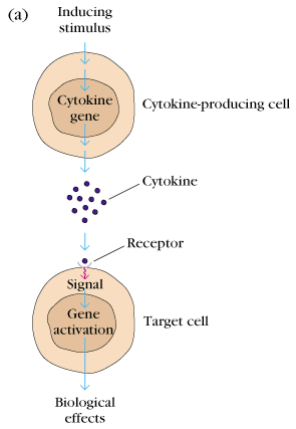
Chapter 13: Cytokines

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.

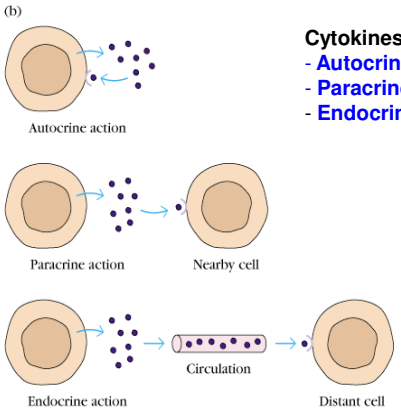
- Don't forget **chemokines**, they are also considered cytokines.



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

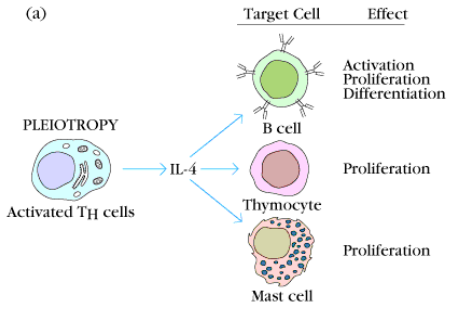
- **Expression of cytokines and their receptors is highly regulated.**

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

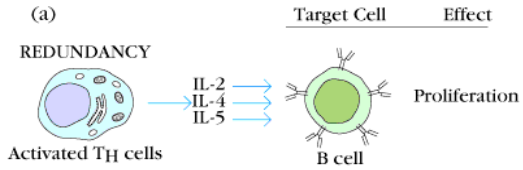


Cytokines can act in an:
- **Autocrine (same cell),**
- **Paracrine (close proximity)**
- **Endocrine (long distance)**

