Lecture 11

T-cell Effector Mechanisms-II: Cytokine Secretion & T-cell Polarization

September 21, 2005

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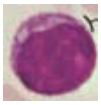
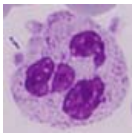
Blood: 4-10,000 WBC per 1 μL

Lymphocytes - 10-15 %

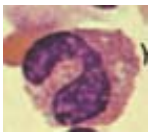


(T-, B- & NK cells)

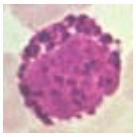
Granulocytes - 35-80 %



PMNs
35-80 %

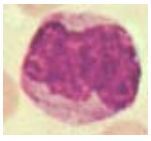


Eos
0-8 %



Basos
0-2 %

Monocytes - 0-15 %



(Macs & DCs)

How did they get there?
Where are they going?
What regulates them?

Think Cytokines, Chemo-kines & Growth Factors !!

What are cytokines and chemokines?

- Small (10-30 kDa), usually secreted and usually glycosylated peptides.
- They bind specific, high affinity (e.g., K_d of 10^{-10}-10^{-12} M) receptors found on target cells.
- Expression of cytokines and their receptors is usually tightly regulated (i.e., temporally/ transiently and geographically).
- Cytokine receptors define the specific type of biological response cytokines stimulate.
- Other more anachronistic terms include monokines and lymphokines. The term interleukin (IL) is now commonly used (e.g., IL-1, IL-2, …).

What do cytokines, chemokines and growth factors do?

- They direct the development, maturation, localization, interactions, activation and life span of immune cells.
- Thus they play an essential role in regulating both immunity adaptive and innate.

How many flavors regulate immunity?

- Growth Factors (e.g., CSF-1, SCF, RANKL,Flt3L)
- IL-1 Family (e.g., IL-1, IL-18 & "Toll-like")
- TNF Family (e.g., TNF-α, CD40L, FasL, LT-β, BAFF)
- TGF-β Family (e.g., TGF-β)
- Chemokines (e.g., CC and CXC families)
- Type I & II Cytokines (a.k.a.Hematopoietins or Four Helix Bundle (e.g., IL-2, IL-4, IL-6, IL-10, IL-12, IL-13, IL-15, GM-CSF, IFN-γ, IFN-α/β)

- Also steroid hormones and prostaglandins

Cytokines & Chemokines can be grouped into functionally related Families

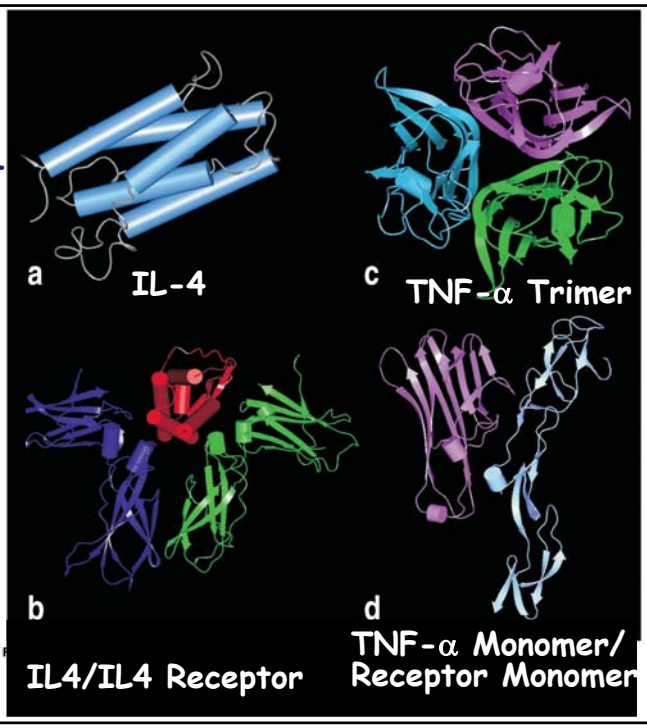
- There are significant functional <u>similarities within</u> each receptor family. The same is true for corresponding ligands (see summary).
- There are important functional <u>differences between</u> between receptor families (see summary).

Cytokine Receptor Classes

Table 11–2. Signal Transduction Mechanisms of Cytokine Receptors

Signal transduction pathway	Cytokine receptors using this pathway	Signaling mechanism
JAK/STAT pathway	Type I and type II cytokine receptors	JAK-mediated phosphorylation and activation of STAT transcription factors (see Box 11–2)
TNF receptor signaling by TRAFs	TNF receptor family: TNR-RII, CD40	Binding of adapter proteins, activation of transcription factors (see Box 11–1)
TNF receptor signaling by death domains	TNF receptor family: TNF-RI, Fas	Binding of adapter proteins, caspase activation (see Box 11–1)
Receptor-associated tyrosine kinases	M-CSF receptor, stem cell factor receptor	Intrinsic tyrosine kinase activity in receptor
G protein signaling	Chemokine receptors	GTP exchange and dissociation of Gα · GTP from Gβγ, Gα · GTP activates various cellular enzymes

Consistent with their significant functional differences, IL-4 & TNF-α, and their corresponding receptors are structurally quite distinct.



