

CYTOKINES

- 1) **Cytokines** are a diverse group of non-antibody proteins that act as mediators between cells. They were initially identified as products of immune cells that act as mediators and regulators of immune processes but many cytokines are now known to be produced by cells other than immune cells and they can have effects on non-immune cells as well. Cytokines are currently being used clinically as biological response modifiers for the treatment of various disorders. The term cytokine is a general term used to describe a large group of proteins but there are other terms that are commonly used to describe particular kinds of cytokines. These include: monokines (cytokines produced by mononuclear phagocytic cells), lymphokines (cytokines produced by activated lymphocytes, especially Th cells), interleukins (cytokines that acts as mediators between leukocytes), and chemokines (small cytokines primarily responsible for leukocyte migration).
- 2) **Cytokines function** as part of a larger inter-related system of proteins and signaling cascades, the cytokine network. These are complex interactions in which different cells can respond differently to the same cytokine depending upon other signals received by the cell. Cytokine signaling is very flexible and can induce both protective and damaging responses. One cytokine often influences the synthesis of other cytokines. They can produce cascades, or enhance or suppress production of other cytokines. In addition, they can often influence the action of other cytokines. The effects can be: **antagonistic**, **additive**, or **synergistic**.

3) Cytokines are not typically stored as preformed proteins. Rather their synthesis is initiated by gene transcription and their mRNAs are short lived. They are produced as needed in immune responses. Genes encoding cytokines can produce variants through alternative splicing to yield proteins with slightly different but biologically significant bioactivities.

4) Many individual cytokines are produced by many cell types involved in both the innate and adaptive immune response. Individual cytokines also act on many cell types (*i.e.*, they are **pleotropic**) and in many cases cytokines have similar actions (*i.e.*, they are **redundant**). Redundancy is due to the nature of the cytokine receptors.

5) Receptors for cytokines are heterodimers (sometimes heterotrimers) many of which can be grouped into families based on common structural features; one subunit is common to all members of a given family. Some examples are shown in Figure 1A (type 1) and Figure 1B (type 2).

