CYTOKINES

The course of immunologic and inflammatory pathways is mediated by several hormones like soluble substances that are secreted by the concerned cell types. These substances have been given a general term: the **cytokine**.

Cytokines are low molecular weight, soluble proteins that are produced in response to an antigen and function as chemical messengers for regulating the innate and adaptive immune systems. They are produced by virtually all cells involved in innate and adaptive immunity, but especially by T helper lymphocytes. The activation of cytokine-producing cells triggers them to synthesize and secrete their cytokines. The cytokines, in turn, are then able to bind to specific cytokine receptors on other cells of the immune system and influence their activity: this includes both enhancing and suppressing responses. Cytokine is a general name; other names include **lymphokine** (cytokines made by lymphocytes), **monokine** (cytokines made by monocytes), **chemokine** (cytokines with chemotactic activities), and **interleukin** (cytokines made by one leukocyte and acting on other leukocytes). Cytokines may act on the cells that secrete them (**autocrine action**), on nearby cells (**paracrine action**), or in some instances on distant cells (**endocrine action**).

It is common for different cell types to secrete the same cytokine or for a single cytokine to act on several different cell types (**pleiotropy**; see the table below.) Cytokines are **redundant** in their activity, meaning similar functions can be stimulated by different cytokines. Cytokines are often produced in a **cascade**, as one cytokine stimulates its target cells to make additional cytokines. Cytokines can also act **synergistically** (two or more cytokines acting together) or **antagonistically** (cytokines causing opposing activities).

There are three functional categories of cytokines depending on whether they
(a) Regulate innate immune responses
(b) Influence adaptive immune responses
(c) Stimulate haematopoiesis

Their short half life, low plasma concentrations, pleiotropy, and redundancy all complicated the isolation and characterization of cytokines. Searches for new cytokines are now often conducted at the DNA level, identifying genes similar to known cytokine genes.

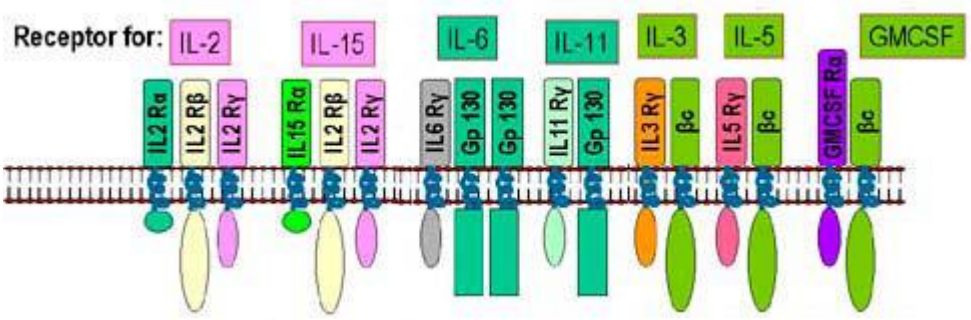
Cytokine Activities.

The largest group of cytokines stimulates immune cell proliferation and differentiation. This group includes Interleukin 1 (IL-1), which activates T cells; IL-2, which stimulates proliferation of antigen-activated T and B cells; IL-4, IL-5, and IL-6, which stimulate proliferation and differentiation of B cells; Interferon gamma (IFN□), which activates macrophages; and IL-3, IL-7 and Granulocyte Monocyte Colony-Stimulating Factor (GM-CSF), which stimulate hematopoiesis.

Cytokine Receptors

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. Cytokines act on

their target cells by binding specific membrane receptors. The receptors and their corresponding cytokines have been divided into several families based on their structure and activities. **Hematopoietin family** Examples are receptors for IL-2 through IL-7 and GM-CSF. **Interferon family** receptors include the receptors for IFN□, IFN□, and IFN□. **Tumor Necrosis Factor family** receptors have four, they include receptors for soluble TNF□ and TNF□ as well as membrane-bound CD40 (important for B cell and macrophage activation) **Chemokine family** receptors have seven transmembrane helices and interact with G protein. This family includes receptors for IL-8, MIP-1 and RANTES.



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.

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: TNF-α, IL-1, IL-10, IL-12, type I interferons (IFN-α and IFN-β), IFN-γ, and chemokines.

   i) TNF-α: Tumor necrosis factor alpha is produced by activated macrophages is 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. TNF- α 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 TNF-α 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.

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.

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.

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 hematopoesis: 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 pluripoietin), which promotes production of PMNs.

d) Interleukin 17: IL-17 is proinflammatory cytokine approximately 150 amino acids long. The IL-17 family includes sex 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.

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.

