M. Sc. Microbiology

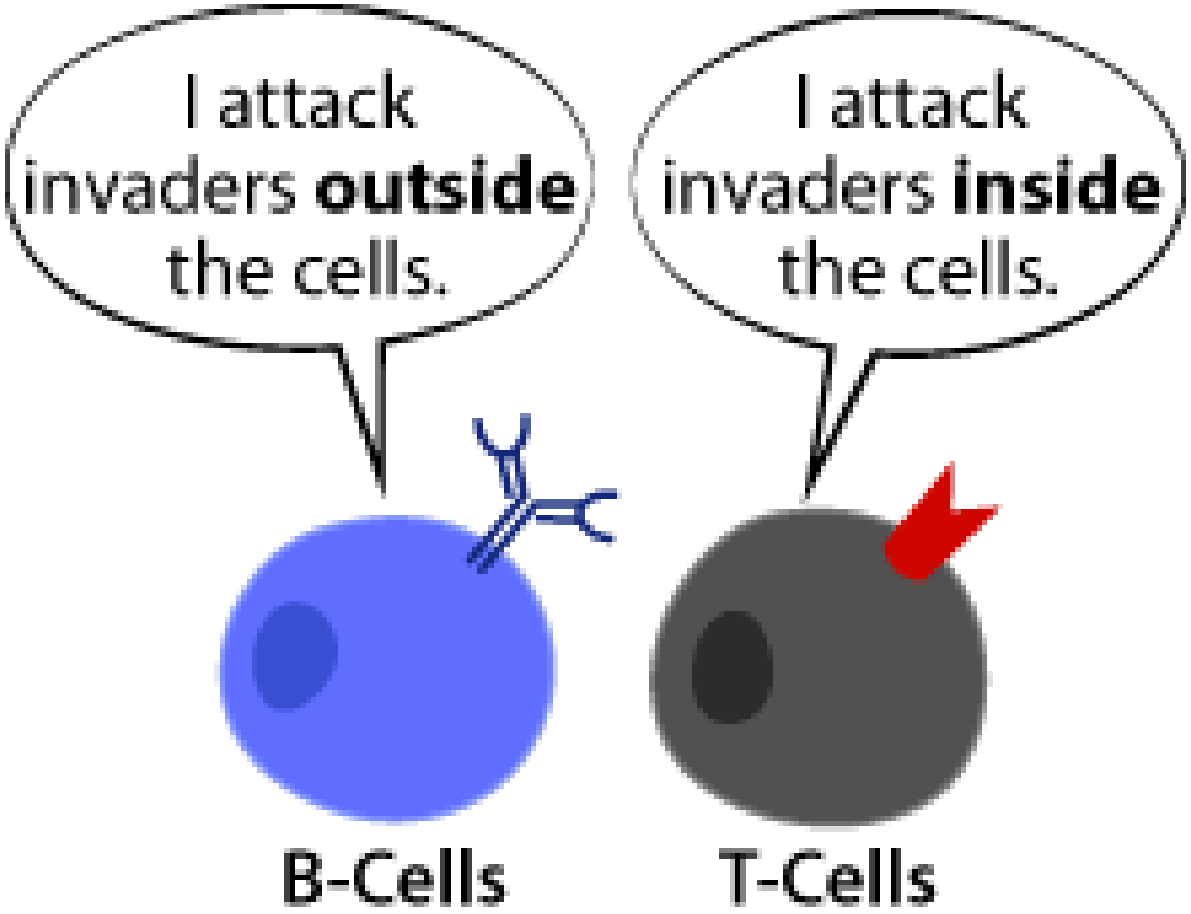
Semester – II

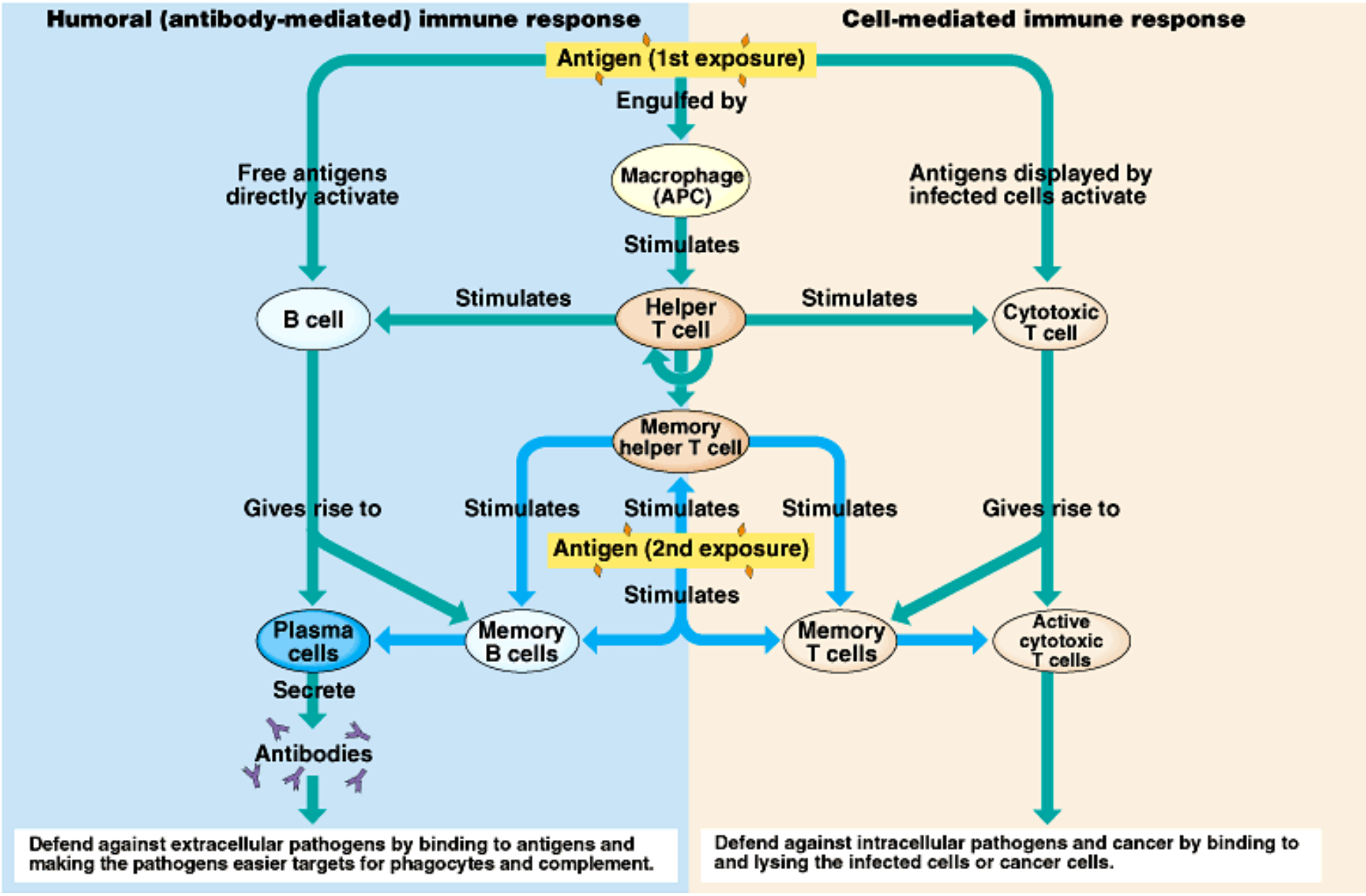
Paper – 201 B

Sub : Immunology

Topic : Immuno competent cells, B & T cell epitope, Antigen processing, Major Histocompatibility complex, Humoral Immunity, Cell mediated immunity

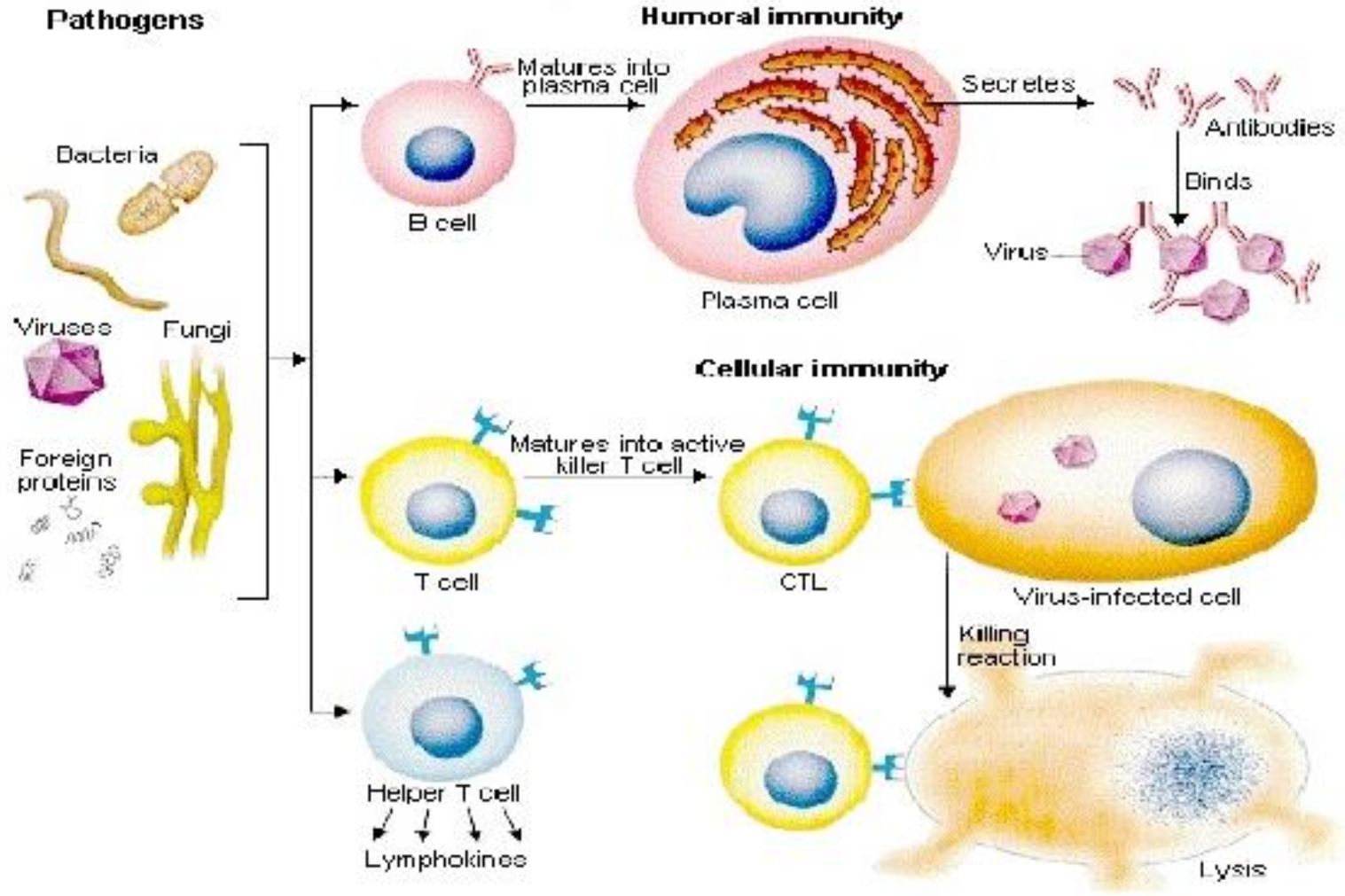
Prof. Keshab Chandra Mondal


I attack invaders **outside** the cells.
I attack invaders **inside** the cells.
B-Cells
T-Cells


Humoral (antibody-mediated) immune response
Cell-mediated immune response
Antigen (1st exposure)
Engulfed by
Free antigens directly activate
Macrophage (APC)
Antigens displayed by infected cells activate
Stimulates
B cell
Stimulates
Helper T cell
Stimulates
Cytotoxic T cell
Memory helper T cell
Gives rise to
Stimulates
Stimulates
Stimulates
Gives rise to
Antigen (2nd exposure)
Stimulates
Plasma cells
Memory B cells
Memory T cells
Active cytotoxic T cells
Secrete
Antibodies
Defend against extracellular pathogens by binding to antigens and making the pathogens easier targets for phagocytes and complement.
Defend against intracellular pathogens and cancer by binding to and lysing the infected cells or cancer cells.

Humoral and cellular immunity

(antibody mediated or cellular)



Adaptive Immunity

Adaptive immune system has two arms

Adaptive Immunity

Humoral Immunity

- Provided by B lymphocytes
- Can recognize protein, polysaccharide, phospholipid and nucleic acid antigens
- Can act against soluble or free antigens
- Provides immunity to extracellular bacteria, viruses and toxins
- Causes Type I, II & III hypersensitivity

Cell mediated Immunity

- Provided by T lymphocytes
- Can recognize only protein antigens
- Recognizes antigens presented by APCs with Class I or Class II MHC molecule
- Provides immunity to intracellular bacteria, viruses, fungi and protozoa
- Causes Type IV hypersensitivity
- Causes acute graft rejection

Cell Surface Markers

- **Leucocytes can be differentiated by the expression of cell surface molecules, which can be identified by monoclonal antibodies. Monoclonal antibody can *differentiate* specific *cluster* of molecules. This system of classification of surface molecules is known as the *Cluster of Differentiation* or CD system**
- **CD** (cluster of differentiation) proteins- molecules on the cells membrane, allow the identification of cells
- The highly important markers in major cell types are – (i) CD4 & CD8 – T cell subsets, (ii) CD 25 – activated T cell, (iii) CD 19 & CD 20 – B cell, (iv) CD 64 & CD 68 – mononuclear phagocytes