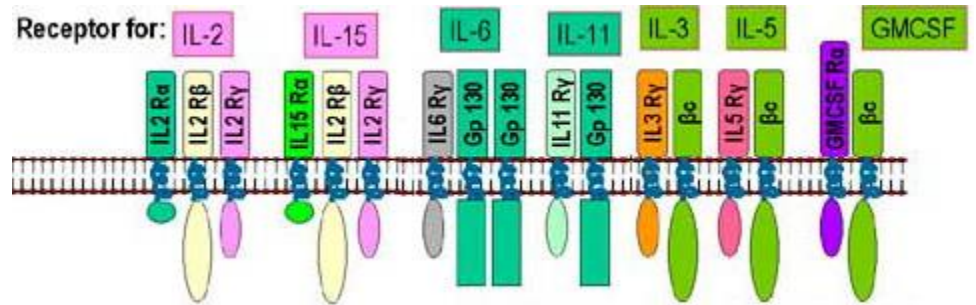


Fig A

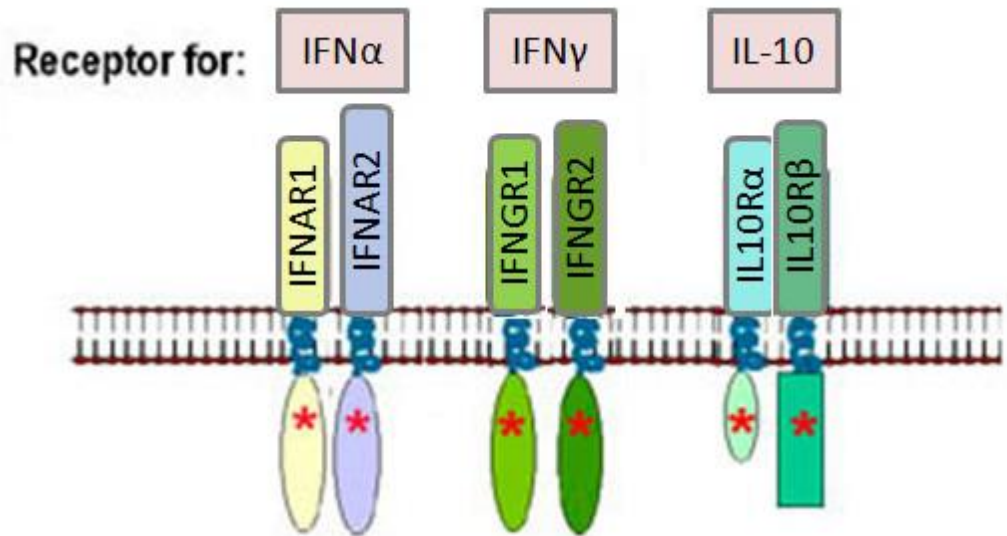


Fig B

- a) Type 1 cytokine receptors (IL-2R family) are the largest family of cytokine receptors. This family is divided into three subsets based on common components: IL2R γ , common β , and gp130 (Figure 1A). These receptors lack intrinsic protein tyrosine kinase activity. Ligand (cytokine) binding leads to receptor dimerization and initiation of intracellular signaling.
- b) Type 2 cytokine receptors (IFNR family) is denoted by conserved cysteines in the extracellular domains of the subunits. The extracellular domains also have tandem Ig-like domains characteristic of this cytokine receptor family. These receptor subunits also have intrinsic tyrosine kinase activity (denoted by the * in Figure 1B).
- c) Chemokine receptors all have seven transmembrane segments linked to GTP-binding proteins. They are selectively expressed on particular lymphocyte populations and are named based on the family of chemokines to which they bind; CCR (the CC receptor) binds CC chemokines as its ligand while the CXCR binds CXC chemokines as its ligand (chemokines naming convention will be discussed below).
- 6)** Since the subunit common to all members of the family functions in binding cytokine and in signal transduction, a receptor for one cytokine can often respond to another cytokine in the same family. Thus, an individual lacking IL-2, for example, is not adversely affected because other cytokines (IL-15, IL-7, IL-9, etc.) assume its function. Similarly, a mutation in a cytokine receptor subunit other than the one in common often has little effect. On the other hand, a mutation in the common subunit has profound effects. For example, a mutation in the gene for the IL-2R γ subunit causes human X-linked severe combined immunodeficiency (XSCID) characterized by a complete or nearly complete T and B cell defects.

7) Cytokines bind to specific receptors on target cells with high affinity and the cells that respond to a cytokine are either: 1) the same cell that secreted cytokine (autocrine); 2) a nearby cell (paracrine) or 3) a distant cell reached through the circulation (endocrine). Cellular responses to cytokines are generally slow (hours) because they require new mRNA and protein synthesis.

8) Categories of cytokines: Cytokines can be grouped into different categories based on their functions or their source but it is important to remember that because they can be produced by many different cells and act on many different cells, any attempt to categorize them will be subject to limitations.

a) Mediators of the innate immune response: Cytokines that play a major role in the innate immune system include: $\text{TNF-}\alpha$, IL-1, IL-10, IL-12, type I interferons (IFN- α and IFN- β), IFN- γ , and chemokines.

i) $\text{TNF-}\alpha$: Tumor necrosis factor alpha is produced by activated macrophages in response to microbes, especially the lipopolysaccharide (LPS) of Gram negative bacteria. It is an important mediator of acute inflammation. It mediates the recruitment of neutrophils and macrophages to sites of infection by stimulating endothelial cells to produce adhesion molecules and by producing chemokines which are chemotactic cytokines. $\text{TNF-}\alpha$ also acts on the hypothalamus to produce fever and it promotes the production of acute phase proteins.

ii) IL-1: Interleukin 1 is another inflammatory cytokine produced by activated macrophages. Its effects are similar to that of $\text{TNF-}\alpha$ and it also helps to activate T cells.

