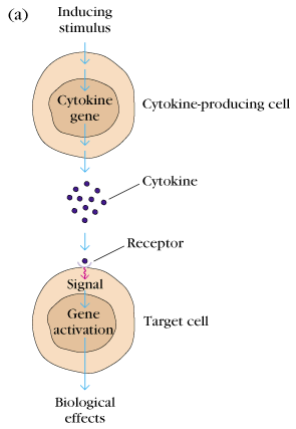
Chapter 13: Cytokines

Definition: secreted, low-molecular-weight proteins that regulate the nature, intensity and duration of the immune response by exerting a variety of effects on lymphocytes and/or other cells.

- Cytokines bind to specific **receptors** on target cells.

- Originally were called **lymphokines** because they were initially thought to be produced only by lymphocytes. Then **monokines** because they were secreted by monocytes and macrophages. Then **interleukin** because they are produced by some leukocytes and affect other leukocytes. The term "**cytokine**" is now used more widely and covers all of the above.

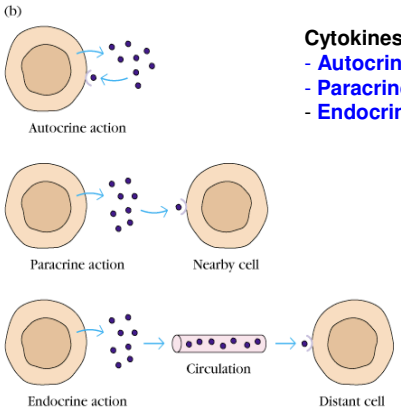
- Don't forget **chemokines**, they are also considered cytokines.



- **Cytokines act only on cells bearing specific receptors.**

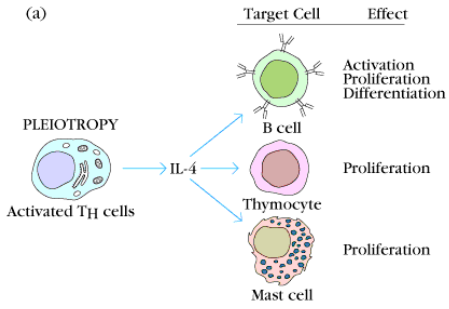
- **Expression of cytokines and their receptors is highly regulated.**

- **E.g. IL-2 receptor**

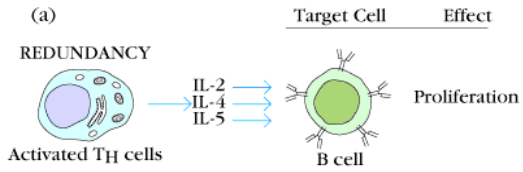


Cytokines can act in an:
- **Autocrine (same cell),**
- **Paracrine (close proximity)**
- **Endocrine (long distance)**