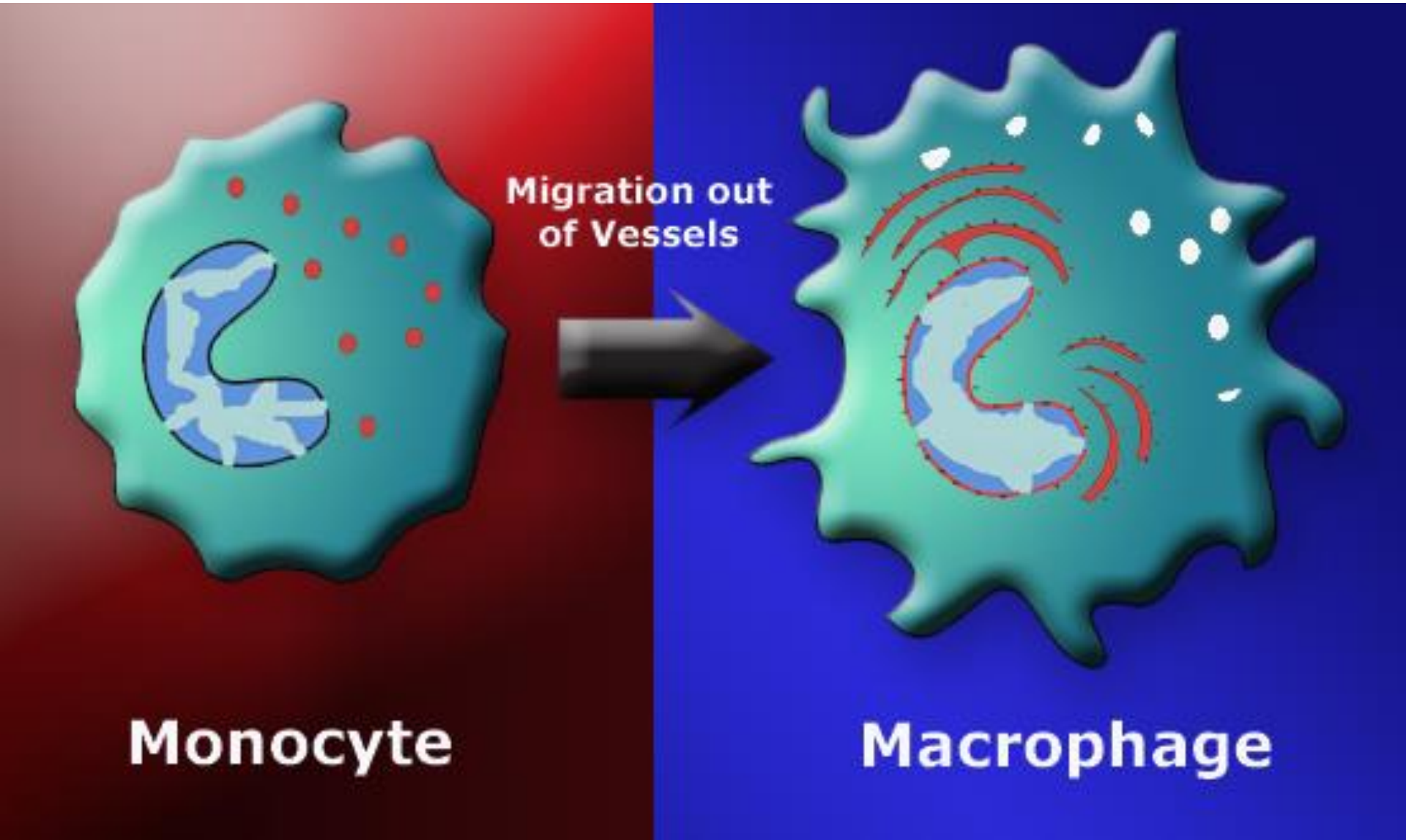
Macrophages- ontogenesis

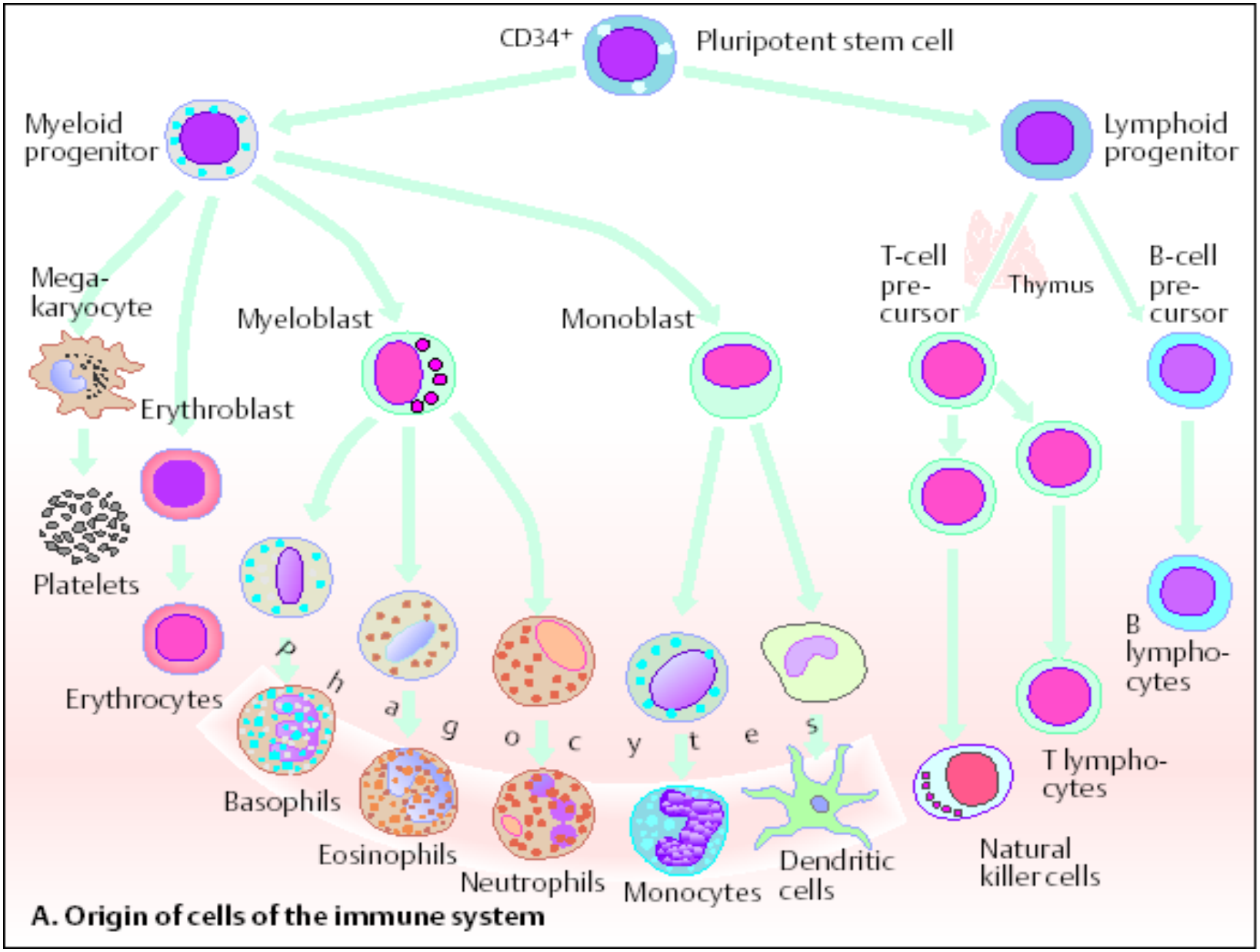
Monocytes- in the blood
Macrophages - in tissues

a monocyte enter damaged tissue through the endothelium of a blood vessel. After migration of monocytes to the tissues they differentiate into different form of macrophages. A macrophages survive several months

- play important roles in innate and adaptive immune responses
- **myeloid progenitor cells** differentiate into the erythrocytic, granulocytic and **monocytic cell lines** and megakaryocytes

Macrophages - terminology

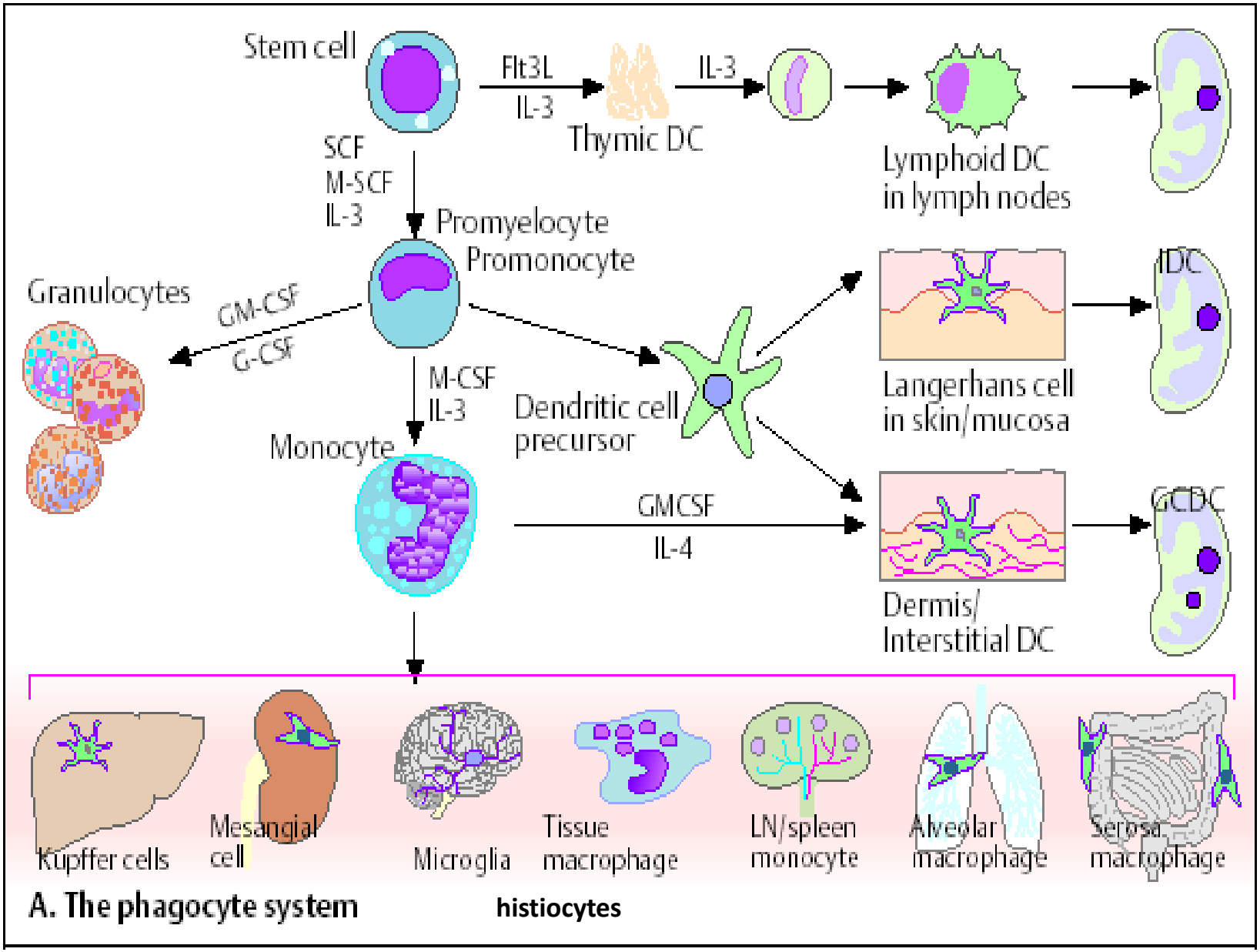



CD34+
Pluripotent stem cell
Myeloid progenitor
Lymphoid progenitor
Mega-karyocyte
Erythroblast
Myeloblast
Monoblast
T-cell pre-cursor
Thymus
B-cell pre-cursor
Platelets
Erythrocytes
Basophils
Eosinophils
Neutrophils
Monocytes
Dendritic cells
B lympho-cytes
T lympho-cytes
Natural killer cells
A. Origin of cells of the immune system

Conversion

The conversion of myeloid precursor cells into monocytes and macrophages is affected by :

- **GM-CSF**: granulocyte-monocyte colony stimulating factor – bone marrow (BM)stromal cells, lymphocytes- production of monocytes from BM
- **M-CSF**: monocyte colony stimulating factor- stromal cells, lymphocytes, endothelial cells, epithelial cells- production and maturation of monocytes
- **IL-3** : lymphocytes- production of monocytes (other blood cells) from BM


Stem cell
Flt3L
IL-3
Thymic DC
IL-3
Lymphoid DC
in lymph nodes
SCF
M-SCF
IL-3
Promyelocyte
Promonocyte
Granulocytes
GM-CSF
G-CSF
M-CSF
IL-3
Monocyte
Dendritic cell
precursor
IDC
Langerhans cell
in skin/mucosa
GMCSF
IL-4
GCDC
Dermis/
Interstitial DC
Kupffer cells
Mesangial
cell
Microglia
Tissue
macrophage
LN/spleen
monocyte
Alveolar
macrophage
Serosa
macrophage
A. The phagocyte system
histiocytes

Macrophage surface molecules

- **MHC gp class I, II** assist in the presentation of epitopes to T lymphocytes
- **CD 35** - complement receptor 1 (**CR 1**), binds complement C3b
- **Receptor** for the **Fc** portion of IgG

Monokines = cytokines produced by macrophages

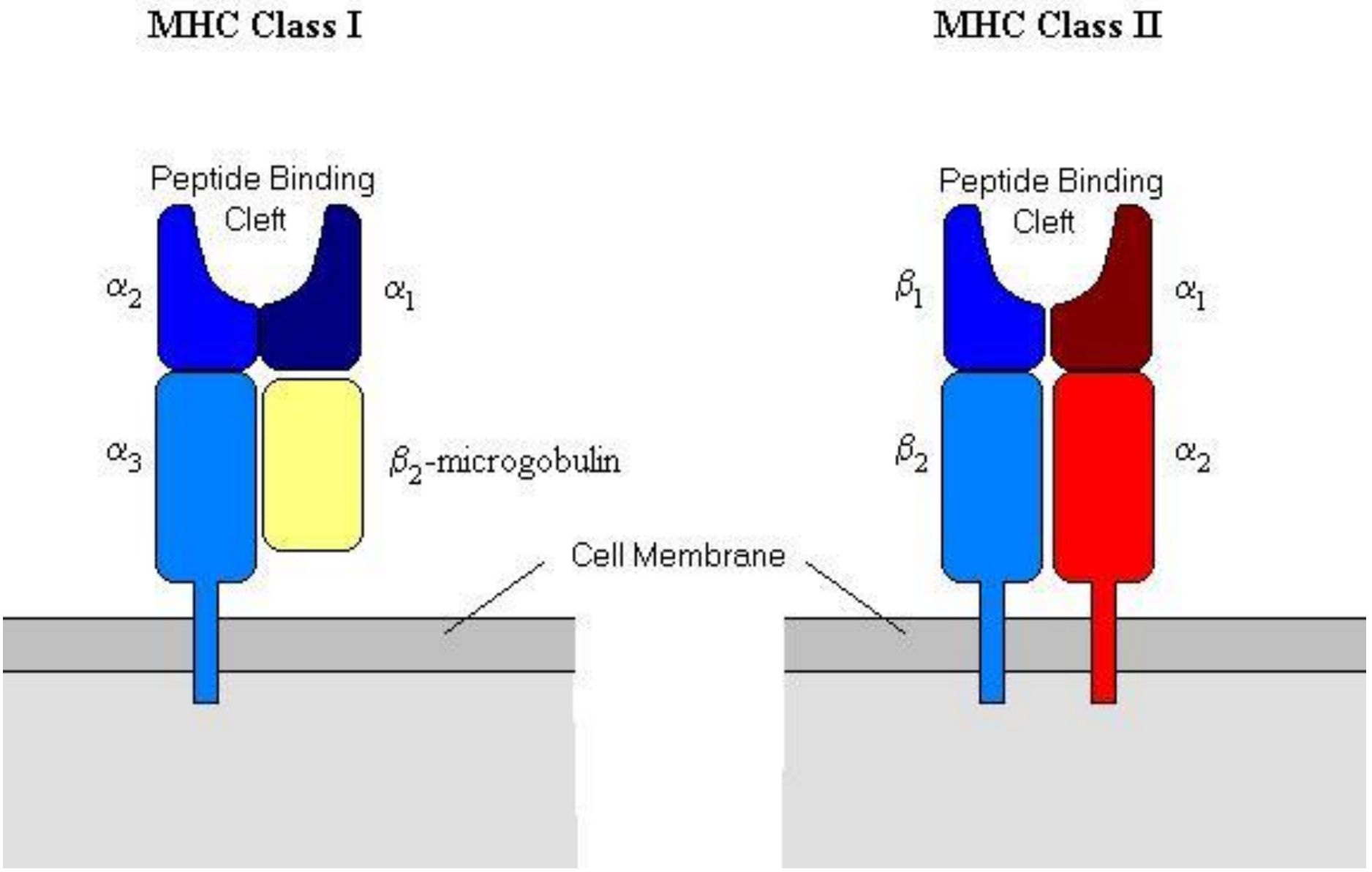
- **IL- 1 α, ß**- stimulate both T and B cells, Ig synthesis, activation of other macrophages, sensitizing cells to IL-2 and IFN
- **TNF- α** - similar in function to IL-1
- **IL- 8** - secreted by activated macrophages
  - chemokine for neutrophils, T cells
- **IL-12** - promotes induction of Th1 cells, inhibits Th2 cells
- **IFN- α**- **activates** host cells to induce enzymes that inhibit protein synthesis needed for viral replication; **increases** expression of MHC gp I class on host cells; **activates** NK cells, T cells, other macrophages

