and mucous secretions of respiratory, digestive and urogenital tract trap and prevent the entry of micro organisms.
  - Gastric secretions (HCl, enzymes etc.) with acidic pH kill the microorganisms. The epithelium of the intestine secretes antimicrobial peptides called *cryptocidins*. These natural antibiotics kill the microorganisms.
  - Acidic pH of vagina kills microorganisms.
  - *Tears* - Contain lysozymes that prevent the entry of pathogens.

BIOLOGICAL BARRIERS

Circulating Phagocytes and NK cells

- Neutrophils and macrophages (monocytes in blood) identify, ingest and destroy the microbes.
- Neutrophils are also called polymorphonuclear leukocytes and each circulated in the blood only for 6 hours.
- Neutrophils are recruited at the site of infection within a few hours of infection otherwise they undergo programmed cell death and usually phagocytosed by resident macrophages in the liver and spleen.
- Macrophages and their circulating precursors, the monocytes play important role in both innate and adaptive immunity.
- Macrophages have a single rounded nucleus and they are phagocytic, hence they are called mononuclear phagocytes.

- All the cells of the mononuclear phagocytic system arise from the bone marrow stem cells and develop into circulating *monocytes*.
- Macrophages show variation from their basic structure.
    - Connective tissue macrophages - *histiocytes,*
    - Macrophages in liver - *kupffer cells,*
    - Brain - *microglial cells,*
    - Lungs - *alveolar macrophages*,
    - kidney - *mesangial cells,*
    - Spleen and lymph node - *macrophages*.
- All they form reticuloendothelial (RE) system.

FUNCTIONS OF PHAGOCYTES

- Active recruitment of inflammatory cells at the site of infection.
- Recognition of microbes.
- Phagocytosis
- Destruction of ingested microbes.
- Production of cytokines.

ACTIVE RECRUITMENT OF INFLAMMATORY CELLS AT THESITE OF INFECTION

- Neutrophils and monocytes are normally present in blood are recruited to the site of infection by binding to adhesion molecules on endothelial cells under influence of chemo attractants produced in response to infection e.g. complement factor C5a, fibrino peptide B, platelet factor 4 etc.
- The chemotactic molecules diffuse from the site of tissue damage and form a concentration gradient.
- Neutrophils move towards the area of highest concentration i.e. the area of tissue damage. *Inflammation* is an important process for the recruitment of cells to the site of infection.
- Next there is *adherence* of circulating leukocytes to the site of infection through a multiple processes involving attachment of cells to endothelium and migration through the endothelium.
- The endothelial cells express the adhesion molecules, which are triggered by bacterial products like lipopolysaccharide or the factors (cytokines, chemokines and vesoreactive factors) released by damaged tissues or resident tissue macrophages.
- These adhesion molecules bind neutrophils and lymphocytes. The neutrophils do not bind tightly but lose their flow speed and roll over the endothelial cell surface and finally escape into tissues by *diapedesis*.
- Some important vasoreactive molecules produced during inflammation are hitamine, serotonin, kinins (bradykinin etc.) and they play important role in inflammation.
- During inflammation, first neutrophils and later monocytes followed by lymphocyte and thrombocytes (platelets) accumulate around the infectious organism.

RECOGNITION OF MICROBES BY NEUTROPHILS ANDMACROPHAGES

- Neutrophils and macrophages express surface receptors that recognize microbes in the blood and tissues and also these receptors activate cells to produce cytokines and microbicidal substances that help in phagocytosis of microbes.
- There are several classes of phagocyte receptors, which bind microbes and mediate their internalization.
- Mannose binding receptors
    - The mannose binding receptor on macrophage surface bind with the terminal mannose and fructose residues of glycoproteins and glycolipids present in the bacterial cell walls.
- Receptors for opsonins
    - Opsonins are various blood proteins, which coat the microbes and helps in phagocytosis, and this process is called ***opsonization***. Opsonic index is the ratio of the amount of opsonin in

the blood of a disease affected animal to the amount of opsonin in the blood of a healthyanimal.

- *Other phagocyte receptors*,which activate the phagocytes but do not participate directly in endocytosis: Example
- Toll like receptors (TLRs)
    - TLRs are a family of membrane proteins found in cell membranes of phagocytes and other cell types. They serve as pattern recognition receptors for a variety of microbe derived molecules and stimulate innate immune response.

PHAGOCYTOSIS OF MICROBES

- Neutrophils and macrophages ingest bound microbes and the process is called ***phagocytosis.***
- A phagocyte crawls towards a microbe, pseudopod advances around and engulf the microbe.
- The cytoplasm of the neutrophil-pseudopod contains a filamentous network of proteins, actin and myosin that determine the fluidity of the cytoplasm.
- Bindings occur between opsonin on the organism and the receptors on the neutrophil (phagocyte).
- Once firmly adhere, the organism is drawn into the cell and cytoplasm engulfs the microbe forming an enclosed vacuole called ***phagosome.***

DESTRUCTION OF INGESTED MICROBES

- Ingested particles are destroyed by the different mechanisms
    - Oxygen dependent pathway:
        - By release of reactive oxygen intermediates (ROIs), called as *respiratory burst*
        - By release of reactive nitrogen intermediates e.g. nitric oxide
    - Oxygen independent pathway:
        - By lysomomal enzymes or other hydrolytic enzymes, defensins and other cationic proteins.