iii) IL-10: Interleukin 10 is produced by activated macrophages and Th2 cells. It is predominantly an inhibitory cytokine. It inhibits production of IFN- γ by Th1 cells, which shifts immune responses toward a Th2 type. It also inhibits cytokine production by activated macrophages and the expression of class II MHC and co-stimulatory molecules on macrophages, resulting in a dampening of immune responses.

iv) IL-12: Interleukin 12 is produced by activated macrophages and dendritic cells. It stimulates the production of IFN- γ and induces the differentiation of Th cells to become Th1 cells. In addition, it enhances the cytolytic functions of Tc and NK cells.

v) Type I interferons: Type I interferons (IFN- α and IFN- β) are produced by many cell types and they function to inhibit viral replication in cells. They also increase expression of class I MHC molecules on cells making them more susceptible to killing by CTLs. Type I interferons also activate NK cells.

vi) INF- γ : Interferon gamma is an important cytokine produced by primarily by Th1 cells, although it can also be produced by Tc and NK cells to a lesser extent. It has numerous functions in both the innate and adaptive immune systems as depicted in Figure 2.

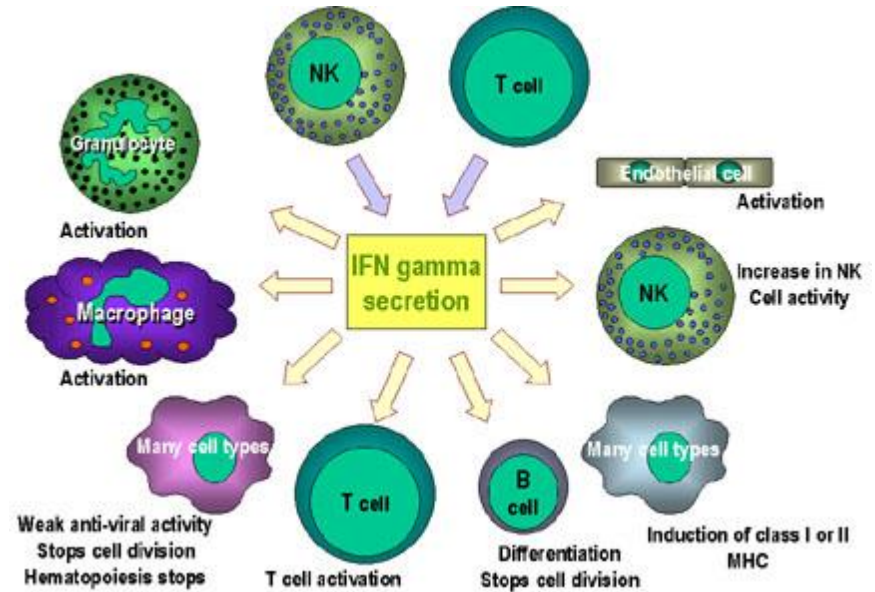


Fig 2:

vii) Chemokines: Chemokines are chemotactic cytokines produced by many kinds of leukocytes and other cell types. They represent a large family of molecules that function to recruit leukocytes to sites of infection and play a role in lymphocyte trafficking by determining which cells will cross the epithelium and where they are directed to go. There are four families of chemokines based on spacing of conserved cysteine. Two examples are the α -chemokines which have a CXC structure (two cysteines with a different amino acid in between) and the β -chemokines which have a CC structure (two neighboring cysteines). Individual chemokines (within the same family) often bind more than one receptor.