Localized release of IL-4 in the cleft between T cell and APC ("The immunological synapse")

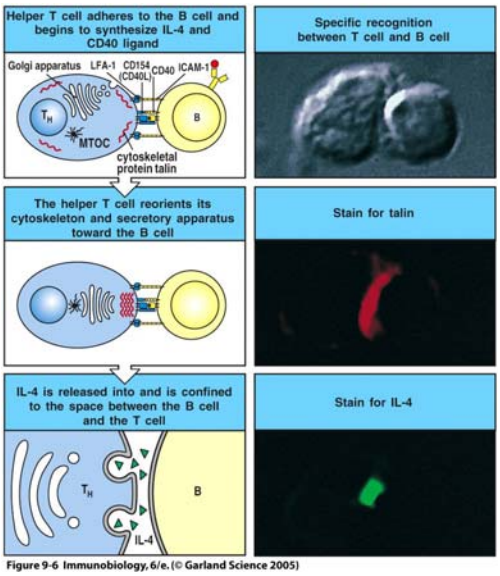


Figure 9-6 Immunobiology, 6/e. (© Garland Science 2005)

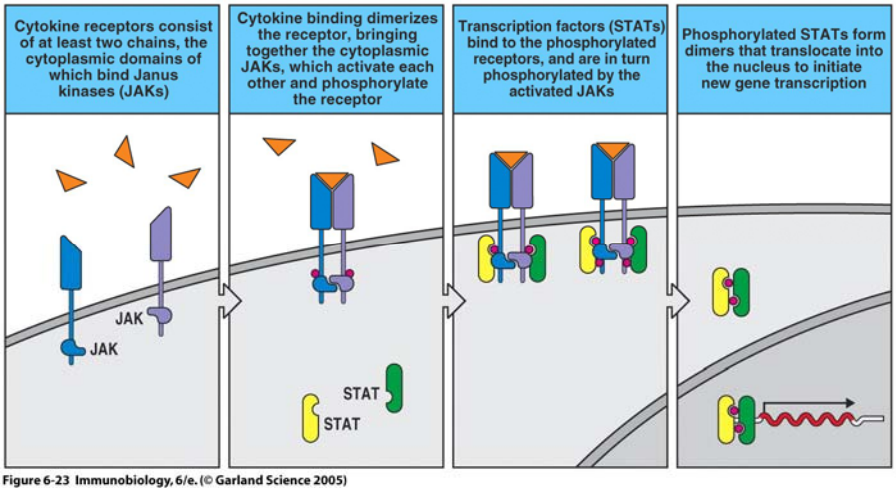
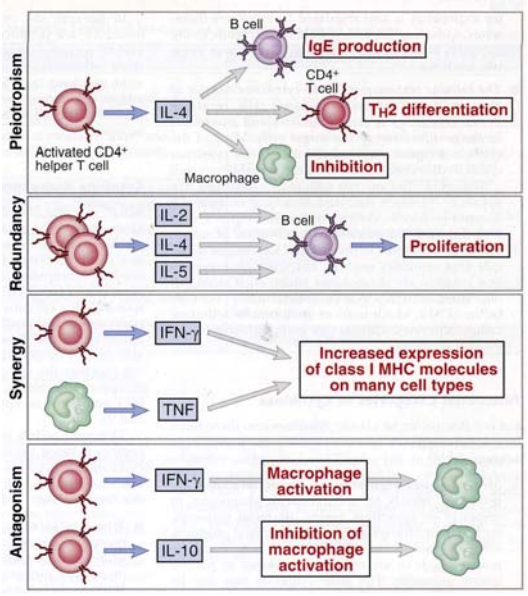


Figure 6-23 Immunobiology, 6/e. (© Garland Science 2005)

General functional properties of Cytokines and Chemokines

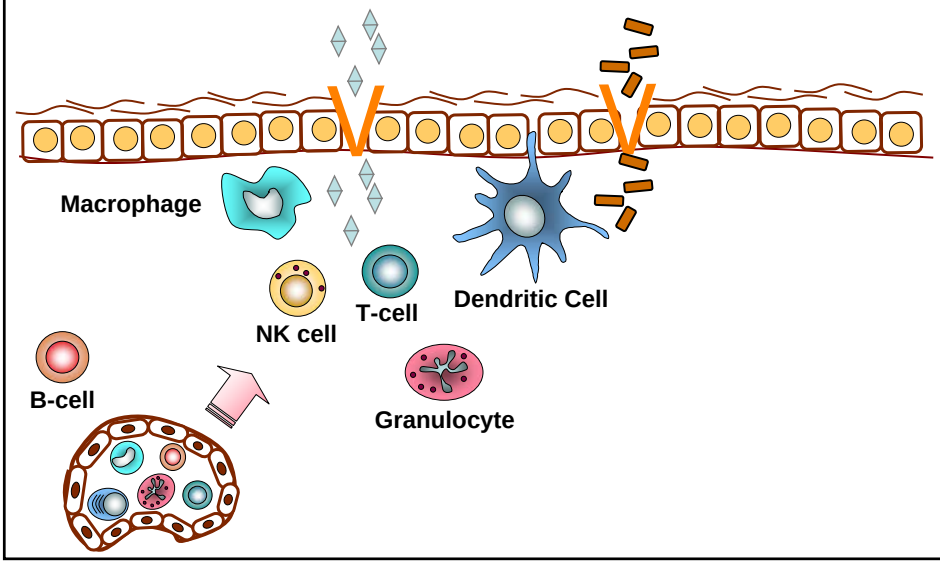
- Usually stimulate transient responses.
- Function at three ranges:
  - Autocrine - “self”
  - Paracrine - adjacent cells
  - Endocrine - through circulatory system
- **Pleitropism** - one ligand activate numerous types of responses (e.g., differentiation, growth & activation).
- **Redundancy** - two or more ligands exhibit functional overlap.
- **Synergy -** two or more ligands synergize to mount a single response.
- **Antagonsism -** two or more cytokines mediating opposite responses to either limit a response or achieve balance (e.g. Feedback loops).

Properties of Cytokines



Some Biology . . . . . .