Function of macrophages

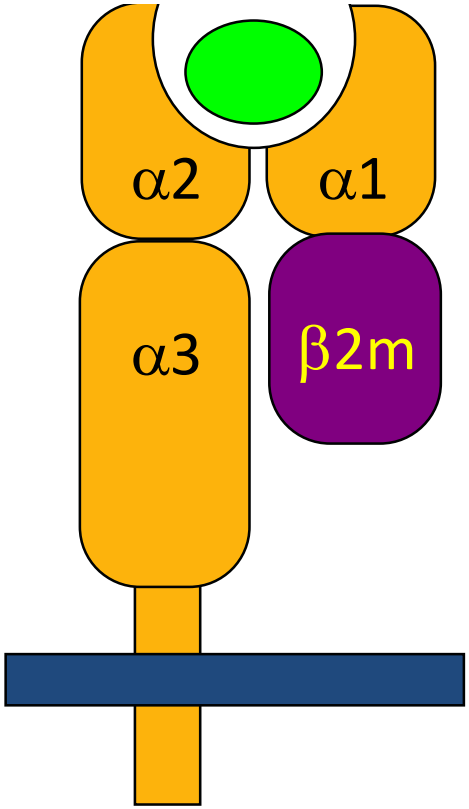
- Phagocytosis
- Production of monokines
- Presentation of epitops with MHC class II
- Presentation of epitops with MHC class I

MHC

- Major Histocompatibility Complex
  - Cluster of genes found in all mammals
  - Its products play role in discriminating self/non-self
  - Participant in both humoral and cell-mediated immunity
- MHC Act As Antigen Presenting Structures
- In Human MHC is found on chromosome 6
  - Referred to as HLA complex in Human
- In Mice MHC is found on chromosome 17
  - Referred to as H-2 complex in Mouse


MHC Class I
Peptide Binding Cleft
α_2
α_1
α_3
β_2-microgobulin
Cell Membrane
MHC Class II
Peptide Binding Cleft
β_1
α_1
β_2
α_2

Overall structure of MHC class I molecules



MHC-encoded α-chain of 43kDa

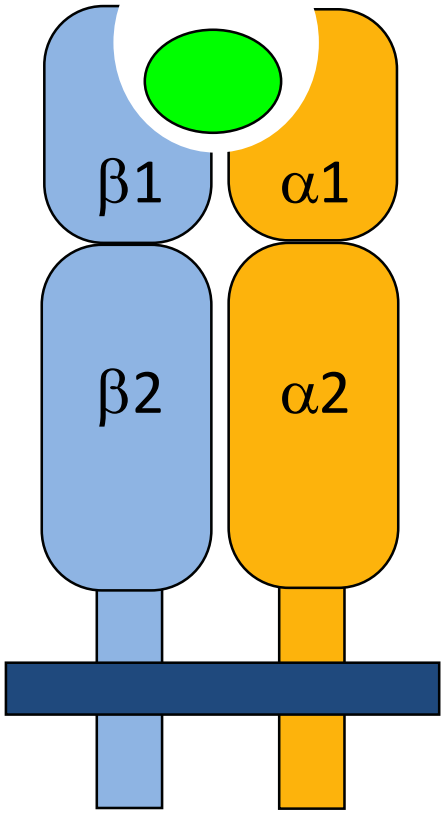
α-chain anchored to the cell membrane

Peptide antigen in a groove formed from a pair of α-helicies on a floor of anti-parallel β strands

β2-microglobulin, 12kDa, non-MHC encoded, non-transmembrane, non covalently bound to α-chain

α3 domain & β2m have structural & amino acid sequence homology with Ig C domains Ig GENE SUPERFAMILY

Overall structure of MHC class II molecules



MHC-encoded, α-chain of 34kDa
and a β-chain of 29kDa

α and β chains anchored to the cell membrane

No β-2 microglobulin

Peptide antigen in a groove formed from a pair of α-helicies on a floor of anti-parallel β strands

α2 & β2 domains have structural & amino acid sequence homology with Ig C domains Ig GENE SUPERFAMILY

Differential distribution of MHC molecules

Tissue	MHC class I	MHC class II
T cells	+++	+/-
B cells	+++	+++
Macrophages	+++	++
Other APC	+++	+++
Thymus epithelium	+	+++
Neutrophils	+++	-
Hepatocytes	+	-
Kidney	+	-
Brain	+	-
Erythrocytes	-	-

Cell activation affects the level of MHC expression.

The pattern of expression reflects the function of MHC molecules:

- **Class I is involved in the regulation of anti-viral/parasite immune responses**
- **Class II involved in regulation of the cells of the immune system**

Anucleate erythrocytes can not support virus replication - hence no MHC class I.

Some pathogens exploit this - e.g. *Plasmodium* species.

TABLE 7-2 Peptide binding by class I and class II MHC molecules

	Class I molecules	Class II molecules
Peptide-binding domain	$\alpha 1/\alpha 2$	$\alpha 1/\beta 1$
Nature of peptide-binding cleft	Closed at both ends	Open at both ends
General size of bound peptides	8–10 amino acids	13–18 amino acids
Peptide motifs involved in binding to MHC molecule	Anchor residues at both ends of peptide; generally hydrophobic carboxyl-terminal anchor	Anchor residues distributed along the length of the peptide
Nature of bound peptide	Extended structure in which both ends interact with MHC cleft but middle arches up away from MHC molecule	Extended structure that is held at a constant elevation above the floor of MHC cleft

Presentation epitopes with MHC gp class II

- After **endocytosis** and **degradation** of the antigen, preservation of its epitopes follows

- epitope is coupled with the MHC gp class II- moved to the cell surface and contact the T-cell receptor

- **MHC** (major histocompatibility complex) = complex of genes that governs the production of the major histocompatibility antigens - in humans are termed HLAs (human leukocyte antigens)

Ag Presentation via Class II MHC Proteins

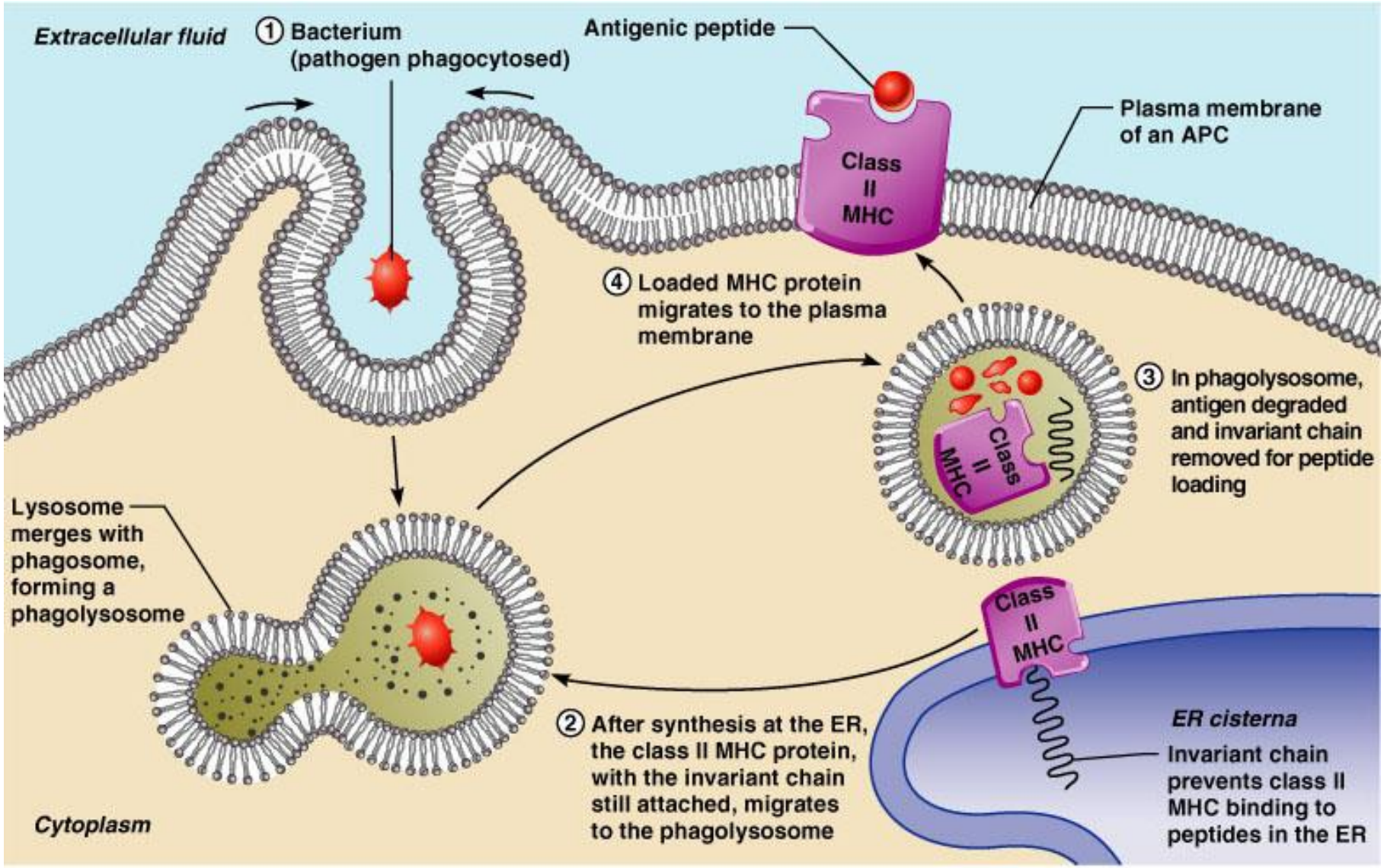


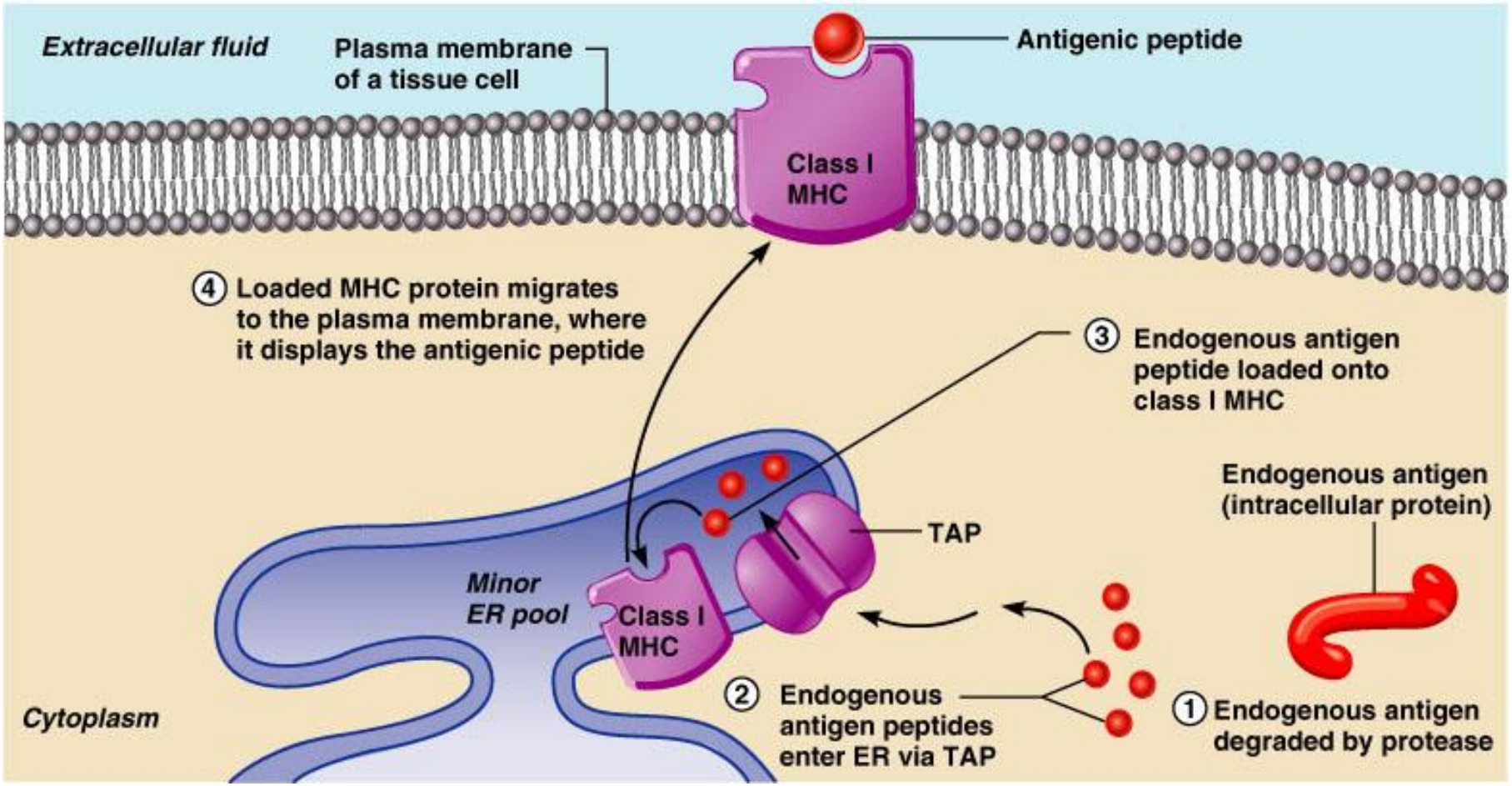
Figure 21.15b

Presentation epitopes with MHC gp class I

- intracelular parasites are **degradeted** in proteasomes of macrophages

- their **peptides** are coupled to **TAP** (transporters associated with antigen processing molecules 1,2) that carry the epitope and **MHC gp class I** to the cell surface- protect epitopes from phagocytic destruction