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

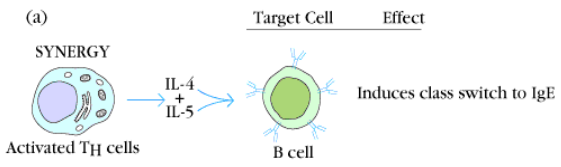


2. Cytokines can be redundant … different cytokines can have the same effects.

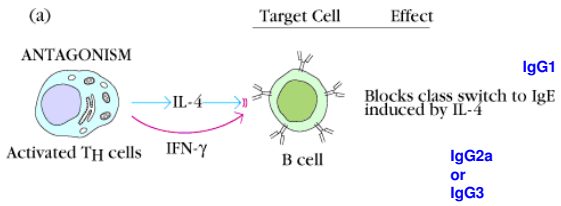


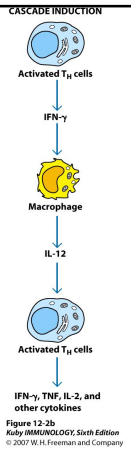
Kuby Fig 12-2a

3. Cytokines can synergize with each other.



4. Cytokines can antagonize each other.





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

6. Cytokines can influence the expression of cytokine receptors.

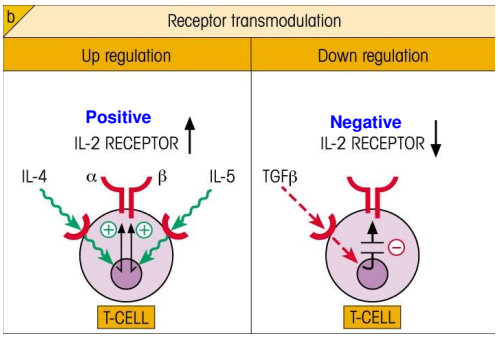


Figure 10.5b

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

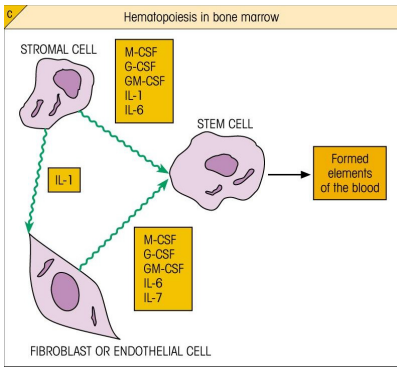
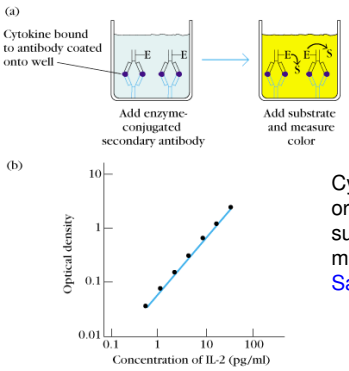


Figure 10.4c

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.

Exam Question!!!!

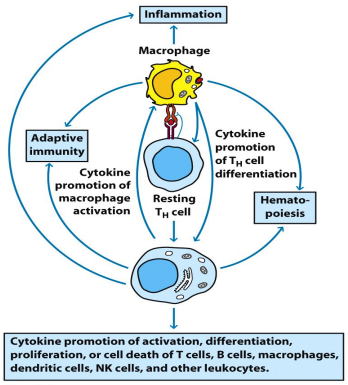


Figure 12-5
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There are many cytokines, including...

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	

TABLE 12-1 Functional groups of selected cytokines*

Cytokine†	Secreted by‡	Targets and effects
	SOME CYTOKINES OF INNATE IMMUNITY	
Interleukin 1 (IL-1)	Monocytes, macrophages, endothelial cells, epithelial cells	Vasculature (inflammation); hypothalamus (fever); liver (induction of acute phase proteins)
Tumor necrosis factor-α (TNF-α)	Macrophages	Vasculature (inflammation); liver (induction of acute phase proteins); loss of muscle, body fat (cachexia); induction of death in many cell types; neutrophil activation
Interleukin 12 (IL-12)	Macrophages, dendritic cells	NK cells; influences adaptive immunity (promotes T_H1 subset)
Interleukin 6 (IL-6)	Macrophages, endothelial cells	Liver (induces acute phase proteins); influences adaptive immunity (proliferation and antibody secretion of B cell lineage)
Interferon α (IFN-α) (this is a family of molecules)	Macrophages	Induces an antiviral state in most nucleated cells; increases MHC class I expression; activates NK cells
Interferon β (IFN-β)	Fibroblasts	Induces an antiviral state in most nucleated cells; increases MHC class I expression; activates NK cells
	SOME CYTOKINES OF ADAPTIVE IMMUNITY	
Interleukin 2 (IL-2)	T cells	T-cell proliferation; can promote AICD, NK cell activation and proliferation; B-cell proliferation
Interleukin 4 (IL-4)	T_H2 cells, mast cells	Promotes T_H2 differentiation; isotype switch to IgE
Interleukin 5 (IL-5)	T_H2 cells	Eosinophil activation and generation
Transforming growth factor β (TGF-β)	T cells, macrophages, other cell types	Inhibits T-cell proliferation and effector functions; inhibits B-cell proliferation; promotes isotype switch to IgA; inhibits macrophages
Interferon γ (IFN-γ)	T_H1 cells, $CD8^+$ cells, NK cells	Activates macrophages; increases expression MHC class I and class II molecules; increases antigen presentation

*Many cytokines play roles in more than one functional category.
†Only the major cell types providing cytokines for the indicated activity are listed; other cell types may also have the capacity to synthesize the given cytokine.
‡Also note that activated cells generally secrete greater amounts of cytokine than unactivated cells.

Table 12-1
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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

Four Structural Families

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

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

- Ig superfamily receptors

- Class I receptors (Hematopoietin receptor family)

- Class II receptors (Interferon receptor family)

- TNF receptor family

- Chemokine receptors

- TGF receptor family

RECEPTOR FAMILY **LIGANDS**

Immunoglobulin superfamily receptors

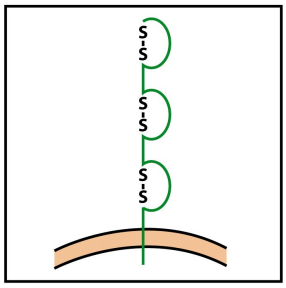

Figure 12-6a
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IL-1
M-CSF
C-Kit
IL-18