How do we protect ourselves from microbes?
The antiviral response



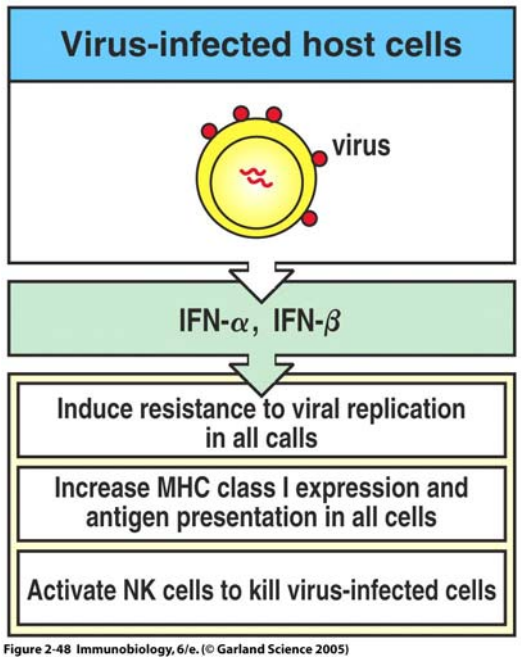


Figure 2-48 Immunobiology, 6/e. (© Garland Science 2005)

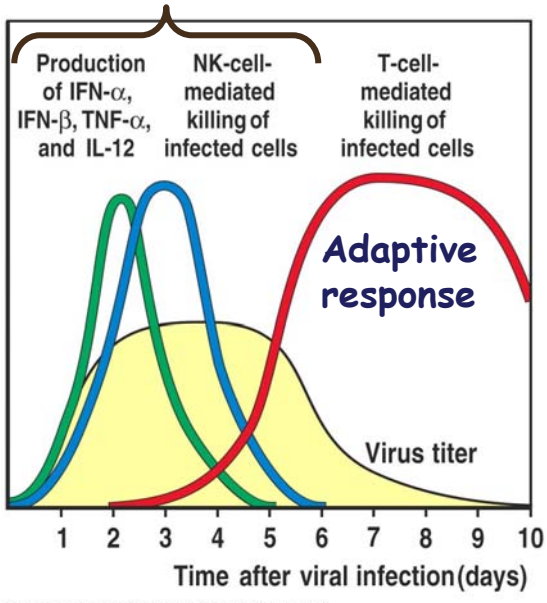
Cytokines initiate the innate immune response during a viral infection.

For adaptive CD8 immunity

An innate response

Innate response to Virus

Inflammatory Cytokines



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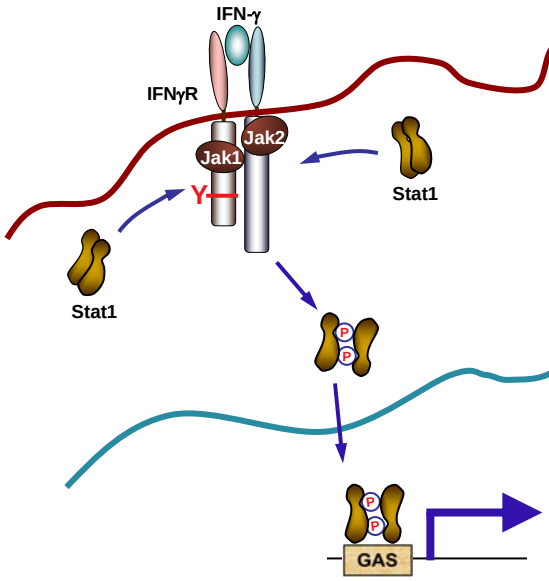
Type I & II Cytokines mediate their biological response through the induction of genes



The JAK-STAT Signaling Paradigm

Cytokines **(IFN-γ, IFN-β)**

Chemokines **(CXCL9, CXCL10)**

Transcription factors **(IRF1, CIITA)**

Enzymes **(iNOS)**

Ag presentation **(MHC-I, MHC-II, TAP)**

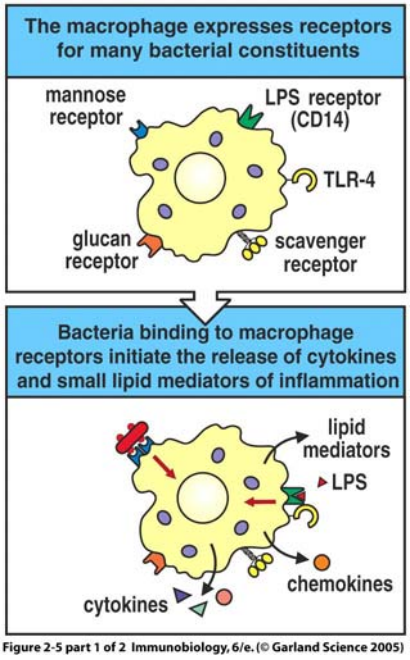
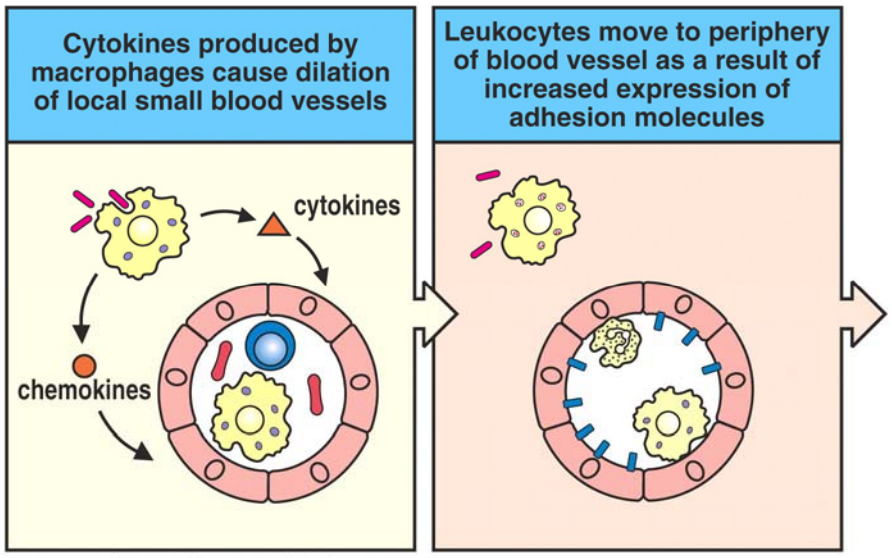


Figure 2-5 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

What about during a bacterial infection, how do macrophage and DC sentries sense and respond?