Ag Presentation via Class I MHC Proteins



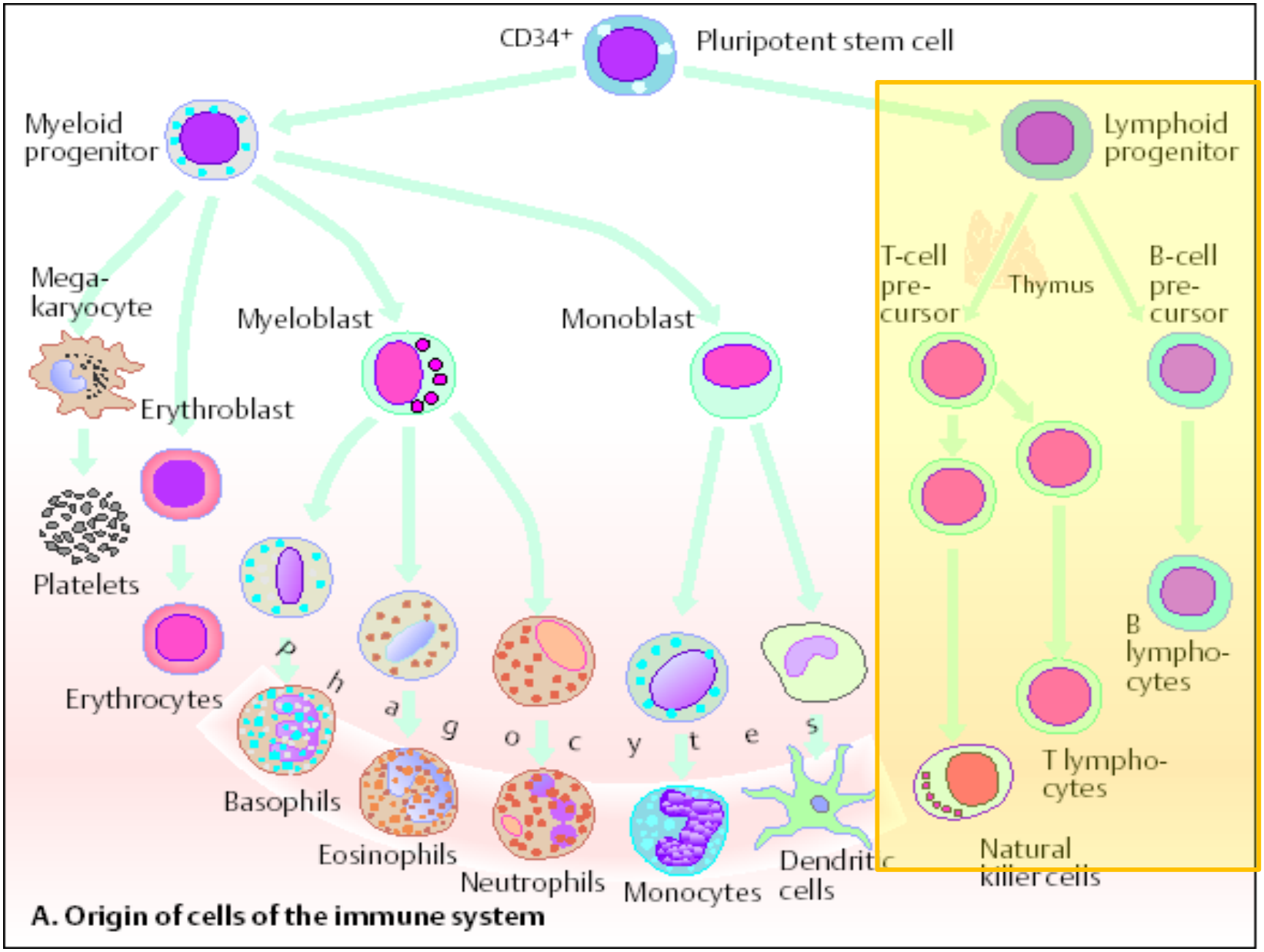
T-lymphocytes, ontogenesis, surface markers. Subpopulations of T-lymphocytes and their functions.

T lymphocytes- ontogenesis

- The undifferentiated stem cell in bone marrow (BM) gives rise to the lymphoid precursor cell which matures into 3 types of lymphocytes:

- **T lymphocytes**
- **B lymphocytes**
- **Natural killer (NK) cell**

- Pro-thymocytes come to the thymus where continue the maturation into T lymphocytes
- Maturation of B lymphocytes continue in BM


CD34+
Pluripotent stem cell
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Erythroblast
Platelets
Erythrocytes
Basophils
Eosinophils
Neutrophils
Monocytes
Dendritic cells
B lympho-cytes
T lympho-cytes
Natural killer cells
A. Origin of cells of the immune system

CD proteins in T-cell

- allow an identification of T-cell subsets
- **CD 2** = adhesion molecule
- **CD 3** = important in intracellular signaling to initiate an immune response; closely associated with TCR
- **CD 5,7**
- **CD 4,8** = are expresed on subclasses of mature T cells; CD4 reacts with MHC gp II.class),CD8 reacts with MHC gp I. class on macrophages
- **CD 28-** receptor for co-stimulator molecules CD80 and 86

Maturation of T lymphocytes

Consist of three types of processes:

- Proliferation of immature cells
- Expression of antigen receptors genes
- Selection of lymphocytes that express useful antigen receptor (TCR)

The pathway of T cell development

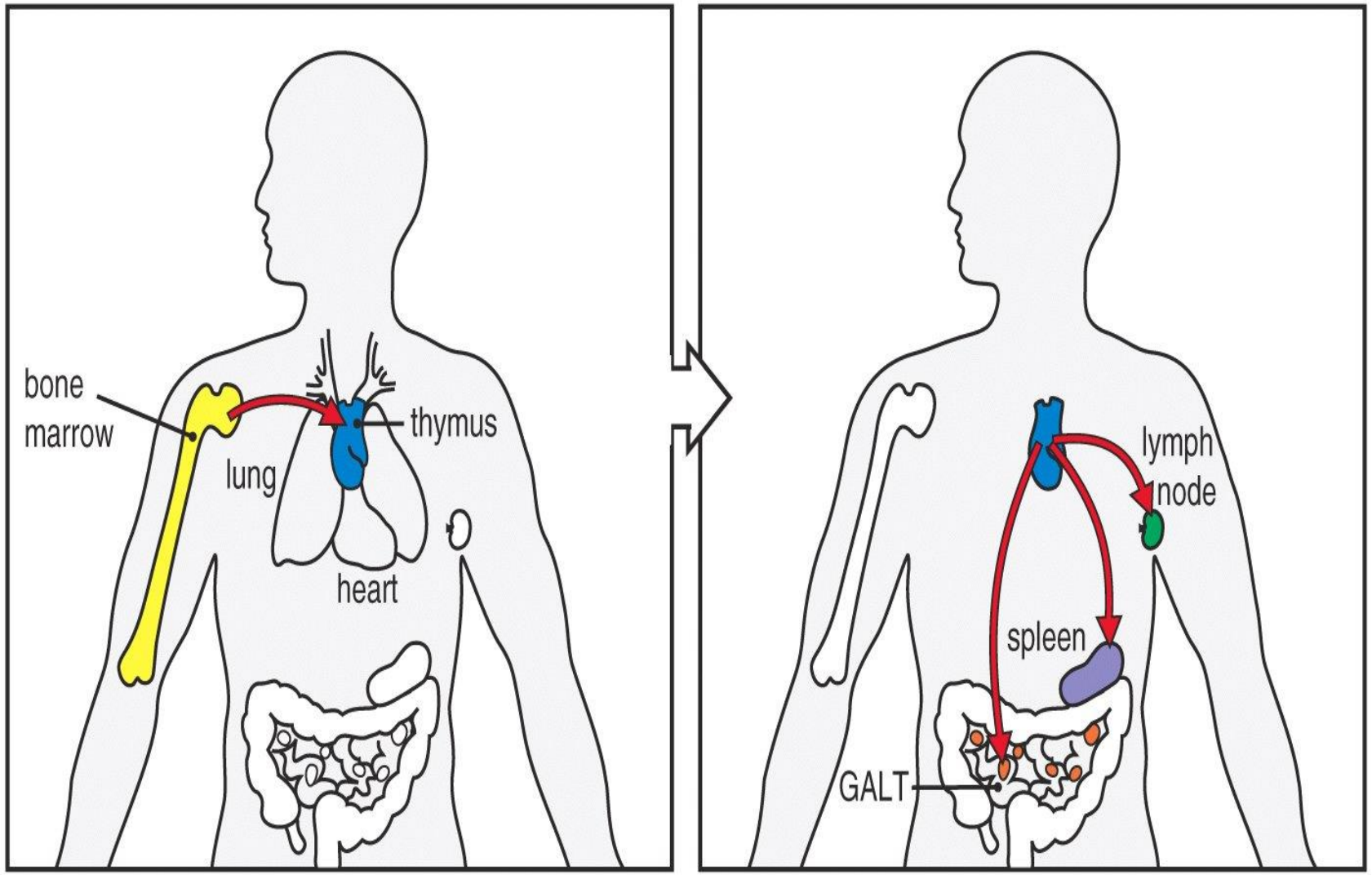
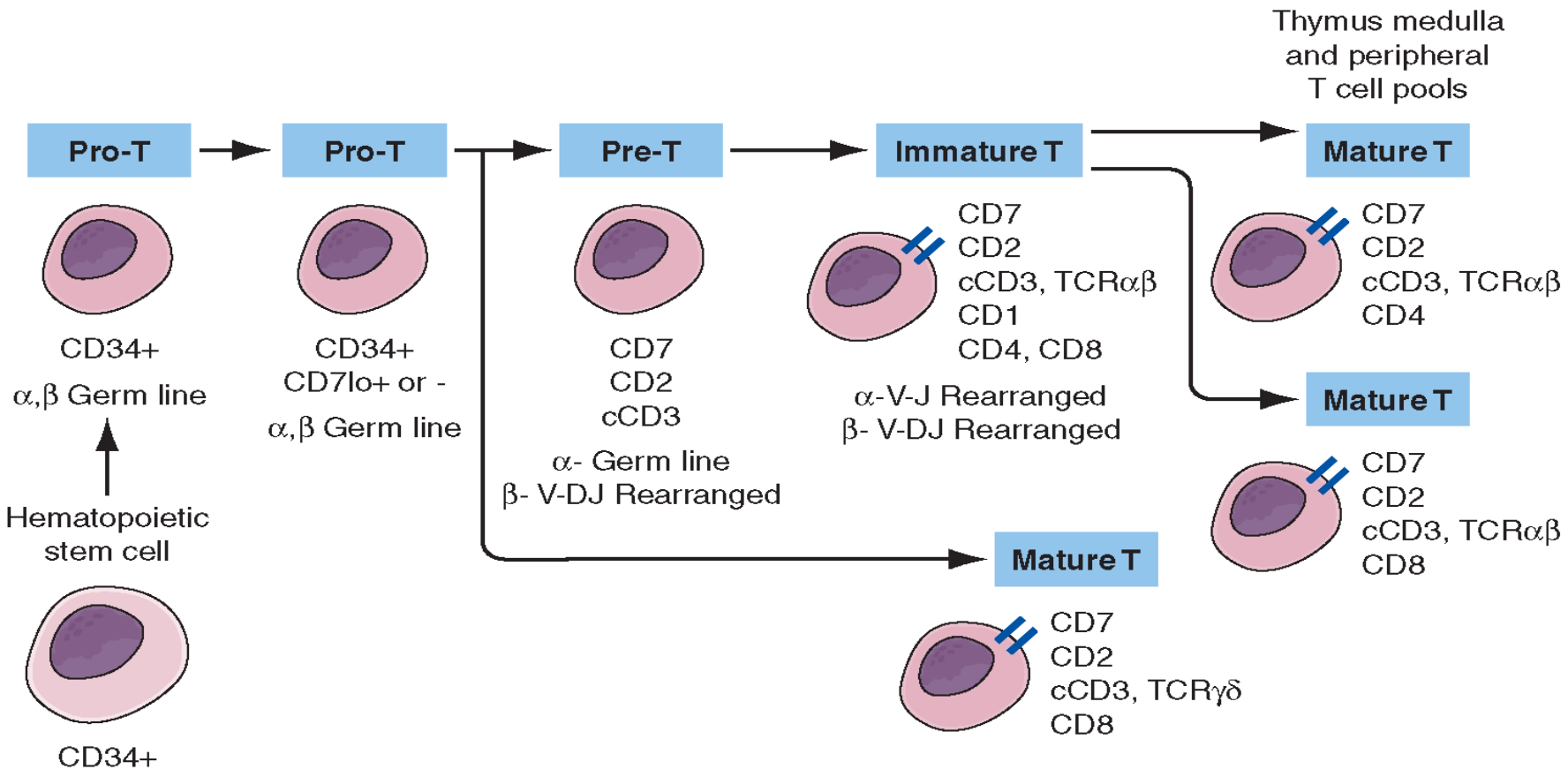
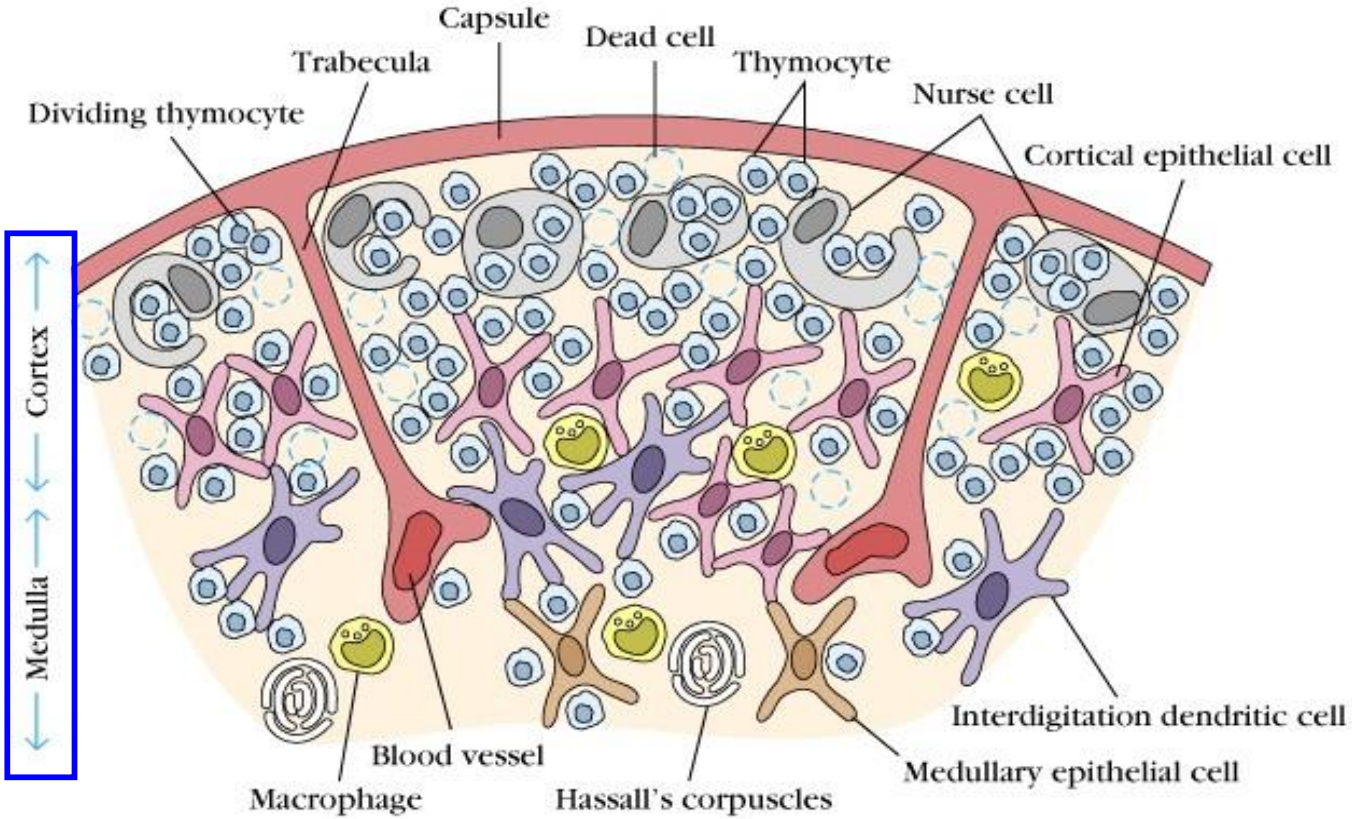