1. Cytokines are pleiotropic … one cytokine can have different effects on different cells.

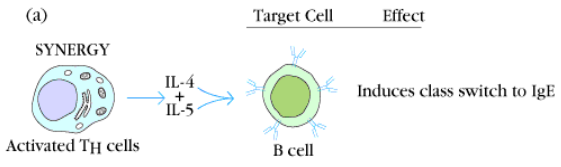


2. Cytokines can be redundant … different cytokines can have the same effects.

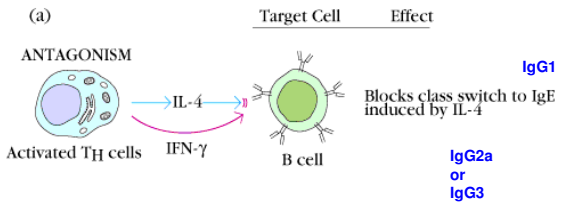


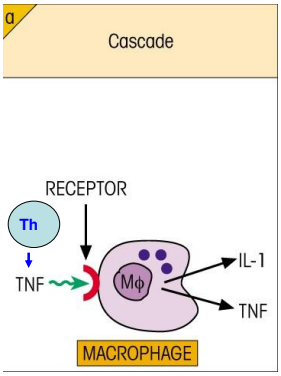
Kuby Fig 12-2a

3. Cytokines can synergize with each other.



4. Cytokines can antagonize each other.





5. Cascade effect, cytokines can stimulate the production of other cytokines.

Figure 10.5a

6. Cytokines can influence the expression of cytokine receptors.

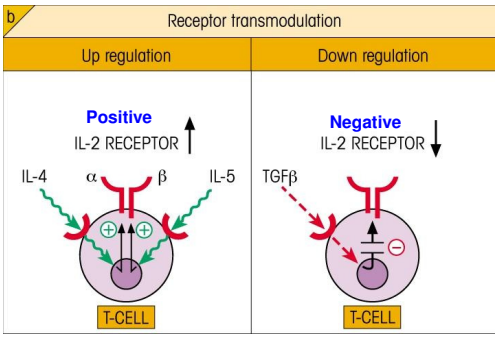


Figure 10.5b

7. Cytokines play key roles in regulating hematopoiesis, innate immunity and acquired immunity.

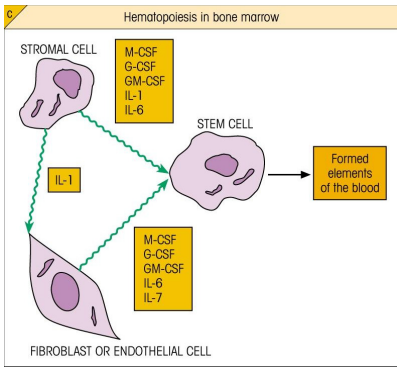
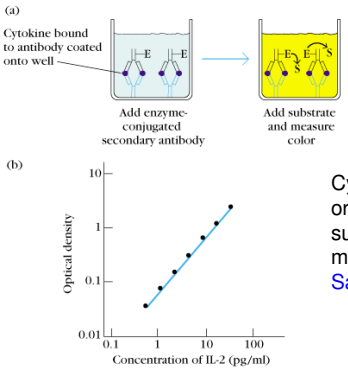


Figure 10.4c

SO…cytokines can have many effects, depending on:

- **the target cell**
- **the state of differentiation/activation of the target cell**
- **the presence or absence of other cytokines**

Sandwich ELISA



Cytokine levels in serum or in tissue culture supernatants can be measured with a Sandwich ELISA assay.

There are many cytokines, including...

IL-1	IL-2	IL-3	IL-4
IL-5	IL-6	IL-7	IL-8
IL-9	IL-10	IL-11	IL-12
IL-13	IL-15	IL-16	IL-17
IL-18	IL-19	IL-20	IL-21
IL-22	IL-23		
IFN-α	IFN-β	IFN-γ	
TNF-α	TNF-β		
TGF-β1			
M-CSF	G-CSF	GM-CSF	

Four Structural Families

- Hematopoietin Family (IL-2, IL-4)
- Interferon Family (IFN-α, β, γ)
- Chemokine Family
- Tumor necrosis family

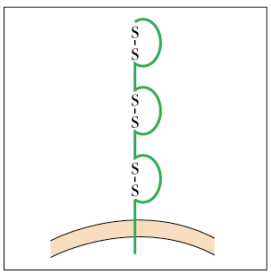
Best way to learn about cytokines…. is by their action !!!

Cytokines and Immune Responses
Proinflammatory cytokines
TNF
IL-1
IL-6
chemokines (many)
Antiinflammatory cytokines
IL-10
IL-1ra
TGF-β
Inhibition of virus replication
IFN-α, -β
Macrophage-activating cytokines
IFN-γ
B cell-activating cytokines
IL-4
IL-5
IL-6
IL-21
T cell-activating cytokines
IL-2
IL-4
IL-12
IFN-γ
Eosinophil- and/or mast cell-activating cytokines
IL-3
IL-4
IL-13
IL-5

Based on structural homology, there are six major cytokine receptor families:

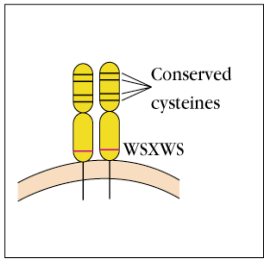
- **Ig superfamily receptors**
- **Interferon receptors**
- **TNF receptor superfamily**
- **Chemokine receptors**
- **TGF receptor family**
- **Hematopoietin receptors (Cytokine receptor superfamily)**

RECEPTOR FAMILY	LIGANDS
(a) Immunoglobulin superfamily receptors	
	IL-1
	M-CSF
	C-Kit



Kuby Fig 12-6a

(b) Class I cytokine receptors (hematopoietin)

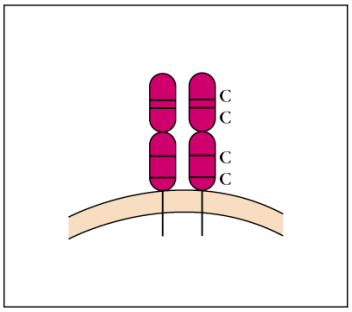


LIGANDS

IL-2	IL-13
IL-3	IL-15
IL-4	GM-CSF
IL-5	G-CSF
IL-6	OSM
IL-7	LIF
IL-9	CNTF
IL-11	Growth hormone
IL-12	Prolactin