Local vs Systemic Response



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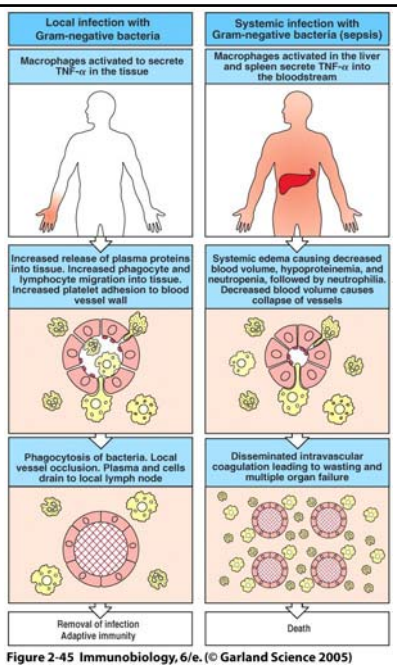
Local vs. systemic infection:

The response to LPS



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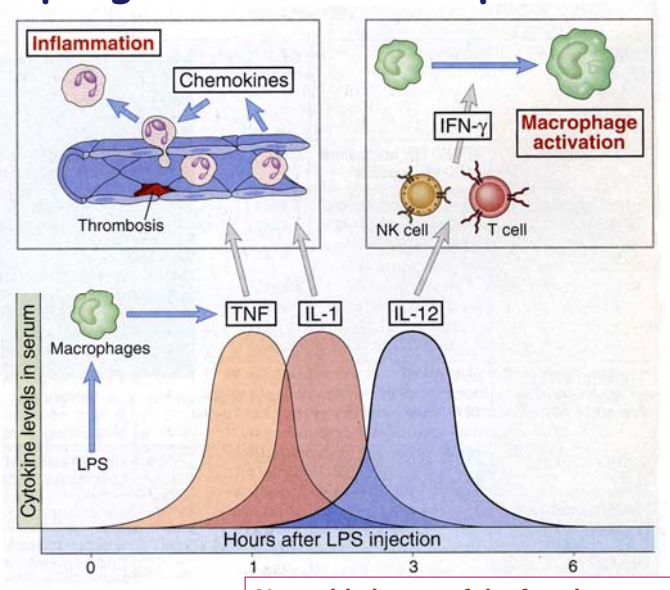
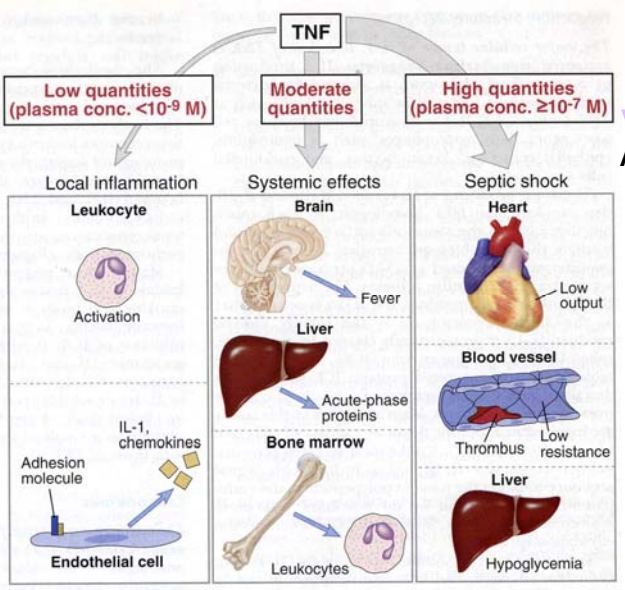
Infection vs. Sepsis

Macrophages critical in response to LPS



Note, this is one of the few times you can meaningfully measure serum cytokine levels!!

Biological actions of TNF



New Anti-inflammatory Agents

Remicade (infliximab)
Anti-TNF

Enbrel (etanercept)
TNF-Trap

Kineret (anakinra)
rIL1-RA

Cytokines and the Th1-Th2 paradigm

Dendritic Cells pick up antigen, get activated and migrate to lymph nodes

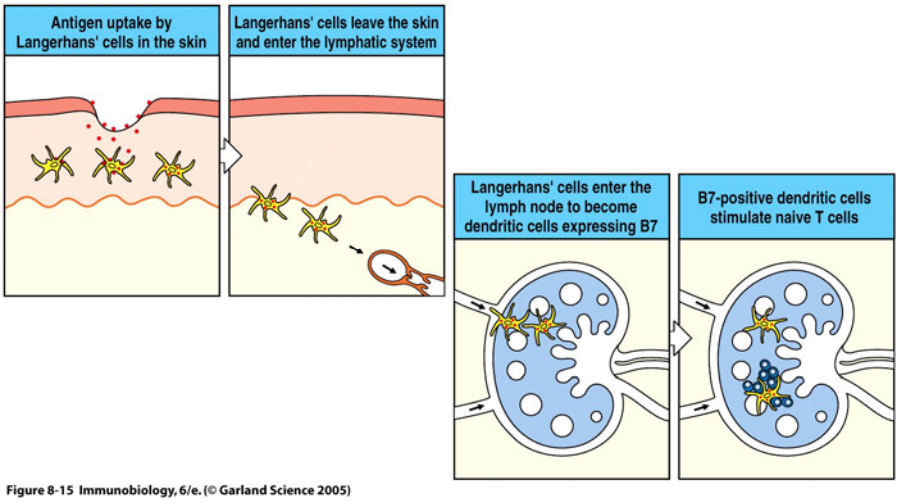


Figure 8-15 Immunobiology, 6/e. (© Garland Science 2005)

Naïve CD4 cells kiss APCs to sample them for right antigen ($1<10^4$-10^6)