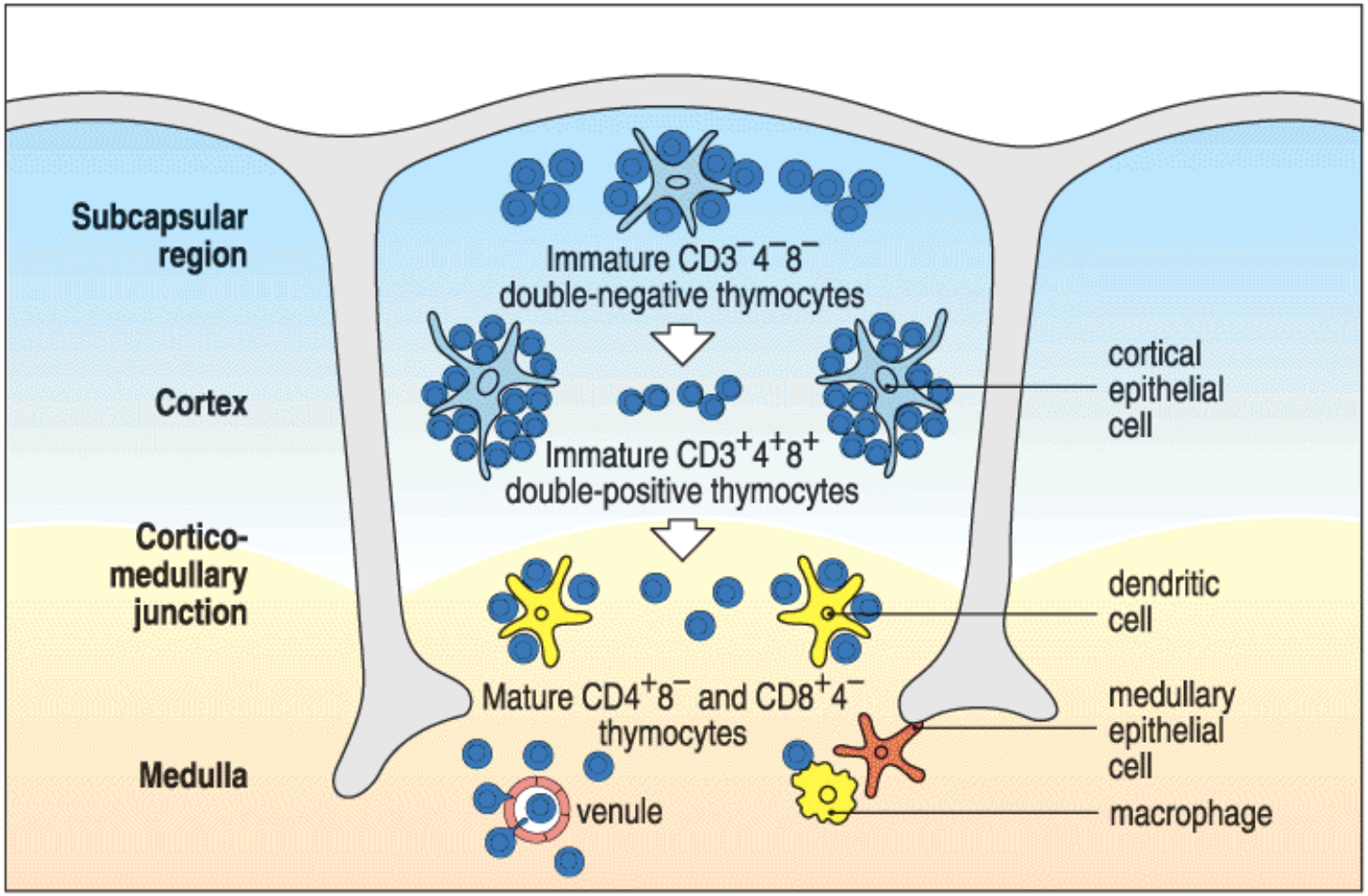
Figure 5-1 The Immune System, 2/e (© Garland Science 2005)


Thymus medulla
and peripheral
T cell pools
Pro-T
Pro-T
Pre-T
Immature T
Mature T
Mature T
Mature T
CD34+
α,β Germ line
Hematopoietic
stem cell
CD34+
CD34+
CD7lo+ or -
α,β Germ line
CD7
CD2
cCD3
α- Germ line
β- V-DJ Rearranged
CD7
CD2
cCD3, TCRαβ
CD1
CD4, CD8
α-V-J Rearranged
β- V-DJ Rearranged
CD7
CD2
cCD3, TCRαβ
CD4
CD7
CD2
cCD3, TCRαβ
CD8
CD7
CD2
cCD3, TCRγδ
CD8

T cell development occurs in the thymus



Thymocytes at different developmental stages are found in distinct parts of the thymus



TCR (T- cells receptor)

- Antigen receptors are encoded by several gene segments that **recombine** during lymphocyte maturation
- Heterodimer consisting of 2 non-identical polypeptide chains linked together by disulfide bonds
- > 95% T cells express the αß heterodimer, 5% γδ
- TCR heterodimer is noncovalently associated with the γ,δ,ε chains of the CD3 molecule
- **COMPLEX TCR- CD3** makes contact with both the **Ag and MHC gp**

TCRαβ lineage comprises the majority of T cells

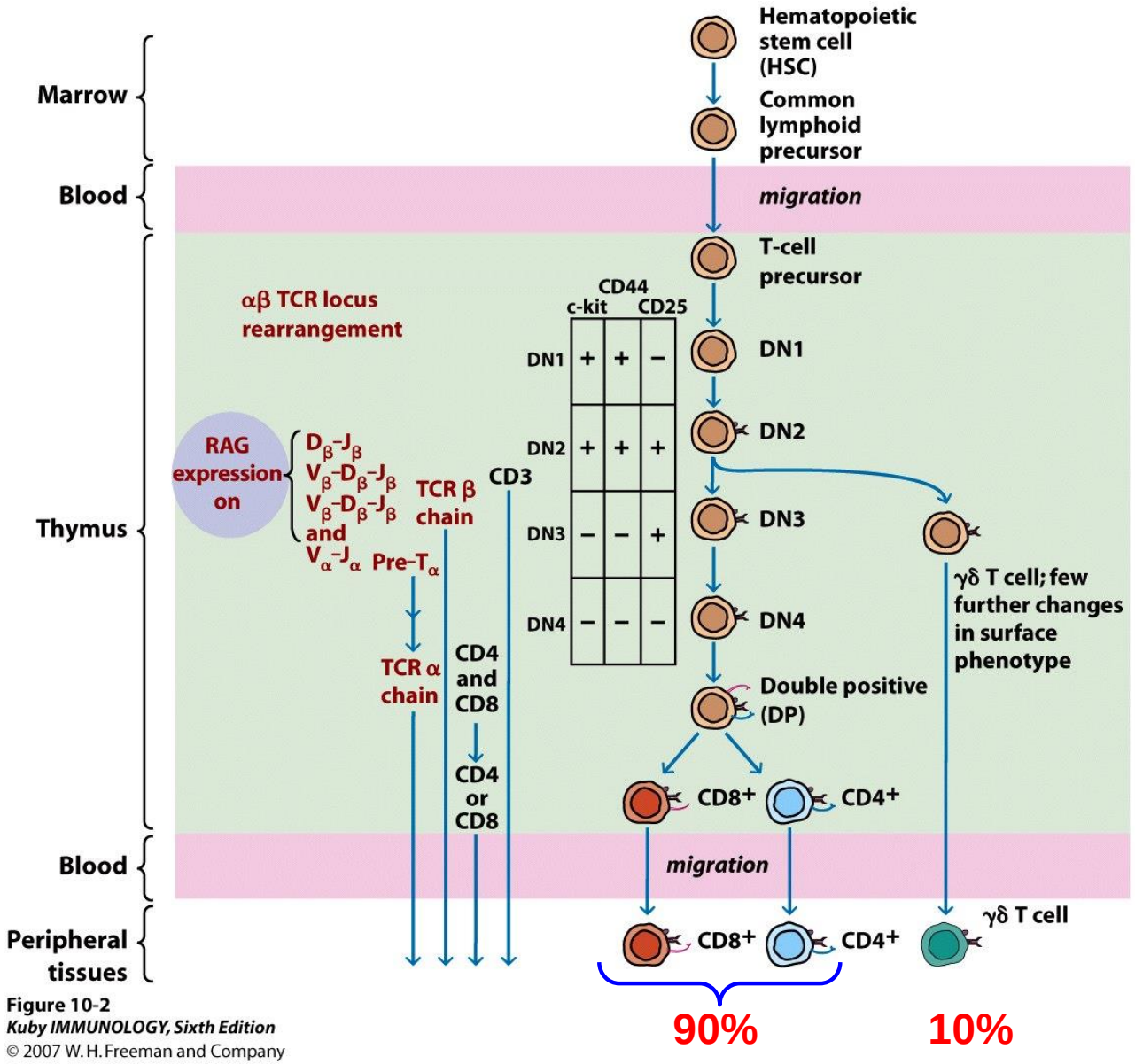


Figure 10-2
Kuby IMMUNOLOGY, Sixth Edition

γδ T cells are favored during early fetal development

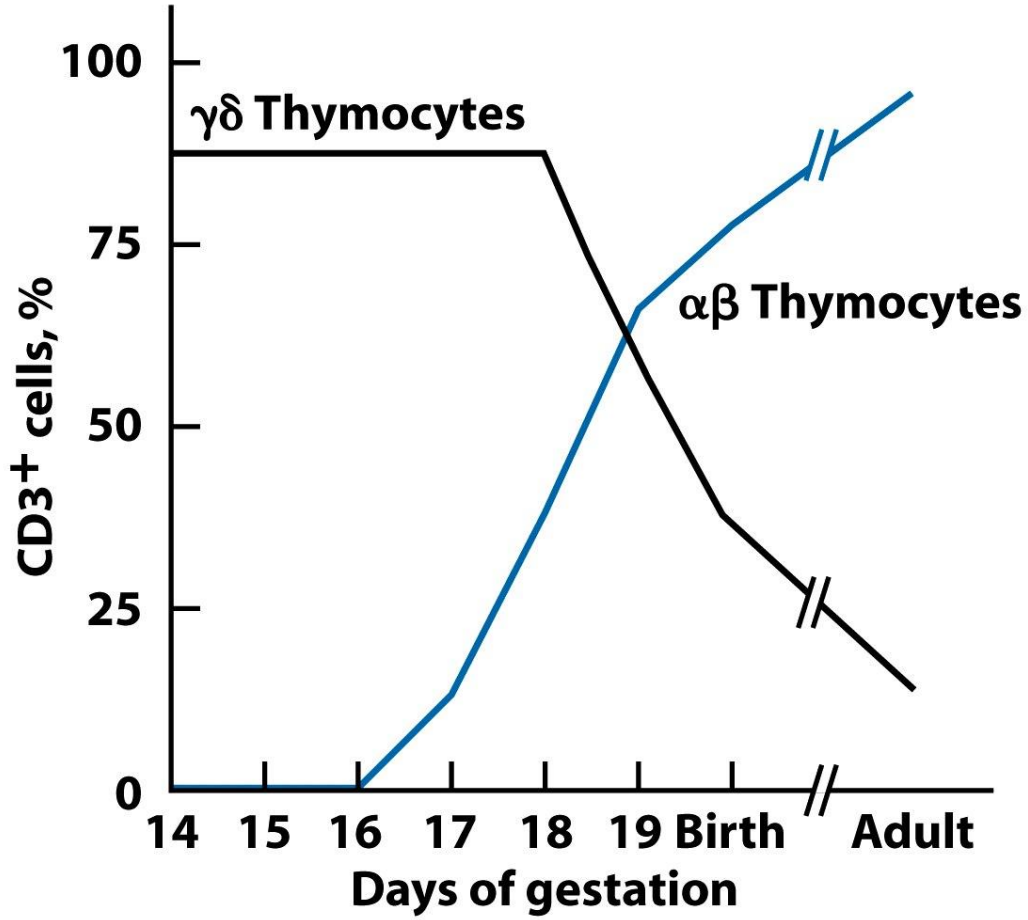


Figure 10-3
Kuby IMMUNOLOGY, Sixth Edition

T cell development involves the sequential generation, assembly and testing of the newly rearranged TCR