Kuby Fig 12-6b

(c) Class II cytokine receptors
(interferon)

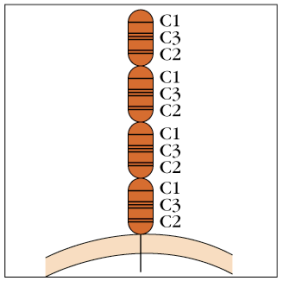


LIGANDS

IFN-α
IFN-β
IFN-γ
IL-10

Kuby Fig 12-6c

(d) TNF receptors

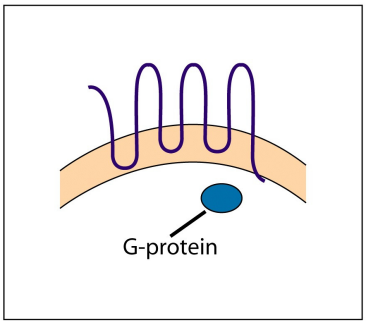


LIGANDS

TNF-α
TNF-β
CD40
Nerve growth factor (NGF)
FAS

Fig 12-6d

(e) Chemokine receptors

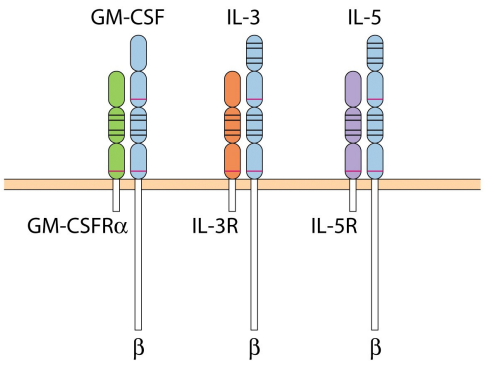


LIGANDS

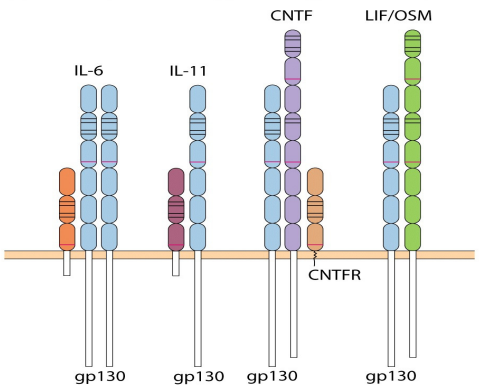
IL-8
RANTES
MIP-1
PF4
MCAF
NAP-2

Three subfamilies of the class I cytokine receptor family (hematopoietin)

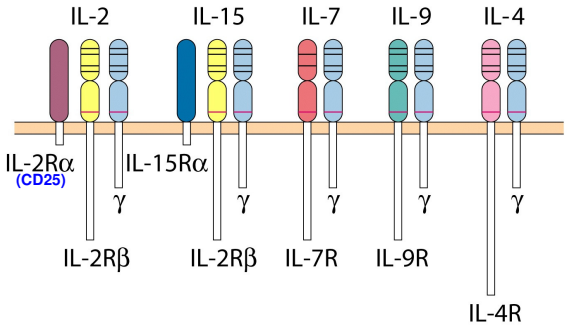
(a) GM-CSF receptor subfamily (common β subunit)



(b) IL-6 Receptor subfamily (common gp130 subunit)



(c) IL-2 receptor subfamily (common γ subunit)

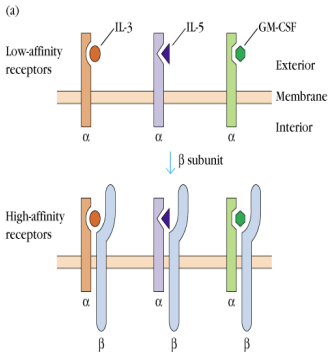


SHARED CYTOKINE RECEPTORS SUBUNITS

Shared Cytokine-Receptor Subunits	
Shared receptor chain	**Cytokines recognized**
γ_c	IL-2, -4, -7, -9, -15, -21
IL-2Rβ	IL-2, IL-15
IL-4Rα	IL-4, -13
IL-13Rα1	IL-4, -13
β_c	IL-3, -5, GM-CSF
gp130	IL-6, -11, -27, -31, LIF, OSM, CNTF, CT-1, CLC
IL-12Rβ1	IL-12, -23
IL-10R2	IL-10, -22
IL-20R2	IL-20, -19, -24
IL-22R	IL-22, -24, -20