Kuby Fig 12-6a

RECEPTOR FAMILY **LIGANDS**

Class I cytokine receptors (hematopoietin)

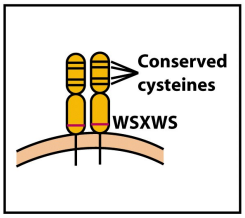

Figure 12-6b
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IL-2
IL-3
IL-4
IL-5
IL-6
IL-7
IL-9
IL-11
IL-12
IL-13
IL-15
IL-21
IL-23
IL-27
GM-CSF
G-CSF
OSM
LIF
CNTF
Growth hormone
Prolactin

RECEPTOR FAMILY **LIGANDS**

Class II cytokine receptors (interferon)

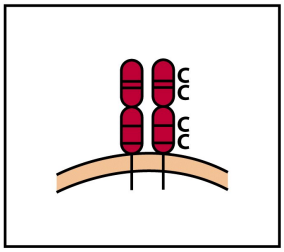

Figure 12-6c
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IFN-α
IFN-β
IFN-γ
IL-10
IL-19
IL-20
IL-22
IL-24
IL-26
IL-28
IL-29

RECEPTOR FAMILY **LIGANDS**

TNF receptors

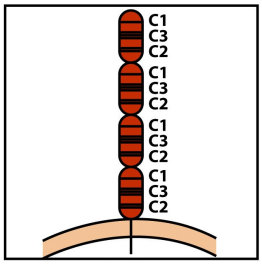

Figure 12-6d
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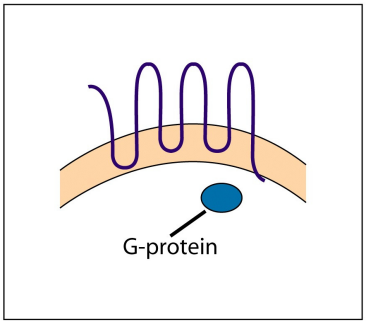
TNF-α
TNF-β
CD27L
CD30L
CD40L
Nerve growth factor (NGF)
FAS

(e) Chemokine receptors



LIGANDS

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

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

GM-CSF receptor subfamily (common β subunit)

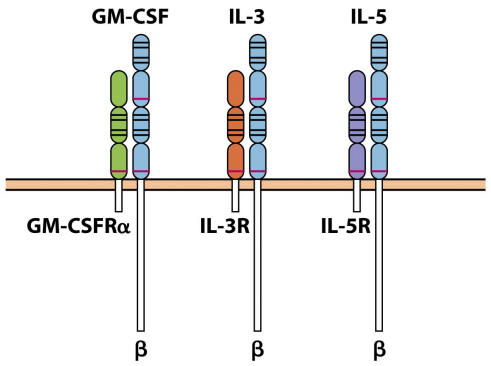


Figure 12-7a
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Hematopoietin Family

IL-6 Receptor subfamily (common gp130 subunit)

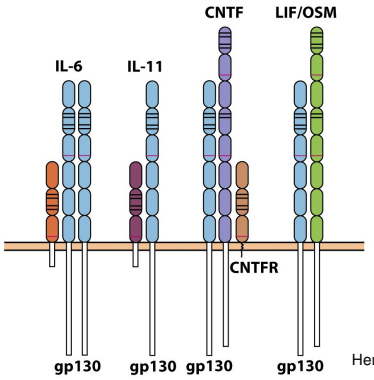

Figure 12-7b
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Hematopoietin Family

IL-2 receptor subfamily (common γ subunit)

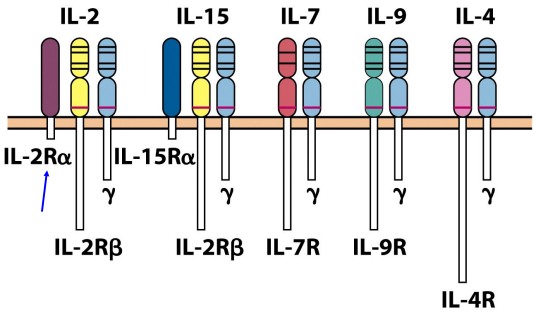

Figure 12-7c
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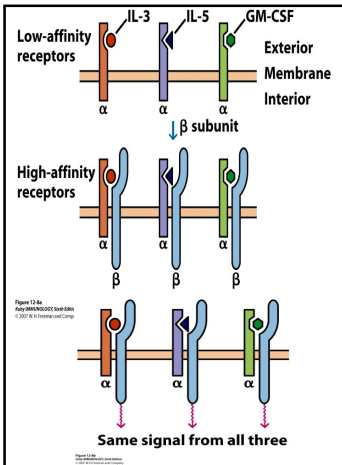
Hematopoietin Family