Selected Immune Cytokines and Their Activities*			
Cytokine	**Producing Cell**	**Target Cell**	**Function****
GM-CSF	Th cells	progenitor cells	growth and differentiation of monocytes and DC
IL-1□ IL-1□	monocytes macrophages B cells DC	Th cells	co-stimulation
		B cells	maturation and proliferation
		NK cells	activation
		Various	inflammation, acute phase response, fever
IL-2	Th1 cells	activated T and B cells, NK cells	growth, proliferation, activation
IL-3	Th cells NK cells	stem cells	growth and differentiation
		mast cells	growth and histamine release
IL-4	Th2 cells	activated B cells	proliferation and differentiation IgG_1 and IgE synthesis
		Macrophages	MHC Class II
		T cells	Proliferation

IL-5	Th2 cells	activated B cells	proliferation and differentiation IgA synthesis
IL-6	monocytes macrophages Th2 cells stromal cells	activated B cells	differentiation into plasma cells
		plasma cells	antibody secretion
		stem cells	Differentiation
		Various	acute phase response
IL-7	marrow stroma thymus stroma	stem cells	differentiation into progenitor B and T cells
IL-8	macrophages endothelial cells	Neutrophils	Chemotaxis
IL-10	Th2 cells	Macrophages	*cytokine production*
		B cells	Activation
IL-12	macrophages B cells	activated Tc cells	differentiation into CTL (with IL-2)
		NK cells	Activation
IFN-□	leukocytes	Various	*viral replication* MHC I expression
IFN-□	fibroblasts	Various	*viral replication* MHC I expression
IFN-□	Th1 cells, Tc cells, NK cells	Various	*Viral replication*
		Macrophages	MHC expression
		activated B cells	Ig class switch to IgG_{2a}
		Th2 cells	*Proliferation*
		Macrophages	pathogen elimination
TGF-□	T cells, monocytes	monocytes, macrophages	Chemotaxis
		activated macrophages	IL-1 synthesis
		activated B cells	IgA synthesis
		Various	*Proliferation*
TNF-□	Th1 and Tc cells	Phagocytes	phagocytosis, NO production
		tumor cells	cell death

Helper T cells have two important functions: to stimulate cellular immunity and inflammation, and to stimulate B cells to produce antibody. Two functionally distinct subsets of T cells secrete cytokines which promote these different activities. Th1 cells produce IL-2, IFN□, and TNF□, which activate Tc and macrophages to stimulate cellular immunity and inflammation. Th1 cells also secrete IL-3 and GM-CSF to stimulate the bone marrow to produce more leukocytes. Th2 cells secrete IL-4, IL-5, IL-6, and IL-10, which stimulate antibody production by B cells.

T cells are initially activated as Th0 cells, which produce IL-2, IL-4 and IFN□. The nearby cytokine environment then influences differentiation into Th1 or Th2 cells. IL-4 stimulates Th2 activity and suppresses Th1 activity, while IL-12 promotes Th1 activities. Th1 and Th2 cytokines are antagonistic in activity. Th1 cytokine IFN□ inhibits proliferation of Th2 cells, while IFN□ and IL-2 stimulate B cells to secrete IgG_{2a} and inhibit secretion of IgG_1 and IgE. Th2 cytokine IL-10 inhibits Th1 secretion of IFN□ and IL-2; it also suppresses Class II MHC expression and production of bacterial killing molecules and inflammatory cytokines by macrophages. IL-4 stimulates B cells to secrete IgE and IgG_1. The balance between Th1 and Th2 activity may steer the immune response in the direction of cell-mediated or humoral immunity.

Cytokine activity can be blocked by **antagonists**, molecules which bind cytokines or their receptors. IL-1 has a specific antagonist that blocks binding of IL-1□ and IL-1□ to their receptor. During immune responses, fragments of membrane receptors may be shed and then compete for cytokine binding. Microbes also influence cytokine activities. For example, Vaccinia virus (Smallpox and Cowpox) encodes soluble molecules which bind IFN□, while Epstein-Barr virus (Infectious Mononucleosis) encodes a molecule homologous to IL-10 that suppresses immune function in the host.

Practice Quiz

Pick the one BEST answer for each question by clicking on the letter of the correct choice.

1. Cytokines may exhibit _____ action, signaling the cells that produce them.
a. antagonistic
b. autocrine
c. endocrine
d. paracrine.
e. synergistic

2. Cytokines are NOT
a. antigen specific.
b. capable of activating more than one cell type.
c. made by lymphocytes.
d. small protein molecules.
e. synthesized de novo in response to antigen or other cytokines.

3. Several cytokines may have the same effect on the cells they bind. This is an example of
a. a cascade.
b. antagonism.
c. pleiotropism.
d. redundancy.
e. synergy.

4. Interferons
a. activate B cells to make virus-specific antibodies.
b. are Th2 cytokines.
c. are virus proteins that interfere with activation of cytotoxic T cells.
d. block virus infection of host cells.
e. inhibit virus replication by infected cells.

5. A cytokine can do all of the following EXCEPT
a. bind to receptors which do not share cytokine-binding subunits.
b. bind to its specific receptor on the same cell that produced it.
c. bind to receptor antagonists produced by pathogenic viruses.
d. compete with other cytokines whose receptors share signal-transducing subunits
e. upregulate (increase) synthesis of high affinity subunits for its receptor.

6. Members of a cytokine receptor family
a. all bind the same cytokines.
b. are grouped together because they share antigen specificity
c. are often found on the same cells
d. are similar in protein structure and sometimes in regions of amino acid sequence.
e. are specific for cytokines produced by a single cell type

7. Cytokines are NOT
a. able to inhibit the function of other cytokines.
b. able to stimulate the synthesis of other cytokines.

c. produced by more than one cell type.
d. small protein molecules.
e. stored in the cell for quick release.