RESPIRATORY BURST

- Several receptors of phagocytes (activated neutrophils and macrophages) recognize microbes, which include, TLRs, G-protein coupled receptors, F_c and C_3 receptors etc.
- Immediately after binding to foreign particles, phagocytes increase their oxygen consumption to about 100 fold and convert molecular oxygen into reactive oxygen intermediates (ROIs), which are oxidizing agents and destroy microbes. This metabolic pathway is also known as ***hexose-monophosphate shunt.***
- The primary free radical generating system is the phagocyte oxidase system. When phagocyte bind with foreign particle, the cell surface enzyme, NADPH (Nicotinamide-adenine dinucleotide phosphate) -oxidase is activated and reduced to NADP with release of electrons. These electrons are accepted bymolecular oxygen and form ROIs such as super oxide radicals where NADP acts as cofactor.

- Super oxide is dismuted enzymatically to form hydrogen peroxide (H_2O_2). Myeloperoxidase enzyme of the phagocyte catalyzes the reaction between hydrogen peroxide and intracellular halide ions to form *hypohalides* that are toxic to the bacteria. The process by which ROIs are produced is calledthe *respiratory burst.*

NITROGEN INTERMEDIATES

- Macrophages also produce nitrogen intermediates mainly nitric oxide by the action of an enzyme inducible nitric oxide synthase (NOS) on stimulation with LPS or other microbial products.
- Nitric oxide combines with hydrogen peroxide or super oxide and produce highly reactive microbicidal radicals within the phagolysosomes.

LYSOSOMAL AND OTHER PROTEOLYTIC ENZYMES

- As soon as a foreign particle is attached to the neutrophil membrane, the primary granules (orlysozymes) from the cytoplasm migrate and fuse with phagosome to form *phagolysosome*.
- The enzymes in the primary granules digest the bacterial cell wall and kill microbes.
- Lysosomal enzymes contain a mixture of proteases (e.g. elastase), lipases and many carbohydratedestroying enzymes and enzymes that act on nucleic acids.

PRODUCTION OF CYTOKINES

- Cytokines are proteins secreted by cells of immune system (both innate and adaptive) in responseto microbes or other antigens e.g. macrophages, neutrophils NK cells etc.
- The nomenclature of cytokines is often based on source of origin e.g. cytokines produced by monocytes or macrophages are called monokines and when produced by lymphocytes are calledlymphokines.
- It is known that the same protein is synthesized by lymphocytes, monocytes, and a variety of cellslike endothelial cells and some epithelial cells.
- Interleukins are cytokines that regulate the interaction between lymphocytes and other leukocytes. They have been numbered chronologically in the order of their discovery (IL-1 to IL-35).
- Function of cytokines
    - Mediators and regulators of innate immunity.
    - Mediators and regulators of adaptive immunity.
    - Stimulators of haematopoiesis and differentiation of immature leukocytes.
    - The cytokines recruit and activates leukocytes to produce systemic reaction and synthesisof effector cells and proteins that potentiate antimicrobial responses.

NATURAL KILLER CELLS (NK CELLS)

- They constitute 5-20% of mononuclear cells of blood and spleen but rare in other lymphoid organs.
- NK cells derived from blood or spleen can kill various target cells without additional activation.
- NK cells are neither T nor B-lymphocytes and do not express receptors like immunoglobulin or T cell receptors. They have activating receptors and inhibitory receptors. When both activating and inhibitory receptors are engaged, inhibitory receptors are dominant and NK cells are not activated. This mechanism prevents the killing of normal host cells. In case of infection, most often there is inhibition of class I MHC molecule expression and ligands for inhibitors receptor of NK cells are lostand they are active.
- NK cells are a subset of large lymphocytes derived from bone marrow precursors with numerouscytoplasmic granules (large granular lymphocytes).
- NK cells also recognize antibody-coated targets. The effectors functions of NK cells are to kill theinfected cells and activate macrophages to destroy the phagocytosed microbes.
- NK cells like T-cytotoxic cells have granules, which contain *perforin* (protein) that creates pores on the target cell membrane and the enzyme called *granzymes,* enter through the pores and cause apoptosis and death of target cells.
- *NK cells* kill, viruses, intracellular bacteria, and IFN-γ produced by NK cells help in phagocytosis by themacrophages. They participate in antibody dependent cell mediated cytotoxicity (ADCC).