Cytokine receptors

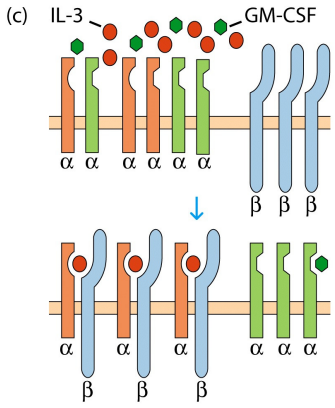
- Sharing of signal transducing molecules explains the redundancy and antagonism exhibited by some cytokines



1. **Cytokine binds to alpha subunit.**
2. **Association with beta subunit.**
3. **Signal transduction through the beta subunit**

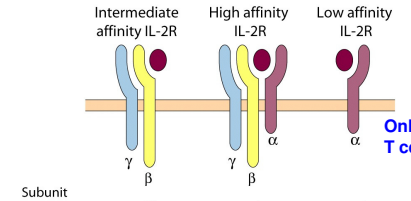
Competition of ligand-binding chains of different receptors for a common subunit.

This can results in antagonistic effects between cytokines



IL-2 Receptor

- Composed of 3 subunits: α, β, and γ chains
- IL-2 receptor is present in 3 forms: low, medium, and high affinity
- The low affinity (monomeric, IL-2Rα), medium affinity (dimeric, IL-2Rαβ), and high affinity (trimeric, IL-2Rαβγ)
- Binding component: α chains
- Transducing components: β and γ chains.



Only in activated T cells

Subunit composition:	IL-2Rβ IL-2Rγ	IL-2Rα IL-2Rβ IL-2Rγ	IL-2Rα
Affinity constant (K_a):	$10^7 M$	$10^{11} M$	$10^8 M$
Dissociation constant (K_d):	$10^{-9} M$	$10^{-11} M$	$10^{-8} M$
Cells expressed by:	NK cells Resting T cells (low numbers)	Activated CD4+ and CD8+ T cells Activated B cells (low numbers)	

A number of cytokine receptors signal via the JAK/STAT pathway.

These include the receptors for IL-2, IL-3, IL-4, IL-6, IL-10, IL-12 and IFN-γ.

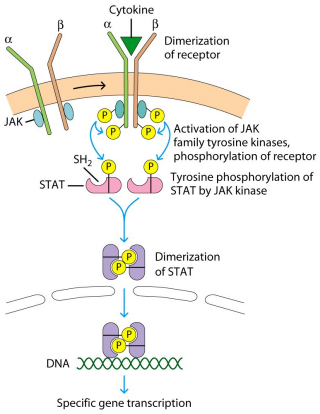
Cytokine receptor subunits are associated with JAK kinases.

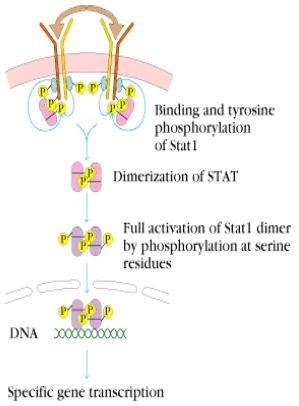
1.- Binding of cytokine causes dimerization of receptors and activation of JAK kinases.

2.- Activated JAK kinases phosphorylate receptor sites and create docking sites for STAT molecules.

JAK = Janus Kinase - OR - Just Another Kinase

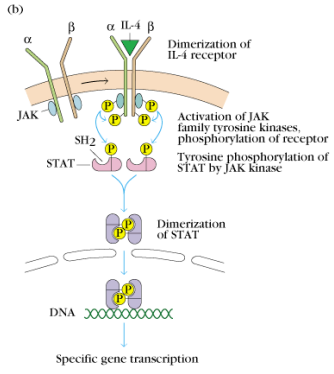
STAT = Signal Transducers and Activators of Transcription





3. After binding to the receptor (α chain), STATs are phosphorylated.

4. They then dissociate from the receptor, dimerize and translocate to the nucleus, where they mediate transcription of target genes.



Kuby Fig 12-10b

Similar JAK/STAT signaling in the IL-4 receptor.