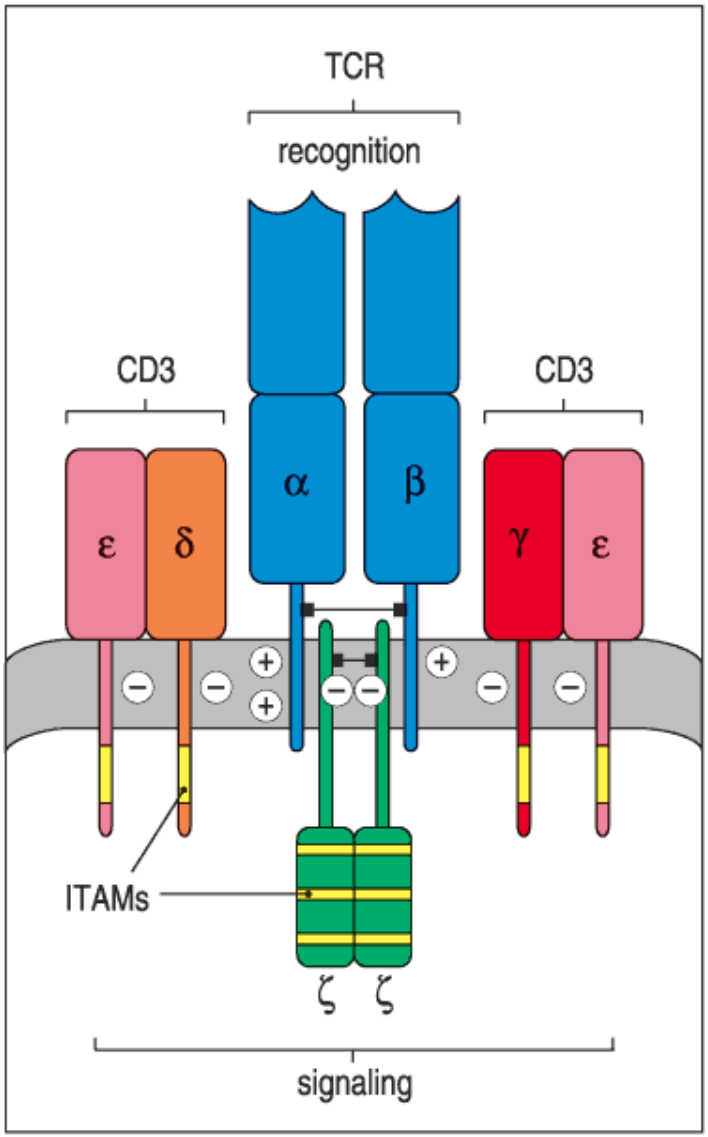
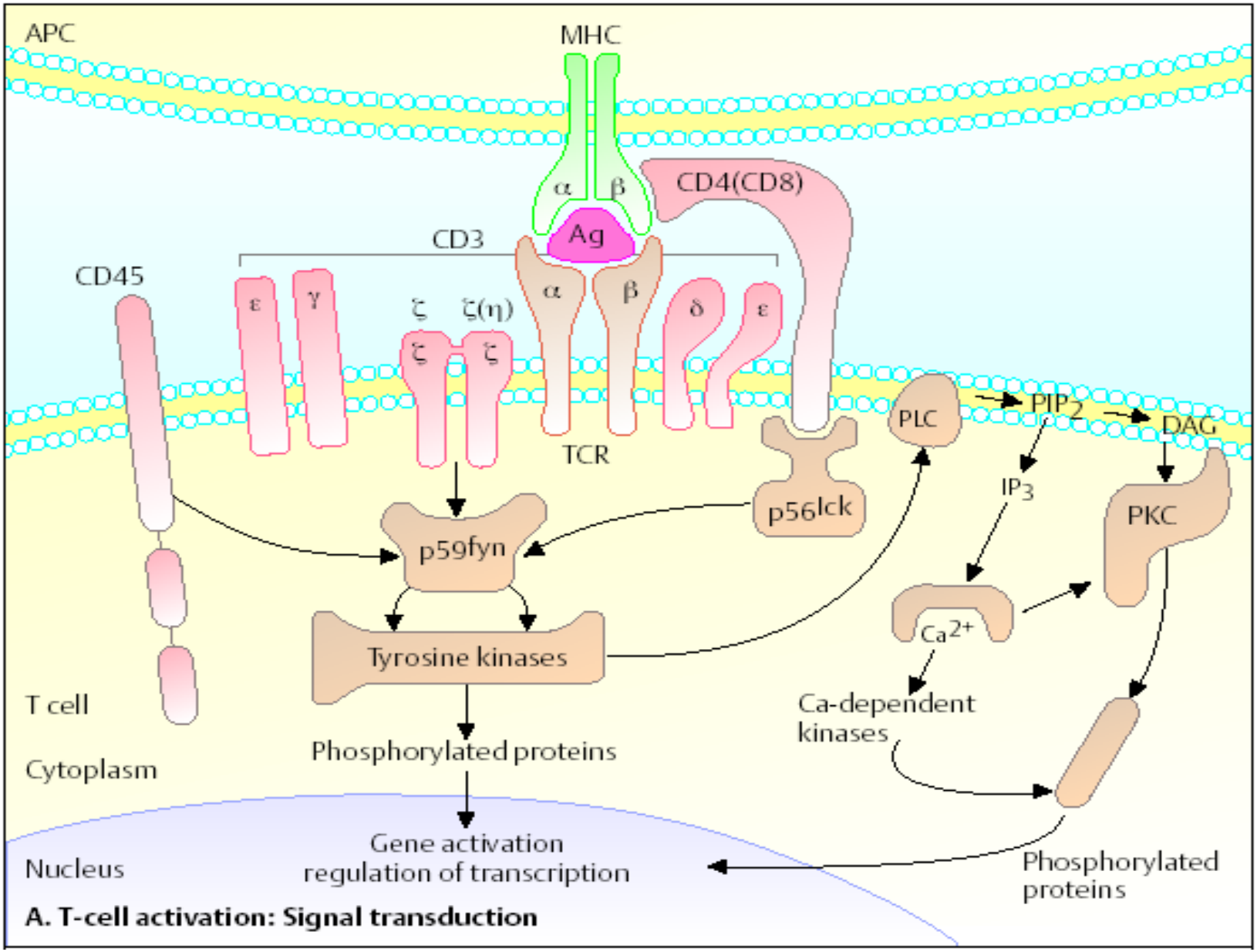


Fig 6.8 © 2001 Garland Science


APC
MHC
α
β
CD4(CD8)
Ag
CD3
CD45
ε
γ
ζ
ζ(η)
ζ
ζ
α
β
δ
ε
TCR
PLC
PIP2
DAG
IP3
PKC
p56lck
p59fyn
Ca2+
Tyrosine kinases
T cell
Cytoplasm
Phosphorylated proteins
Ca-dependent kinases
Nucleus
Gene activation
regulation of transcription
Phosphorylated proteins
A. T-cell activation: Signal transduction

Subpopulation of T cells

- Subpopulation of T cells have been defined according to their particular function and their CD membrane markers

- **Cytotoxic T lymphocytes = Tc;CD8+**

  - recognize the foreign epitope in association with **class I MHC** molecules

- **Helper T-lymphocytes = Th; CD4+**

  - recognize the epitopes in association with **class II MHC** molecules

Cytotoxic T lymphocytes (Tc;**CD8+**)

- cause lysis of target cells; are active against tumors, virus-infected cells, transplanted allogenetic tissue
- release TNF- depresses proteosynthesis
- recognize the foreign epitope in association with **class I MHC** molecules
- destroy their target cells by releasing perforin (create poresin the cell membrane and cytoplasm escapes) and granzymes (degrading essential macromolecules)

Helper T-lymphocytes (Th; CD4+)

- recognize the epitopes in association with II MHC p II.class
- help B cells to produce antibodies and help phagocytes to destroy ingested microbes
- subsets of Th cells: Th1, Th2 cells

Helper T Cells (T_H)

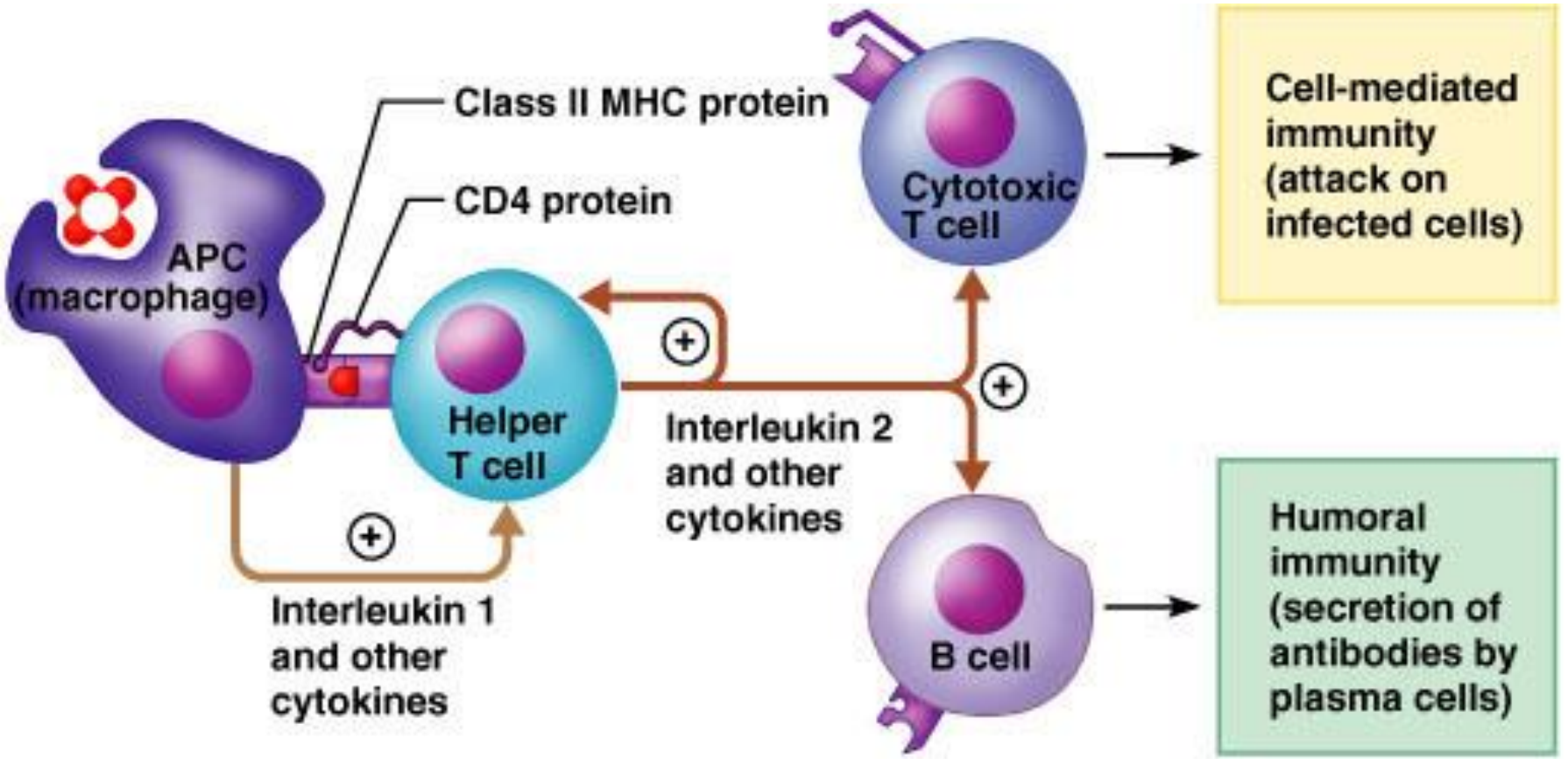
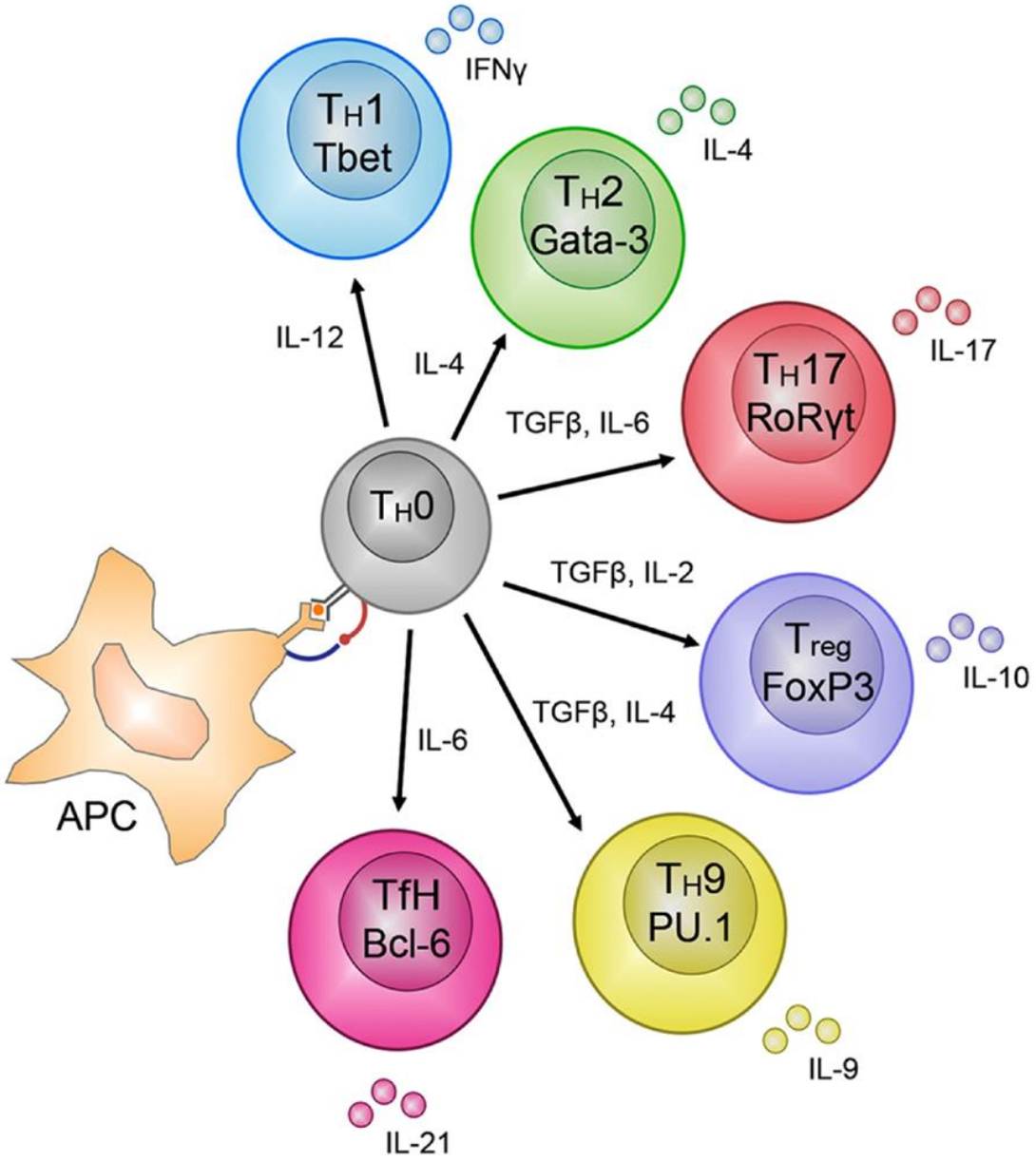


Figure 21.17a


IFNγ
TH1
Tbet
IL-4
TH2
Gata-3
IL-12
IL-4
IL-17
TH17
RoRγt
TGFβ, IL-6
TH0
TGFβ, IL-2
Treg
FoxP3
IL-10
APC
IL-6
TGFβ, IL-4
TfH
Bcl-6
TH9
PU.1
IL-9
IL-21

- Viral infections or microbes that infect macrophages or natural killer (NK) cells elicit a Th1 response. Th1 lymphocytes secrete interferon gamma (IFN-γ) and tumor necrosis factor beta (TNF-β).

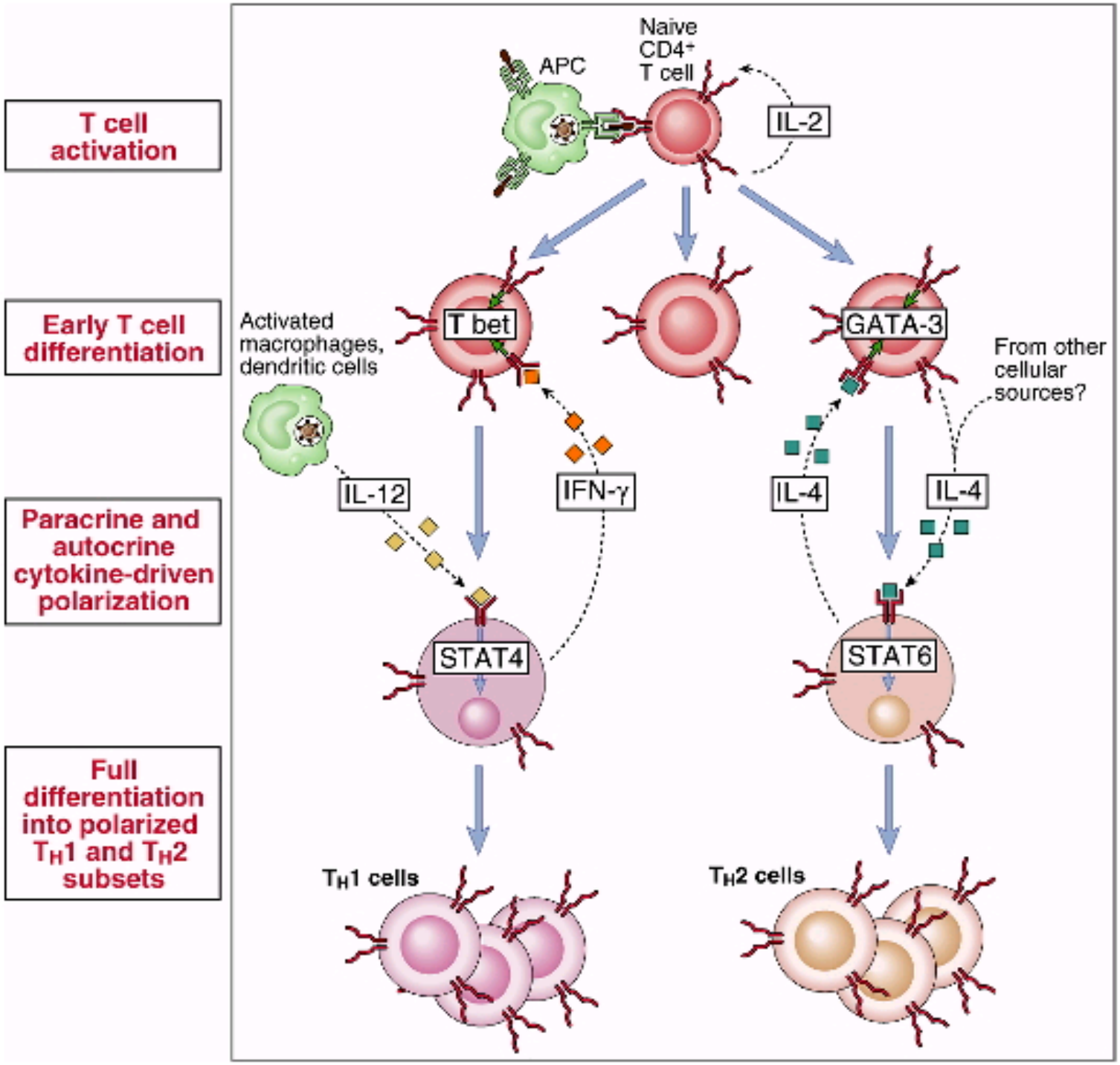
- A Th2 response elicits Th2 lymphocytes in response to helminths, allergens, and extracellular microbes. Th2 lymphocytes produce cytokines interleukin (IL)-4, 5, and 13, among others.