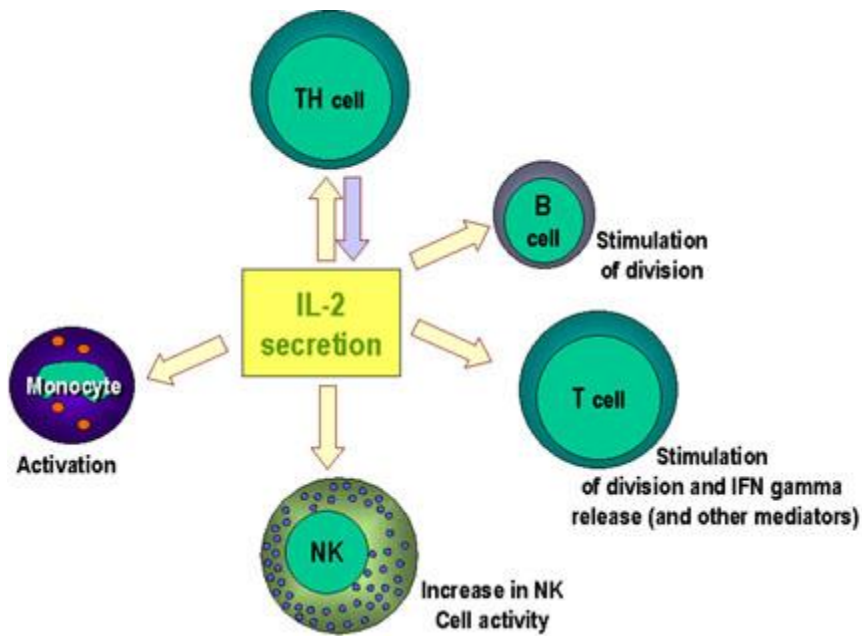


Fig 3

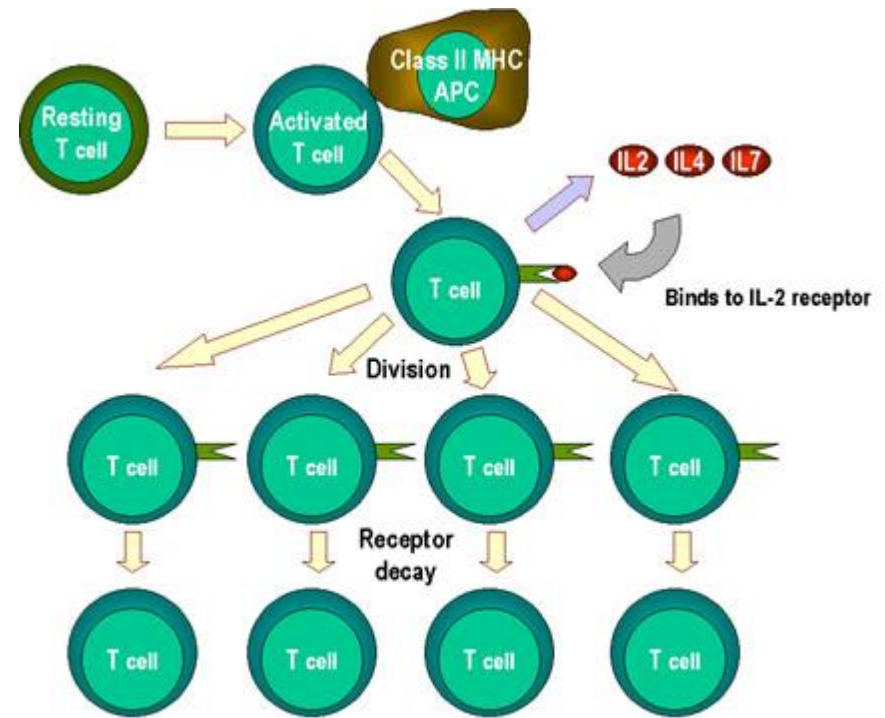


Fig 4

b) Mediators of the adaptive immune response: Cytokines that play a major role in the adaptive immune system include: IL-2, IL-4, IL-5, TGF- β , IL-10 and IFN- γ .

i) IL-2: Interleukin 2 is produced by Th cells, although it can also be produced by Tc cells to a lesser extent. It is the major growth factor for T cells. It also promotes the growth of B cells and can activate NK cells and monocytes as depicted in Figure 3. IL-2 acts on T cells in an autocrine fashion. Activation of T cells results in expression of IL-2R and the production of IL-2. The IL-2 binds to the IL-R and promotes cell division. When the T cells are no longer being stimulated by antigen, the IL-2R will eventually decay and the proliferative phase ends Figure 4.