SHARED CYTOKINE RECEPTORS SUBUNITS

	Shared Cytokine-Receptor Subunits	
	Shared receptor chain	**Cytokines recognized**
C	γ_c	IL-2, -4, -7, -9, -15, -21
A	IL-2Rβ	IL-2, IL-15
	IL-4Rα	IL-4, -13
	IL-13Rα1	IL-4, -13
	β_c	IL-3, -5, GM-CSF
B	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

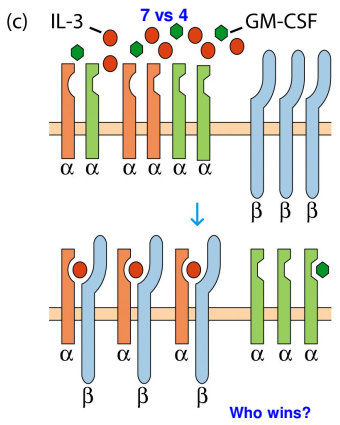
- Sharing of signal transducing molecules explains the **redundancy** and **antagonism** exhibited by some cytokines!



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.

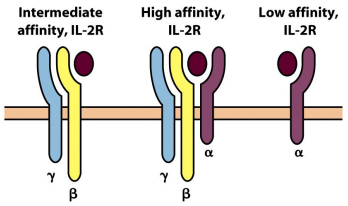
This can results in antagonistic effects between cytokines



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.

* Boy in the Bubble → the absence of a functional IL-2R -chain protein.



Only in activated T cells

	Intermediate affinity, IL-2R	High affinity, IL-2R	Low affinity, IL-2R
Subunit composition:	IL-2Rβ IL-2Rγ	IL-2Rα IL-2Rβ IL-2Rγ	IL-2Rα
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)	

Figure 12-9a
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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

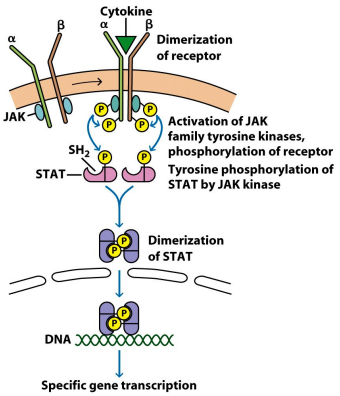

Figure 12-10
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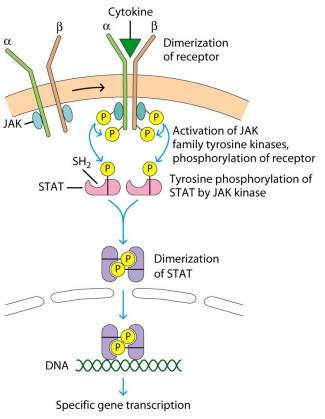


3. After binding to the receptor (α chain), STATs are phosphorylated by JAK kinases.

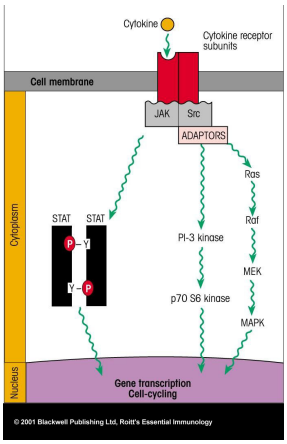
4. They then dissociate from the receptor, dimerize and translocate to the nucleus, where they mediate transcription of target genes.

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.