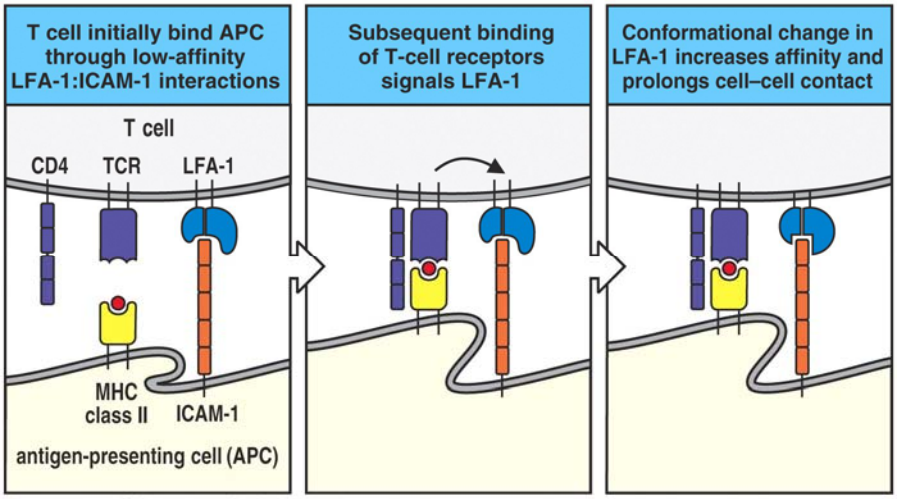


Figure 8-9 Immunobiology, 6/e. (© Garland Science 2005)

Signal #1 (TCR) and Signal #2 (coreceptors) direct activation of naïve T-cells.

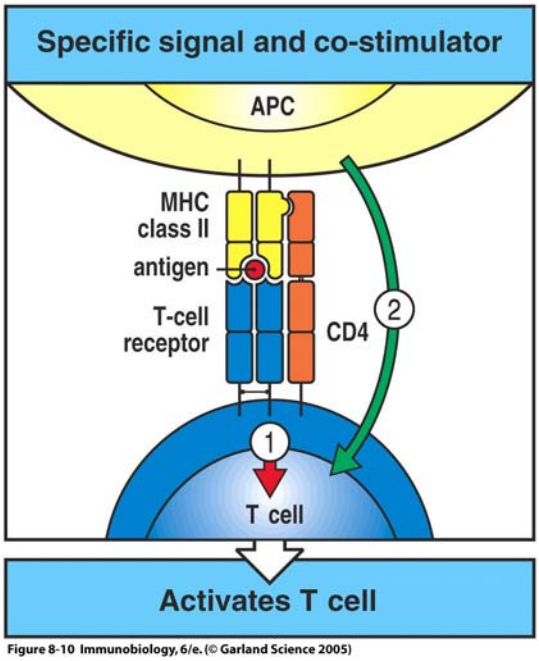
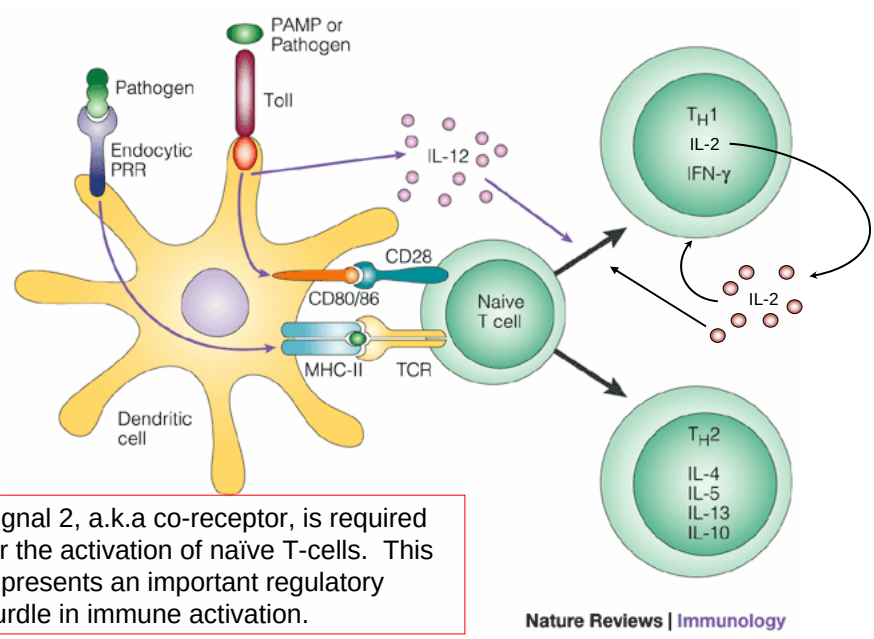


Figure 8-10 Immunobiology, 6/e. (© Garland Science 2005)

Cytokines direct Th1-Th2 polarization



Signal 2, a.k.a co-receptor, is required for the activation of naïve T-cells. This represents an important regulatory hurdle in immune activation.

The important IL-2 "autocrine loop"

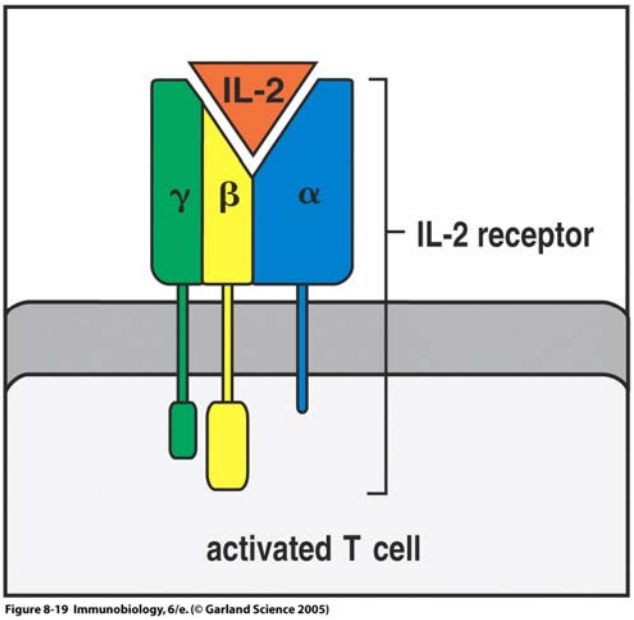


Figure 8-19 Immunobiology, 6/e. (© Garland Science 2005)

The IL-2 autocrine loop

TCR stimulation leads to induction of IL-2 and IL-2 receptor α-chain to generate high affinity receptor, culminating in potent T-cell proliferation.