ii) IL-4: Interleukin 4 is produced by macrophages and Th2 cells. It stimulates the development of Th2 cells from naïve Th cells and it promotes the growth of differentiated Th2 cells resulting in the production of an antibody response. It also stimulates Ig class switching to the IgE isotype.

iii) IL-5: Interleukin 5 is produced by Th2 cells and it functions to promote the growth and differentiation of B cells and eosinophils. It also activates mature eosinophils.

iv) TGF- β : Transforming growth factor beta is produced by T cells and many other cell types. It is primarily an inhibitory cytokine. It inhibits the proliferation of T cells and the activation of macrophages. It also acts on PMNs and endothelial cells to block the effects of pro-inflammatory cytokines

c) Stimulators of hematopoiesis: Some cytokines stimulate the differentiation of hematopoietic cells. These include GM-CSF which promotes the differentiation of bone marrow progenitors, M-CSF, which promotes growth and differentiation of progenitors into monocytes and macrophages and G-CSF (also known as pluripoyetin), which promotes production of PMNs.

d) Interleukin 17: IL-17 is proinflammatory cytokine approximately 150 amino acids long. The IL-17 family includes six members which share sequence homology but differential tissue expression. IL-17 is produced by Th17 cells and its overexpression has been associated with autoimmune disease including multiple sclerosis, rheumatoid arthritis, and inflammatory bowel disease.

9) Cytokine networks: Although the focus of most research and this paper has been on the production and action of cytokines on cells of the immune system, it is important to remember that many of them have effects on other cells and organ systems. A schematic diagram showing some of the interactions in the cytokine network is presented in Figure 5. In fact, the cytokine network is quite complex and represents a series of overlapping and inter-related connections amongst cytokines. Within this network, one cytokine may induce or suppress its own synthesis, induce or suppress the synthesis of other cytokines, induce or suppress synthesis of cytokine receptors (both its own and other cytokine Rs), and antagonize or synergize with other cytokines.