Cytokine Antagonists

Action:

1) Blocking the receptor (IL-1Ra), and
2) Binding to the cytokine (IL-2, IFN-γ)

TABLE 12-3 Viral mimics of cytokines and cytokine receptors

Virus	Products
Leporipoxvirus (a myxoma virus)	Soluble IFN-γ receptor
Several poxviruses	Soluble IFN-γ receptor
Vaccinia, smallpox virus	Soluble IL-1β receptor
Epstein-Barr	IL-10 homolog
Human herpesvirus-8	IL-6 homolog; also homologs of the chemokines MIP-I and MIP-II
Cytomegalovirus	Three different chemokine receptor homologs, one of which binds three different soluble chemokines (RANTES, MCP-1, and MIP-1α)

Table 12-3
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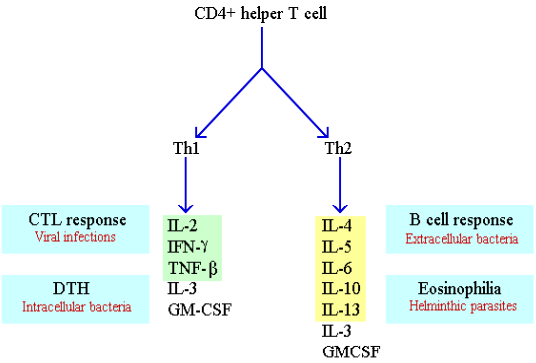

TABLE 12-4 Cytokine secretion and principal functions of mouse T_H1 and T_H2 subsets

	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 R. L. Coffman, 1993, *Immunology Today* 14:270.

Table 12-4
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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, macrophage activation, opsonizing Abs).

T_H2 cells produce cytokines (IL-4, IL-5, IL-6, IL-13) that promote immune responses against extracellular pathogens (antibody responses IgE/IgG1, 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.

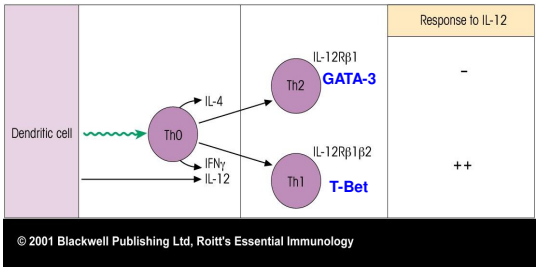
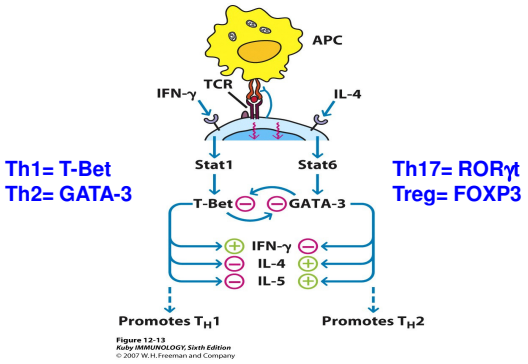


Figure 10.7

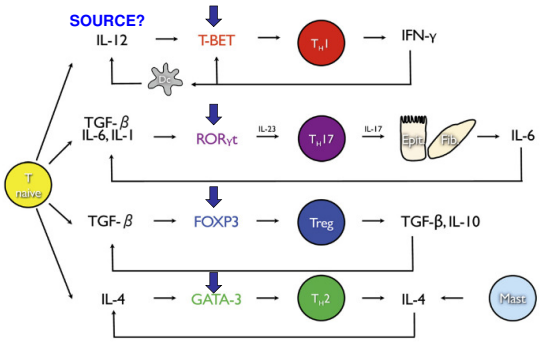
Cytokine cross-regulation

- 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 IgG1 and IgE by B cells
- IL-4 (Th-2) promotes production of IgE and IgG1 by B cells and decreases IgG2a.

Transcriptional Regulation of Cytokines



Transcription factors can be used to characterize Th lineage



Cytokine & Diseases

- **Bacterial Septic Shock**
  - Due to several Gram (-) bacteria
  - Stimulation of Macrophages & DCs by LPS → ↑ TNF-α, IL-1β
  - Drop in blood pressure, fever, diarrhea, systemic blood clotting in various organs, increased respiratory rate, capillary leakage, etc
- **Bacterial Toxic Shock**
  - Caused by superantigens (wide variety of toxins)
  - Activation of T cells → ↑ cytokines from T cells and activated MØ (↑ TNF-α, IL-1β)
- Infectious Diseases
  - Leprosy, Chagas Disease (↓IL-2Rα).