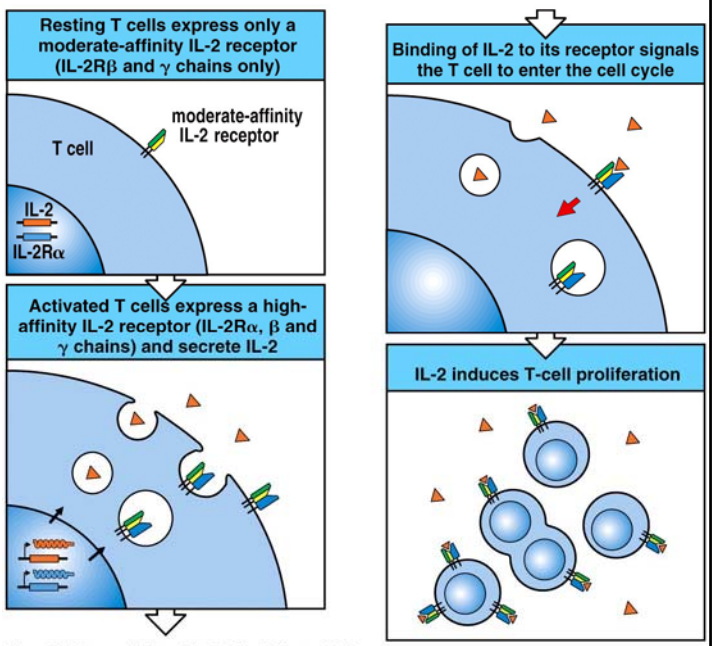


Figure 8-20 Immunobiology, 6/e. (© Garland Science 2005)

Now let's digress to review how TCR signaling directs cytokine production. This is an important drug target!

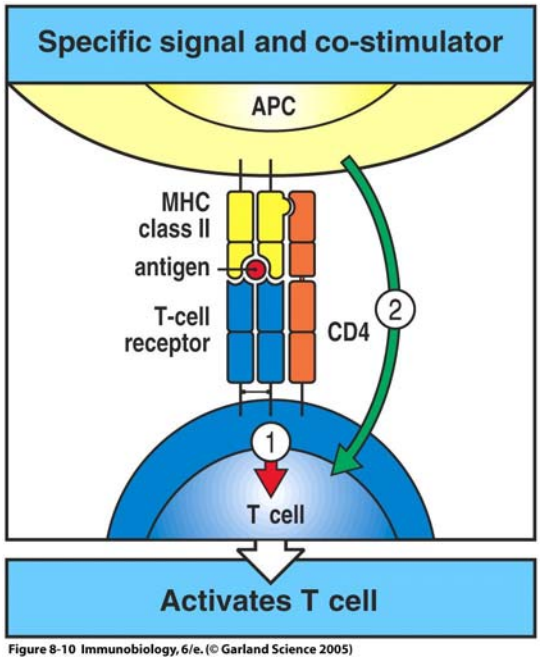
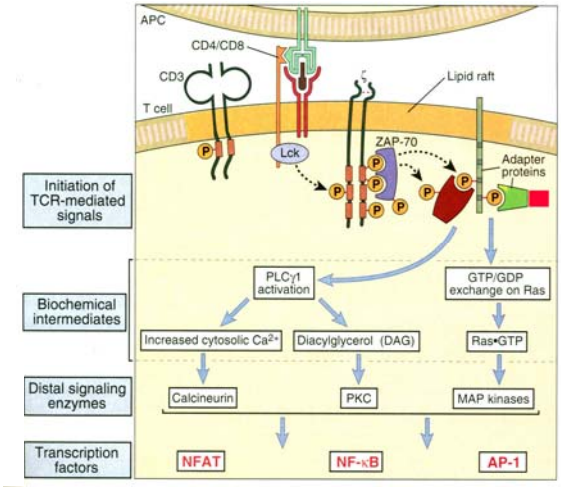


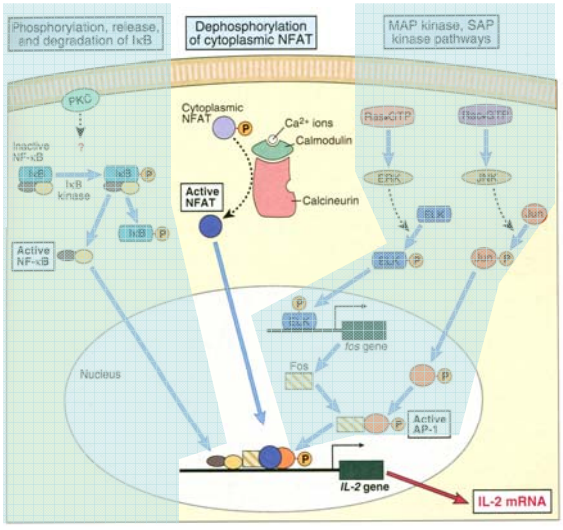
Figure 8-10 Immunobiology, 6/e. (© Garland Science 2005)