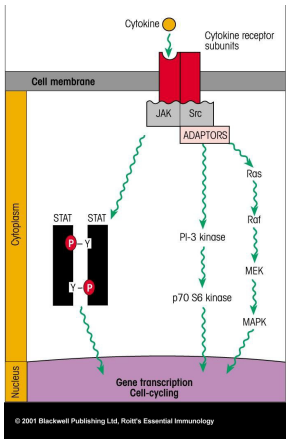
Different receptors associate with different JAK/STAT combinations

TABLE 12-2 STAT AND JAK INTERACTION WITH SELECTED CYTOKINE RECEPTORS DURING SIGNAL TRANSDUCTION

Cytokine receptor	JAK	STAT
IFN-γ	JAK1 and JAK2	Stat1*
IFN-α/β	JAK1 and Tyk-2	Stat2
IL-2	JAK1 and JAK3	Stat5
IL-3	JAK2	Stat5
IL-4	JAK1 and JAK3	Stat6*
IL-6	JAK1 (and sometimes others)	Stat3
IL-10	JAK1 and Tyk-2*	Stat3
IL-12	JAK2 and Tyk-2*	Stat4*

*Despite its name, Tyk-2 is also a Janus kinase.

SOURCE: Adapted from Bach, Aguet, and Schreiber, 1997, *Annu. Rev. Immun.* **15:**563.



Other pathways may also be involved in cytokine signaling.

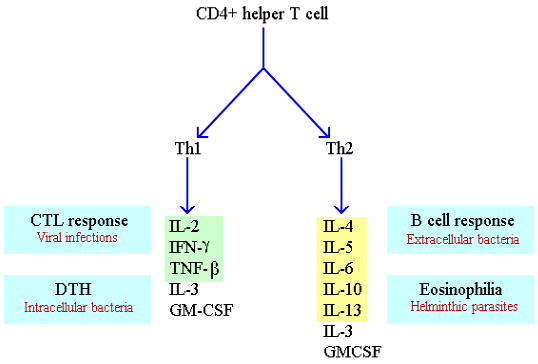


TABLE 12-4 CYTOKINE SECRETION AND PRINCIPAL FUNCTIONS OF MOUSE T_H1 AND T_H2 SUBSETS

Cytokine/function	T_H1	T_H2
Cytokine secretion		
IL-2	+	–
IFN-γ	++	–
TNF-β	++	–
GM-CSF	++	+
IL-3	++	++
IL-4	–	++
IL-5	–	++
IL-10	–	++
IL-13	–	++
Functions		
Help for total antibody production	+	++
Help for IgE production	–	++
Help for IgG2a production	++	+
Eosinophil and mast-cell production	–	++
Macrophage activation	++	–
Delayed-type hypersensitivity	++	–
T_C-cell activation	++	–

SOURCE: Adapted from F Powrie and RL Coffman, 1993, *Immunol. Today* 14:270.

Helper T cells can be divided into two main types - T_H1 and T_H2 - with distinct patterns of cytokine secretion.

T_H1 cells produce cytokines (IFN-γ and IL-2) that promote immune responses against intracellular pathogens (DTH, cytotoxic T cell responses, opsonizing Abs).

T_H2 cells produce cytokines (IL-4, IL-5, IL-6, IL-13) that promote immune responses against extracellular pathogens (antibody responses, eosinophilic responses, allergic reactions).

Some cytokines are produced by both T_H1 and T_H2 cells. These cytokines - GM-CSF and IL-3 - act on the bone marrow to increase production of leukocytes - so they are needed no matter what type of pathogen is present.

T_H1/T_H2 differentiation is influenced by the levels of key cytokines.

- IL-4 promotes T_H2 differentiation.

- IFN-γ and IL-12 promotes T_H1 differentiation.

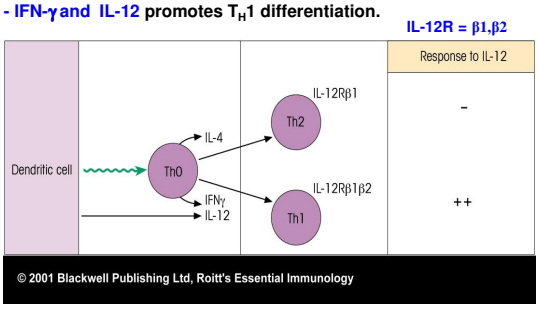


Figure 10.7

Cytokine cross-regulation

- IFN-γ (Th-1) inhibits proliferation of Th-2
- IL-4 and IL-10 (Th-2) inhibits proliferation of Th-1 by decreasing IL-12 production
- INF-γ (Th-1) promotes IgG2a production and decreases IgE by B cells
- IL-4 (Th-2) promotes production of IgE and IgG1 by B cells and decreases IgG2a.