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

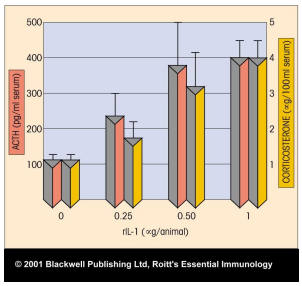


Figure 11.19

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

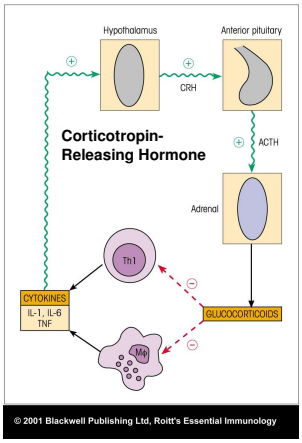


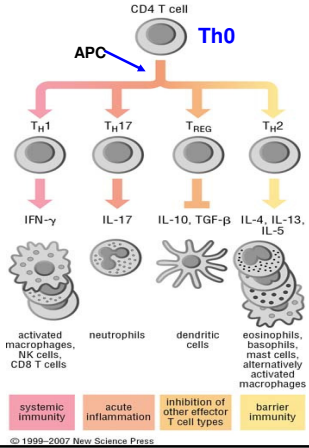
Figure 11.20

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.

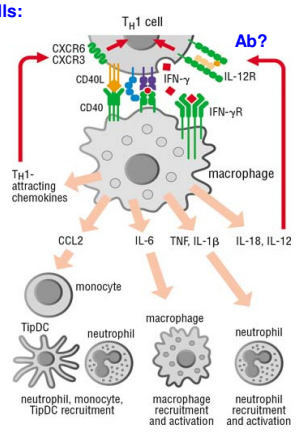
Stress may suppress Th1 immune responses.

The End, but interesting material next!!

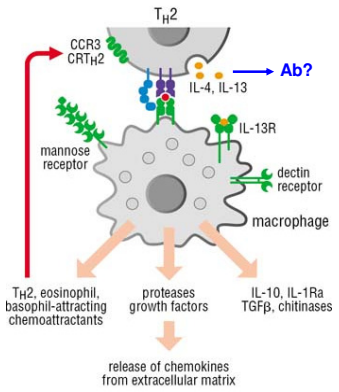


- Outcome of CD4 T cells
- Th_0 will give rise to at least 3 Th populations
- Th populations differentiated based on cytokine production
- Treg

Macrophage activation by Th1 Cells:

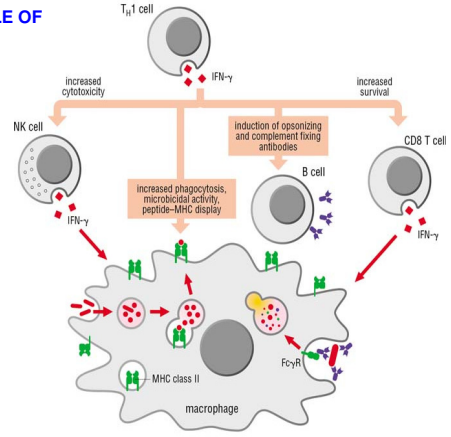


Macrophage activation by Th2 Cells:



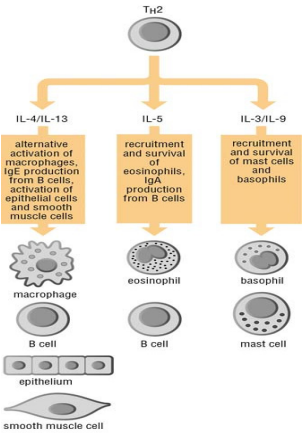
EFFECTOR ROLE OF Th1 CELLS:

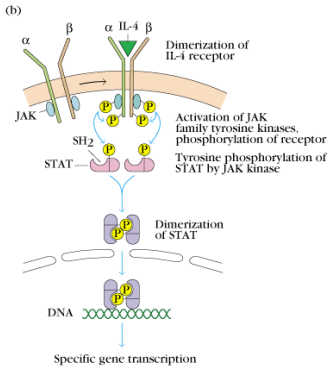
1) Cytotoxicity
2) ↑ Phagocytosis
3) ↑ opzonizing & complement fixing Abs



EFFECTOR ROLE OF Th2 CELLS:

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





Kuby Fig 12-10b

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