TCR-mediated Signal Transduction: A Tyrosine Kinase Cascade

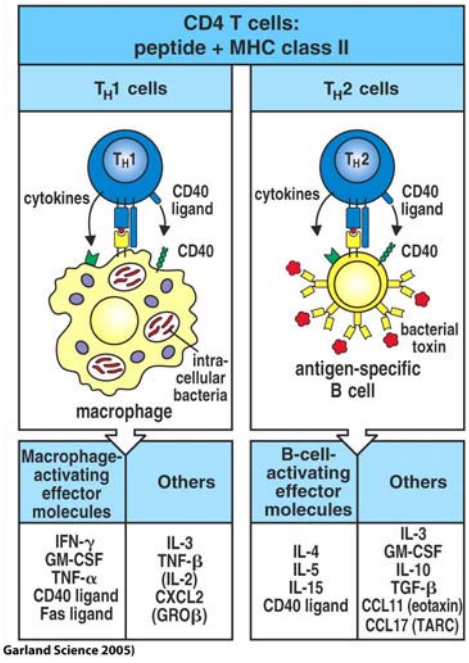


NF-AT & TCR-mediated signal transduction culminate in cytokine production

Cyclosporin A (CyA) & Tacrolimus (FK506) are two important drugs that block calcineurin activation ⇒ NFAT activation ⇒ **IL-2** production! They are therefore potent immunosuppressive drugs.



Th1 and Th2 cells each secrete signature cytokines & chemokines that define their effector functions.



Important Th1 effector cytokines

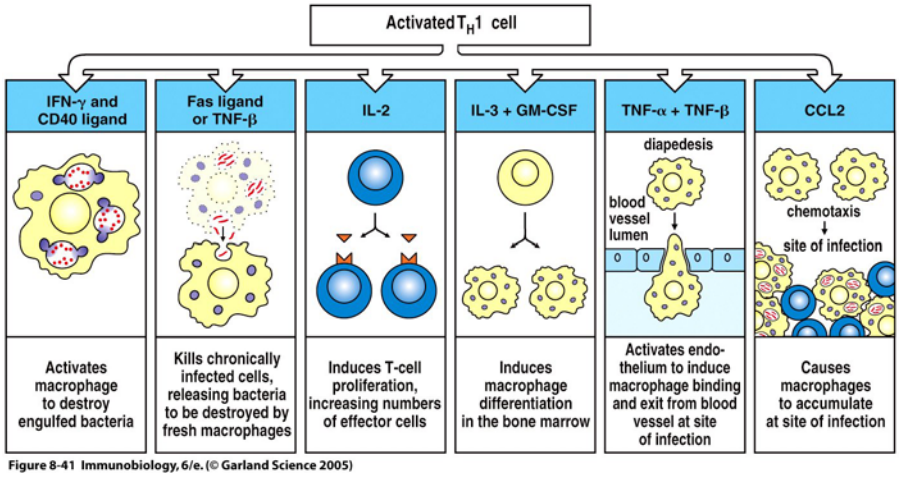


Figure 8-41 Immunobiology, 6/e. (© Garland Science 2005)

IFN-γ is the Th1 signature cytokine

Important Th2 effector cytokines IL-4, IL-5 & IL-6 promote humoral immunity

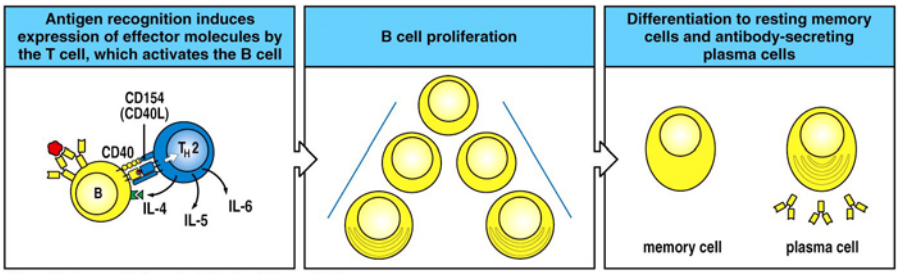
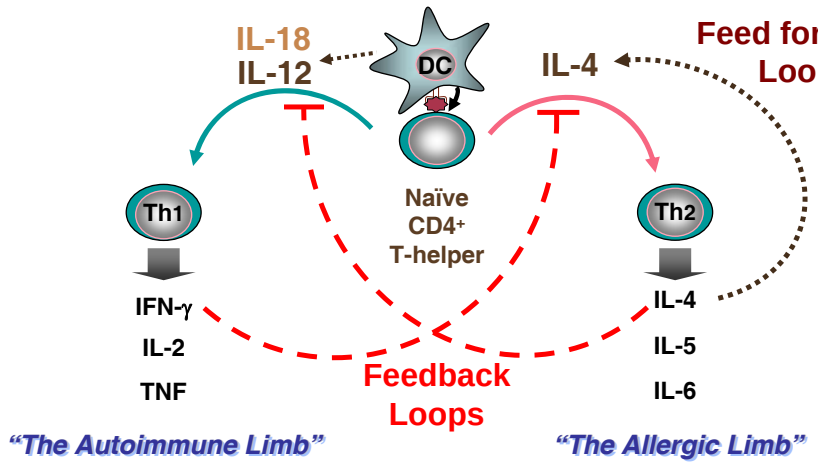


Figure 9-5 Immunobiology, 6/e. (© Garland Science 2005)

IL-4, the signature Th2 effector cytokine regulates B-cells, . . . and IL-10 & TGF-β potently antagonize cellular immunity (think regulatory T-cells).