- Th17 cells are lymphocytes that produce IL-17 to recruit neutrophils and macrophages and to trigger an inflammatory response in different organs in order to remove extracellular pathogens from the body.

- Additionally, there is a regulatory arm characterized by regulatory T cells (Tregs) that produce IL-10 to downregulate all other immune arms with the objective of preventing an exacerbated and injury-provoking immune response. Achieving a balance between all these arms is what enables an adequate immune response to fight pathogens and prevents immune-mediated diseases

TH1
vs
TH2

Property	T_H1 subset	T_H2 subset
Cytokines produced		
IFN-γ	+++	–
IL-4, IL-5, IL-13	–	+++
IL-10	+/–	++
IL-3, GM-CSF	++	++
Cytokine receptor expression		
IL-12R β chain	++	–
IL-18R	++	–
Chemokine receptor expression		
CCR4	+/–	++
CXCR3, CCR5	++	+/–
Ligands for E- and P-selectin	++	+/–
Antibody isotypes stimulated	IgG2a (mouse)	IgE, IgG1 (mouse)/IgG4 (humans)
Macrophage activation	+++	–


T cell activation
Early T cell differentiation
Paracrine and autocrine cytokine-driven polarization
Full differentiation into polarized T_H1 and T_H2 subsets
APC
Naive CD4+ T cell
IL-2
T bet
GATA-3
Activated macrophages, dendritic cells
From other cellular sources?
IL-12
IFN-γ
IL-4
IL-4
STAT4
STAT6
T_H1 cells
T_H2 cells

Th1 cells

secrete:

- **INF-γ** (gamma interferon) : activates macrophages to become more effective at killing phagocytosed microbes, supresses the development of Th2 cells
- **IL- 2 :** stimulates survival and proliferation of T cells, called T-cell growth factor
- **TNF** (tumor necrosis factor)- stimulates the recruitment of neutrophils and monocytes to sites of infection, activates these cells to eradicate microbes
- **IL-3 :** promotes expansion of immature marrow progenitors of all blood cells
- **GM-CSF :** acts on progenitors in the bone marrow to increase production of neutrophils and monocytes

Th2 cells

secrete:

- **IL-4** : induces differentiation of Th2 cells from naive CD4+ precursors, stimulation of IgE production by B cells
- **IL-5** : activates mast cells
- **IL-6** : stimulates the synthesis of acute phase proteins by hepatocytes
- **IL-10** : inhibits activated macrophages, supresses Th1 production
- **IL-3, GM-CSF**

Regulatory T cells

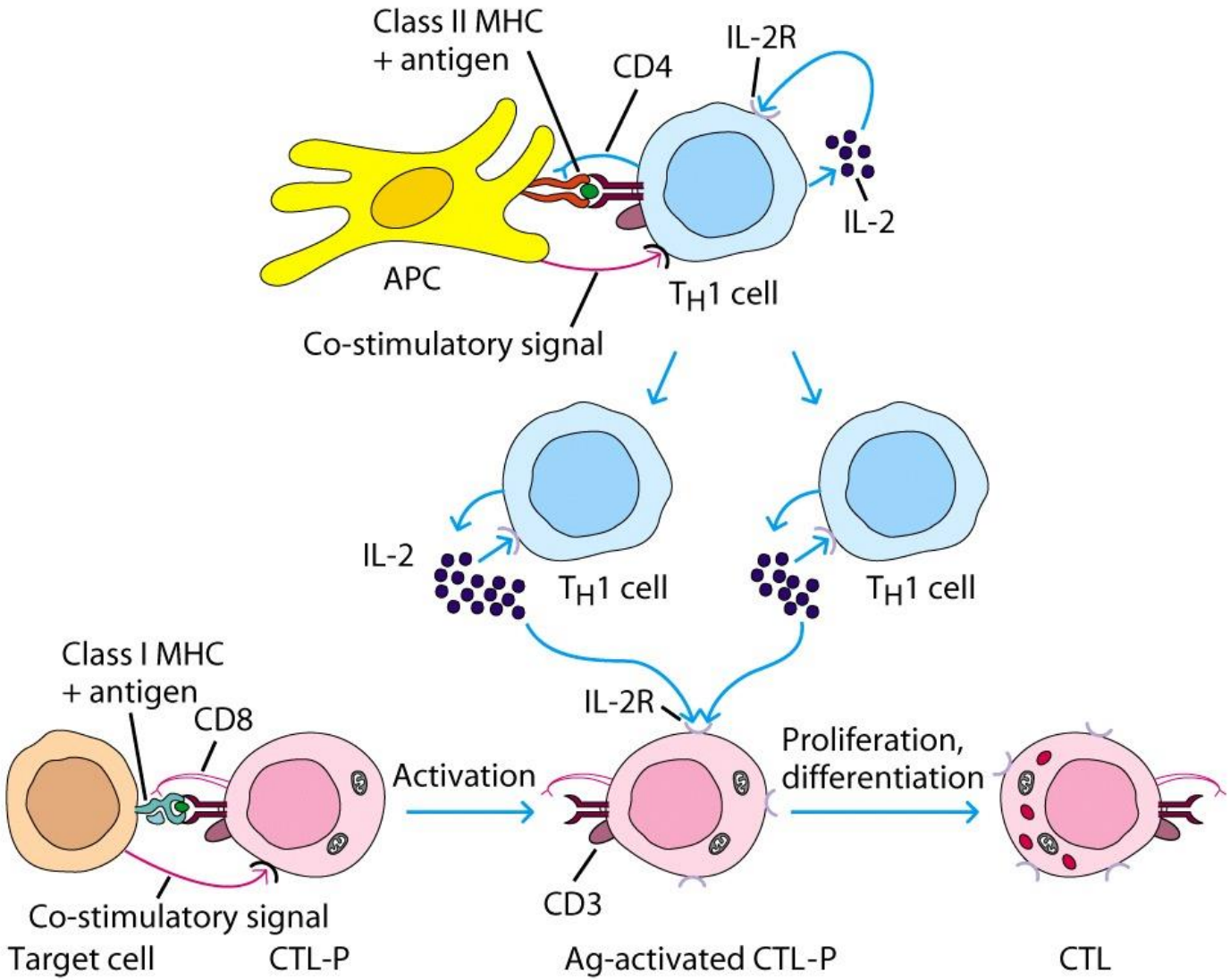
- Express CD4, CD25, FoxP3
- Regulate the activation or effector function of other T cells
- Are necessary to maintain tolerance to self antigens

Cell Mediated Immunity

- General responses by CMI, include:
  - Facilitate innate immune response to bacteria
  - Anti-viral
  - Anti-fungal
  - Anti-tumor
  - Transplantation rejection

Cytotoxic T Cells

- CTLs Recognize Cells That Have Been infected
  - Virus
  - Transformed to tumor
- CTL Activation Is Divided Into 2 Phases
  - Activation and differentiation of naïve CTL
  - Effector recognizes Class I MHC/peptide and destroys target
- Naïve CTLs Cannot Kill
  - Referred to as CTL-Ps (precursors)
  - 3 signals needed for activation
    - Ag specific signal thru TCR/MHC I+Ag
    - Co-stimulatory signal CD28(CTL)/B7 (APC)
    - IL-2 signaling inducing proliferation (CTL-P do not express IL-2 R)
      - IL-2 is provided by T_{H1} or CTL-P itself
      - IL-2R is expressed only after activation



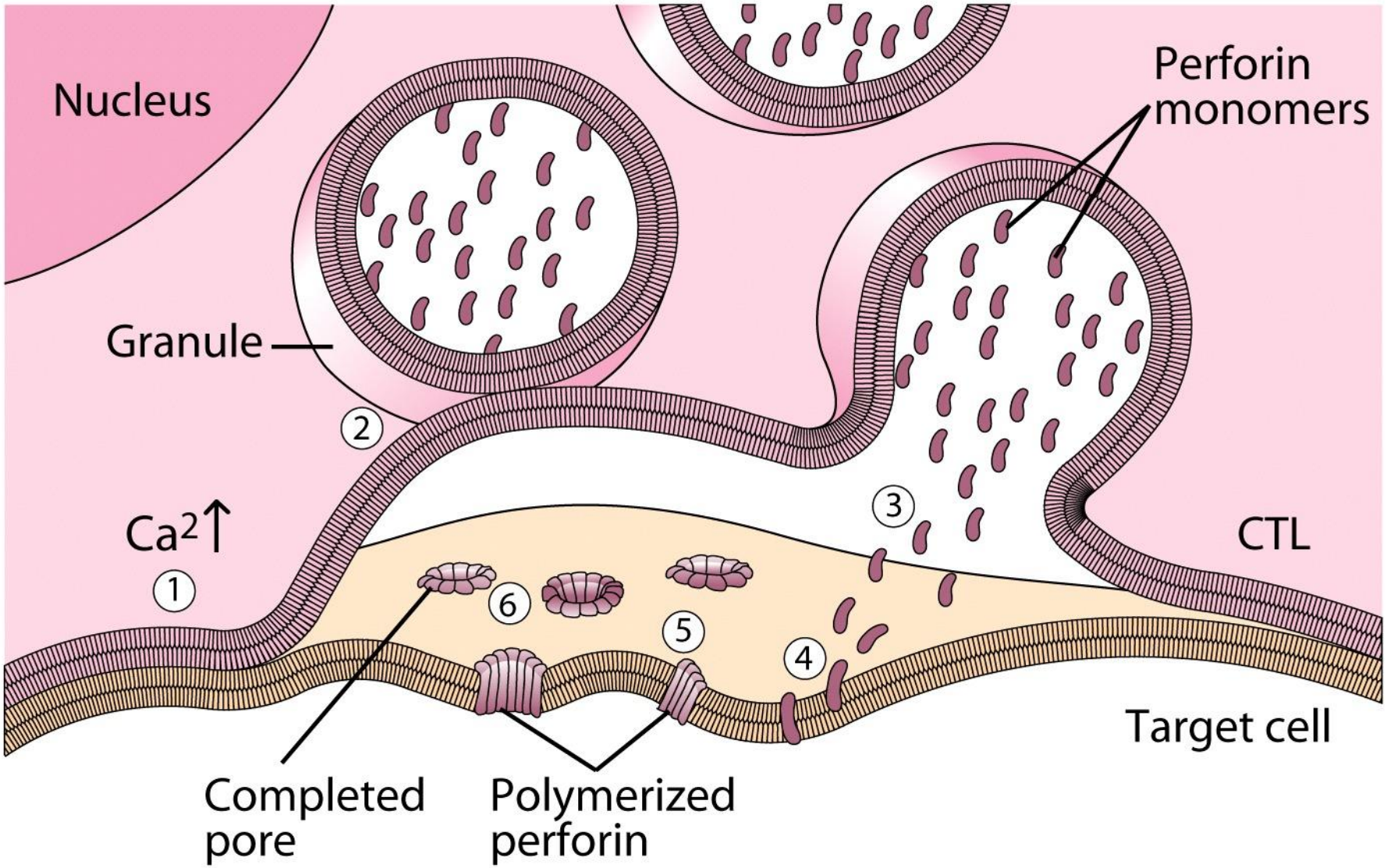
IL-2R expression	–	+	+
IL-2 expression	–	±	±
Proliferation	–	–	+
Effector cytotoxic function	–	–	+

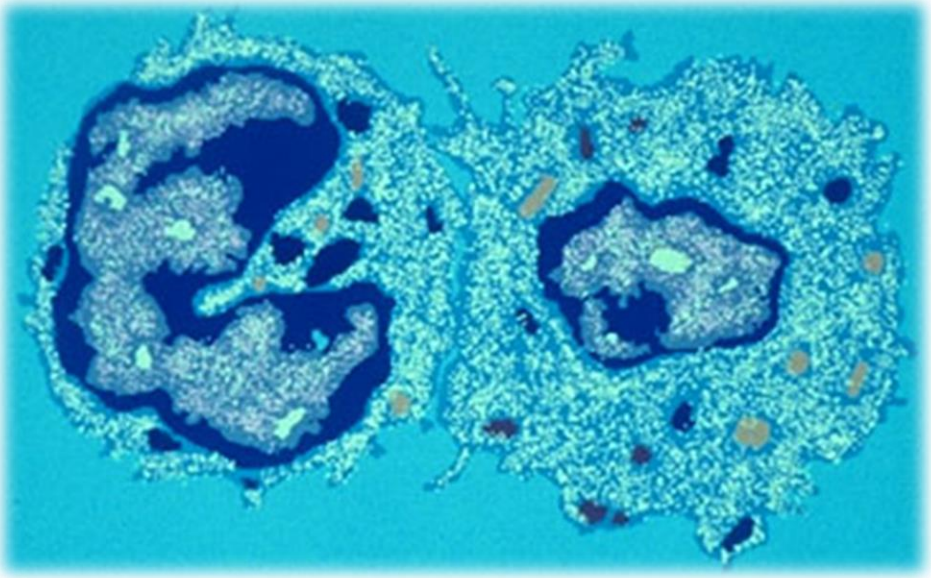
Cytotoxic T Cells

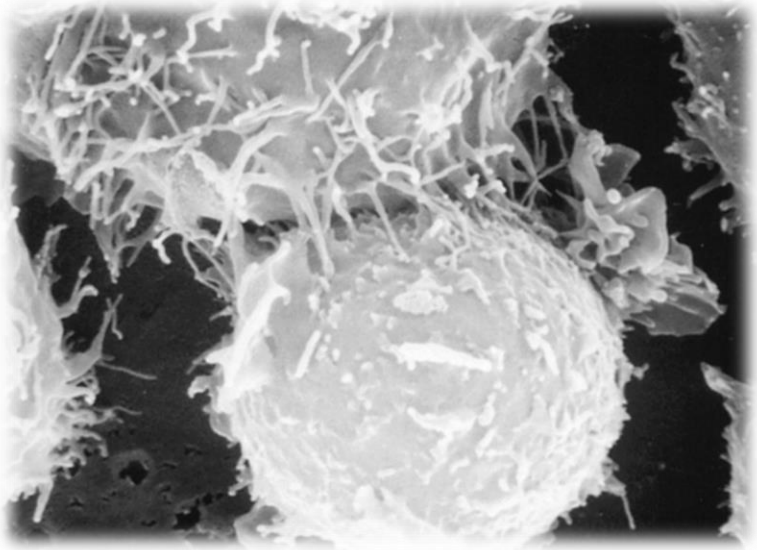
- T_{H1} And CTL Collaborate To Induce Effector CTL
  - IL-2 seems to be crucial (knock out data)
  - Lack of IL-2R In CTL-P Ensures Ag Specificity
- Upon clearance of antigen CTLs undergo apoptosis
- T_{H1} Induce up-regulation of co-stimulatory molecules on APCs enhancing APC/CTL Co-stimulation