Cytokine & Diseases

- **Bacterial Septic Shock**
  - Due to several Gram (-) bacteria
  - Stimulation of Macrophages by LPS → ↑ TNF-α, IL-1β
  - Drop in blood pressure, fever, diarrhea, systemic blood clotting in various organs
- **Bacterial Toxic Shock**
  - Caused by superantigens (wide variety of toxins)
  - Activation of T cells → ↑ cytokines from T cells and activated MØ
- Infectious Diseases
  - Leprosy, Chagas Disease.

Relative predominance of T_H1 vs T_H2 helper T cells can influence the course of infectious disease (*Mycobacterium leprae*)

T_H1 activity		T_H2 activity	
Tuberculoid	Lepromatous	Tuberculoid	Lepromatous
IL-2		IL-4	
IFN-γ		IL-5	
TNF-β		IL-10	

Tuberculoid - ↑ CMI (granulomas)
- No RIP

Lepromatous - ↑ HI (dissemination)
- RIP

Neuroendocrine regulation

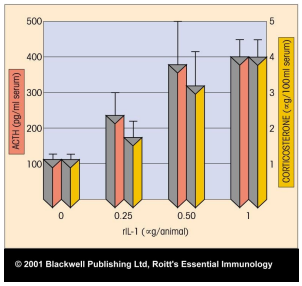


Figure 11.19

IL-1, IL-6 and TNF-α can induce production of glucocorticoids by acting on the hypothalamic-pituitary-adrenal (HPA) axis.

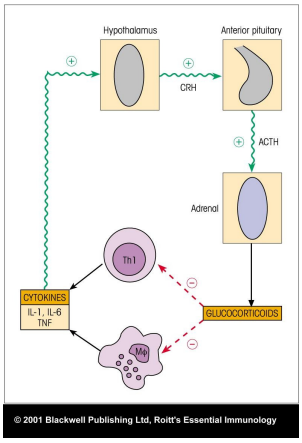


Figure 11.20

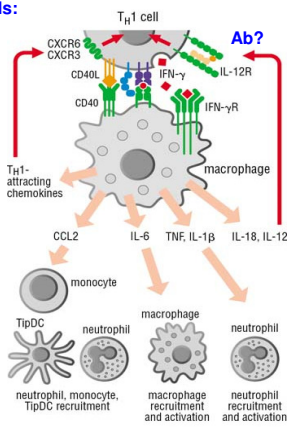
Glucocorticoid hormones can influence ongoing immune responses - particularly suppressing inflammatory responses.

Sex hormones also influence immune responses - e.g. females tend to be more prone to autoimmune disorders than males.

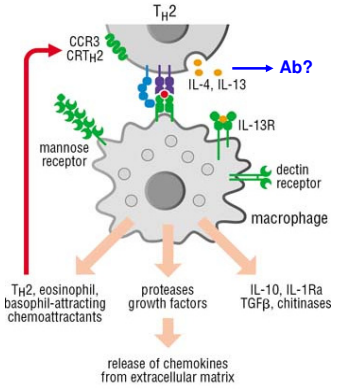
Stress may suppress Th1 immune responses.

The End, but interesting material next!!

Macrophage activation by Th1 Cells:

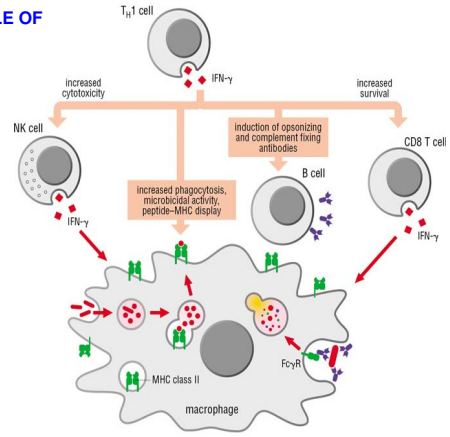


Macrophage activation by Th2 Cells:



EFFECTOR ROLE OF Th1 CELLS:

1) Cytotoxicity
2) ↑ Phagocytosis
3) ↑ opzonizing & complement fixing Abs



EFFECTOR ROLE OF Th2 CELLS:

1) IgE production
2) IgA production
3) Eosinophil recruitment
4) Basophil & Mast cell recruitment