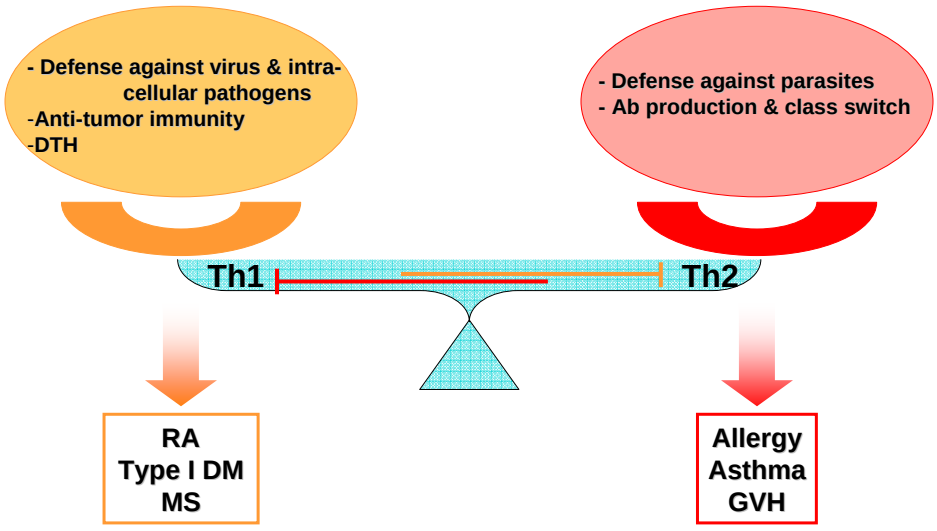
CCL11/17

Polarization of CD4+ T-cells into functional Th1 and Th2 subsets



Other factors that influence the decision to become Th1 vs. Th2 include co-stimulators and nature of peptide:MHC interaction

Failure to balance Th1 and Th2



RA: Rheumatoid arthritis
DM: Dibetes mellitus

MS: Multiple sclerosis
GVH: Graft-vs-Host disease

DTH: Delayed-type hypersensitivity

Chemokines

Chemokines signal through G-protein coupled receptors making them desirable drug targets

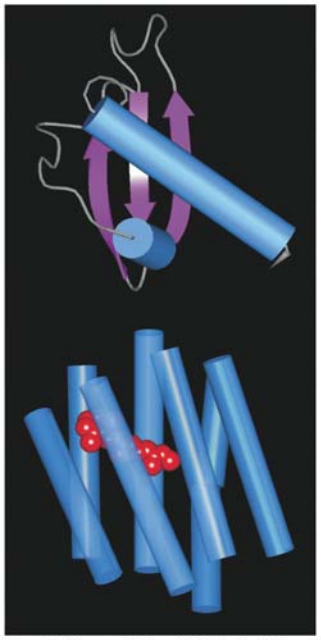
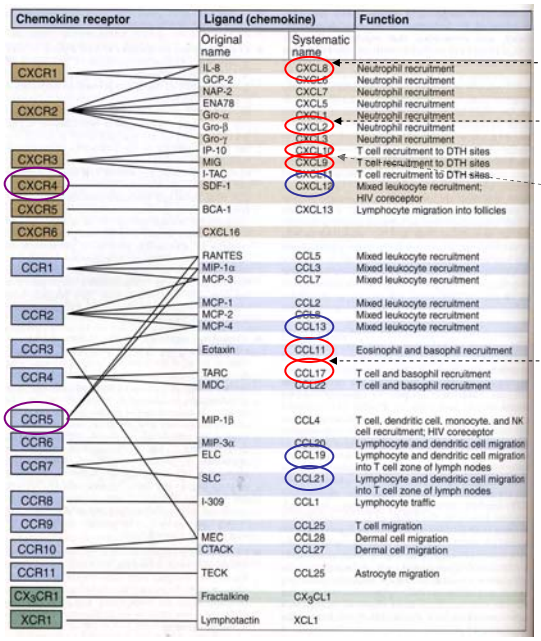


Figure 2-40 Immunobiology, 6/e. (© Garland Science 2005)

Chemokine Redundancy



Abbas Chpt. 11

***Inflammatory**

***Homeostatic**

Leukocytes express unique sets of chemokines receptor signatures allowing them to be targeted to the appropriate tissues either homeostatically or drive an inflammatory response.

QuickTime™ and a GIF decompressor are needed to see this picture.

Homeostatic targeting of lymphocytes and APCs in the spleen

QuickTime™ and a
GIF decompressor
are needed to see this picture.

Cell	Chemokine receptor	Chemokine sensed
DC	CCR7	ELC, SLC
naïve T	CCR7	ELC, SLC
naïve B	CXCL5	BLC

Chemokines are much more than just chemo-attractants

CXCL12

QuickTime™ and a
GIF decompressor
are needed to see this picture.

CXCR4

Of Note . . . . .

- **Two chemokine receptors serve as co-receptors for HIV infection (CXCR4 and CCR5)**

Cytokines you should know

Type I & II Cytokine Receptors (JAK-STAT)

- **IL-2** -Th1 cytokine ⇒T-cell proliferation
- **IL-4** -Th2 cytokine ⇒ B-cell proliferation; Th2 polarization
- **IL-6** -Th2 cytokine ⇒ B-cell proliferation; Plasma cell growth
- **IL-10** -Th2 cytokine ⇒ antagonizes cellular immunity
- **IL-12** -DC cytokine ⇒ drives Th1 polarization
- **IFN-γ** -Th1 cytokine ⇒drives inflammation; Mac. Activation; DTH
- **IFN-α** -All cells make this antiviral cytokine

Toll (TLR) /IL-1 Receptors (NFκB)

- **IL-1** -Potent activator of inflammation & innate immunity
- TLR -Potent activators of innate and adaptive immunity

TNF Related Receptors (NFκB vs. Caspases)

- **TNF** -Potent activator of inflammation & innate immunity (arthritis)
- **CD40L** - T-cell help (survival/proliferation) to B-cells
- **FasL** -Induces cell death: to achieve negative selection; to terminate an immune response

TGF-β Receptors

- **TGF-β** -Antagonizes cellular immunity and promotes wound healing

Chemokine Receptors (GPCRs*)

- Chemokines (*see Fig. 11.6*)
  - **Inflammatory** (e.g., CCL11, CCL17, CXCL2, CXCL8/9/10)
  - **Non-inflammatory** (i.e. homeostatic; e.g., CCL19, CCL21,CXCL-12, CXCL-13, S-1P)

*G-Protein Coupled Receptors -Good drug targets