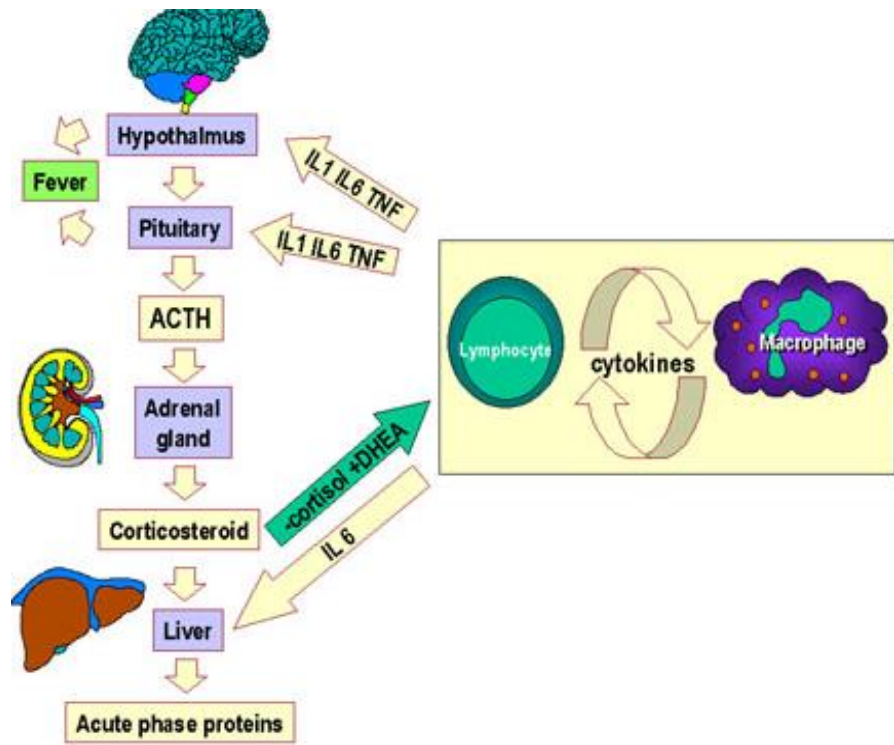


Figure 5A

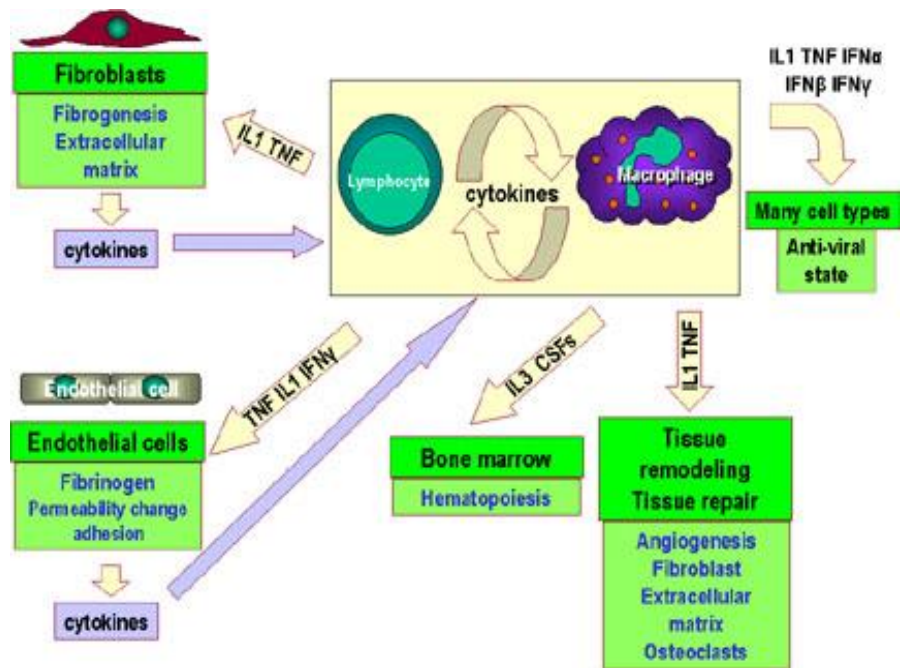


Figure 5B

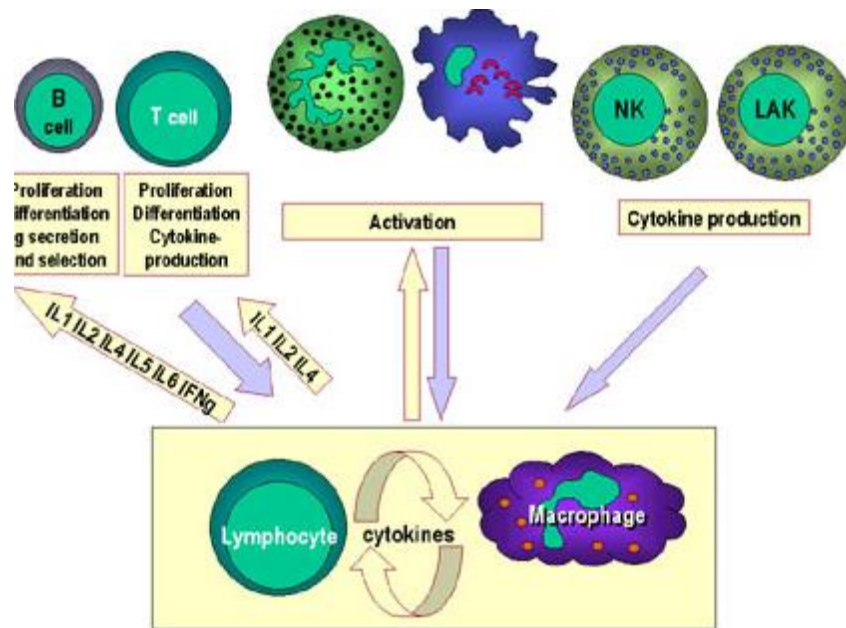


Figure 5C