How CTLs Kill

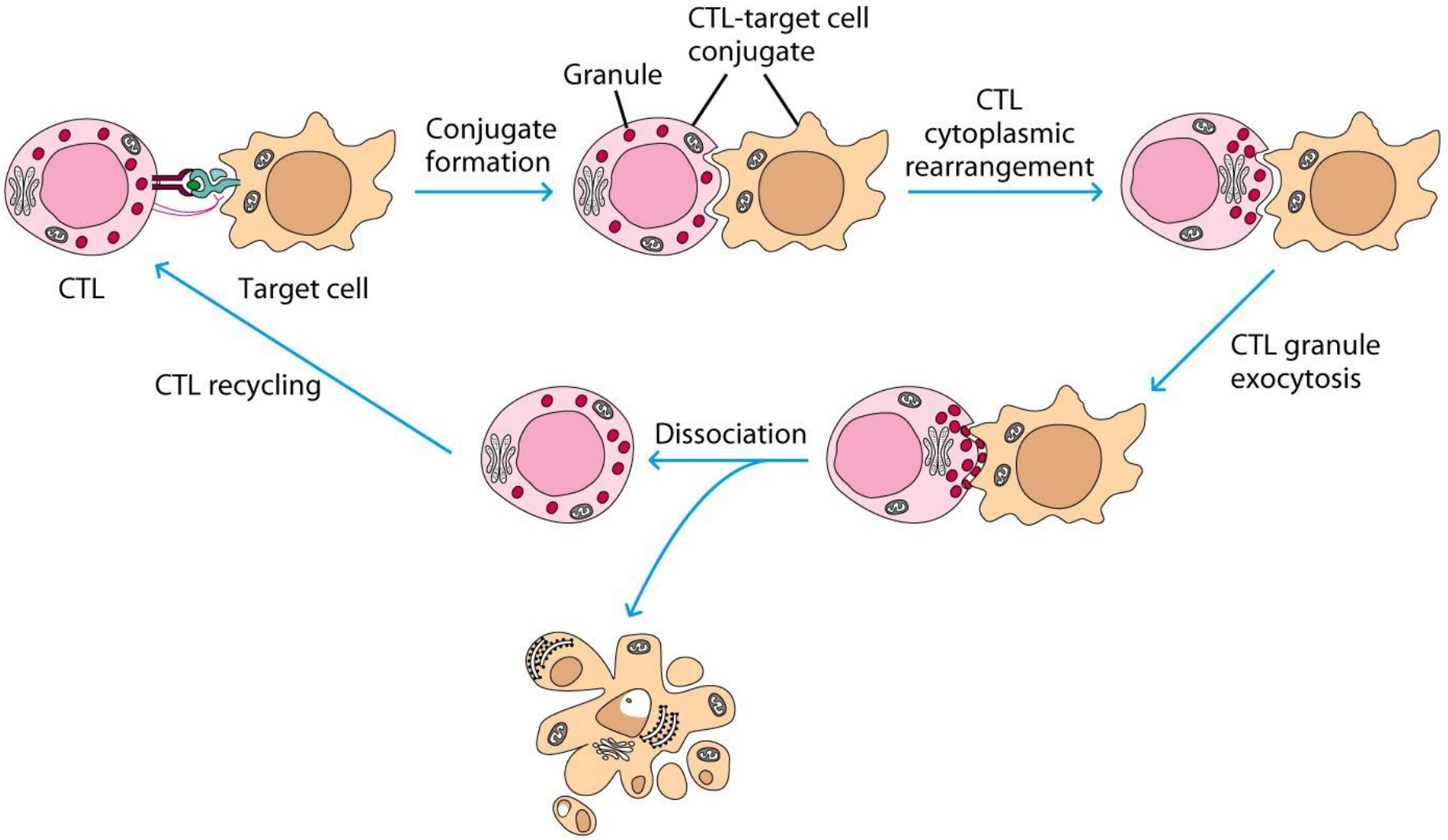
- 4 Phases In CTL Killing
  - Conjugate formation
    - Lymphocyte function-associated antigen 1 [LFA-1] of CTL binds ICAMs (Intercellular Adhesion Molecule 1 also known as CD54) (Target)
      - LFA-1 changes to high avidity if Ag Is Recognized
      - Activated LFA-1 persists for 5-10 mins
  - Membrane attack
    - Requires Ca^{2+} and energy
      - Granules release Perforins (65 kDa) and Granzymes (serine proteases) at the junctional space
      - Perforins polymerize forming cylindrical pores (5-20 nm), Ca^{2+} is needed
      - Granzymes enter target cell
      - Granzyme B can enter thru mannose-6-phosphate receptor in a vesicle
      - DNA fragmentation
  - CTL dissociation
  - Target cell destruction
    - Apoptotic death within a few hours


Nucleus
Perforin monomers
Granule
2
$Ca^{2}\uparrow$
1
3
CTL
6
5
4
Target cell
Completed pore
Polymerized perforin



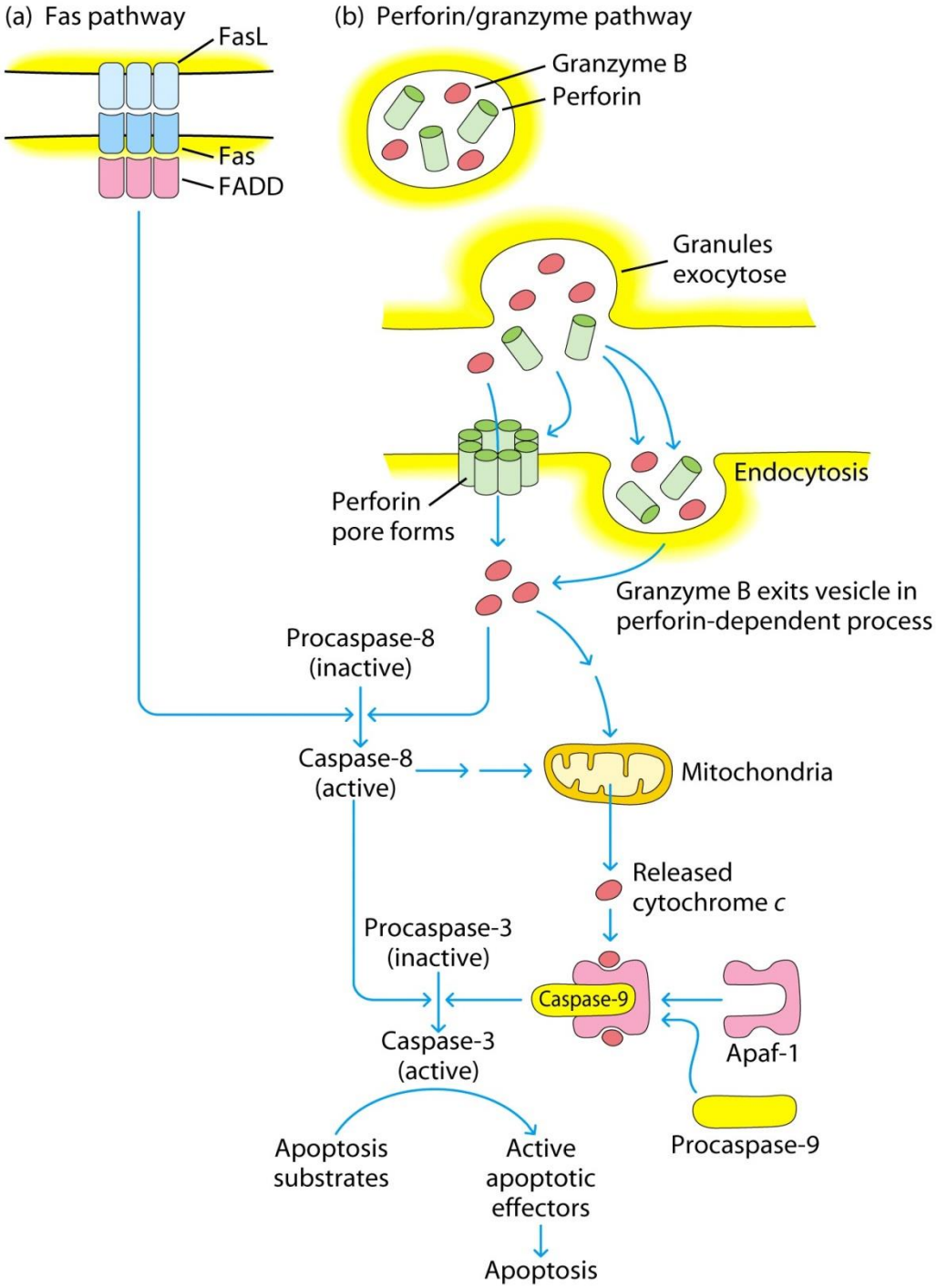


CTL Making Contact With Tumor Cell (small cell)


CTL-target cell conjugate
Granule
Conjugate formation
CTL cytoplasmic rearrangement
CTL
Target cell
CTL granule exocytosis
CTL recycling
Dissociation

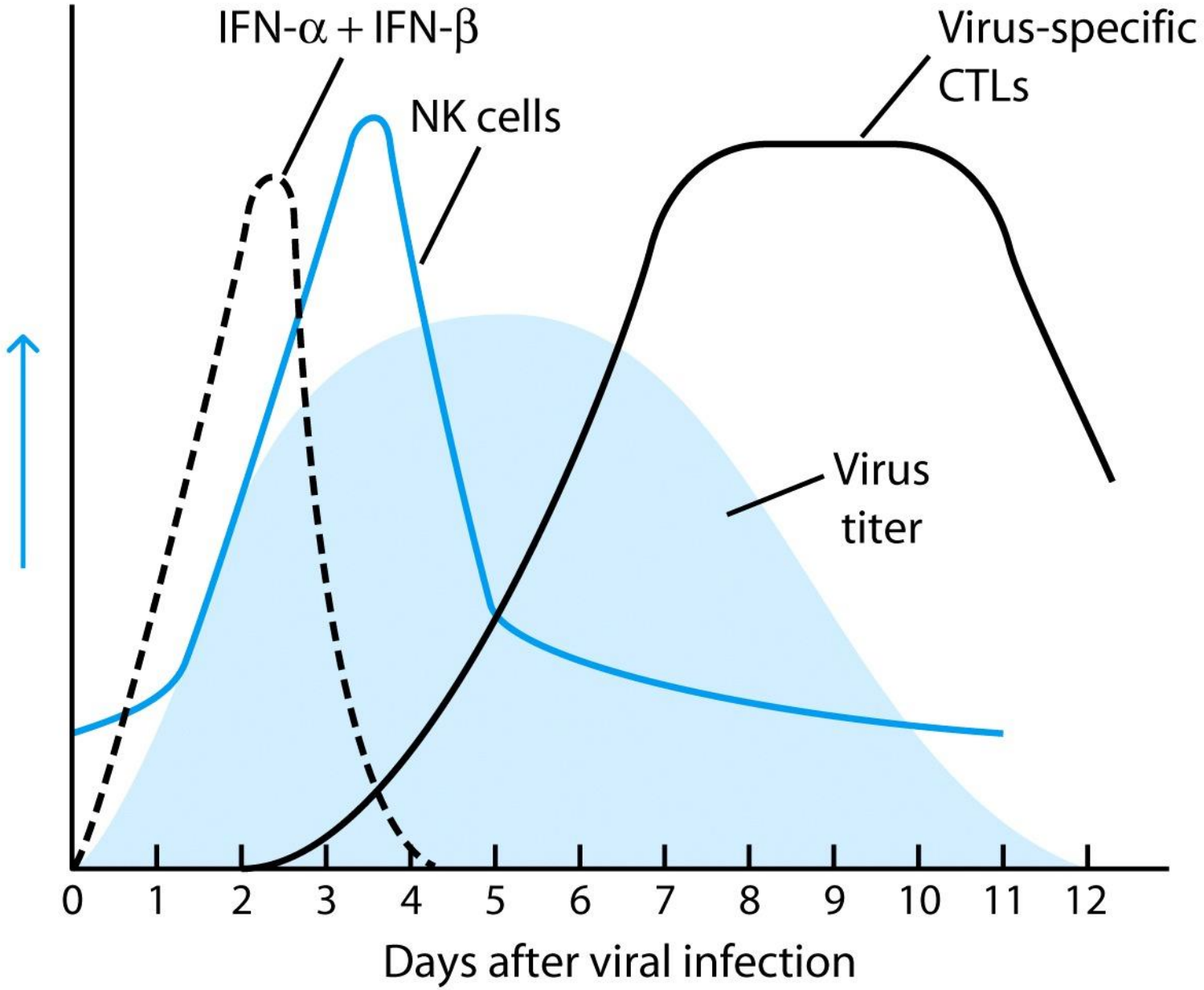
FasL Mediated Cytotoxicity

- Fas ligand (FasL), also known as CD178 or CD95L, is expressed on CTLs and functions by engaging the death receptor Fas (CD95), also known as apoptosis antigen 1 (APO-1 or APT) on target cells and triggering apoptosis.
- Some CTLs Lack Granzyme And Perforin
- They Kill Using FasL-Fas Interaction
  - FasL is found on CTLs
  - Fas is found on target cell
  - FasL-Fas interaction induces apoptosis
- 2 Mechanisms Are Responsible For CTL Induced Apoptosis
  - FasL-Fas (FADD Activation leading to pro-caspase 8 activation)
  - Perforin and granzyme
  - During apoptosis caspases (cysteine proteases that cleave aspartic acid) are activated
  - Family of more than 12 caspases exist
  - Activation of caspases results in orderly destruction of target cell


(a) Fas pathway
FasL
Fas
FADD
(b) Perforin/granzyme pathway
Granzyme B
Perforin
Granules exocytose
Endocytosis
Perforin pore forms
Granzyme B exits vesicle in perforin-dependent process
Procaspase-8 (inactive)
Caspase-8 (active)
Mitochondria
Released cytochrome c
Procaspase-3 (inactive)
Caspase-9
Apaf-1
Caspase-3 (active)
Procaspase-9
Apoptosis substrates
Active apoptotic effectors
Apoptosis

Natural Killer Cells

- NK Make Up 5-10% Of Circulating Lymphocytes
  - Major Producers of IFNγ
  - Thru IFNγ they influence innate immunity (MΦ)
  - They also influence adaptive, favor T_{H1}
  - Eliminate viruses and tumor cells
- Early Responders To Viral Infections
  - IFNα and IFNβ produced by pDCs Stimulates NK activity
  - IFNγ production induces MΦ To Make IL-12
  - IL-12 Results In More IFNγ Pushing Towards T_{H1}
  - T_{H1} Thru IL-2 Induces CTL activation

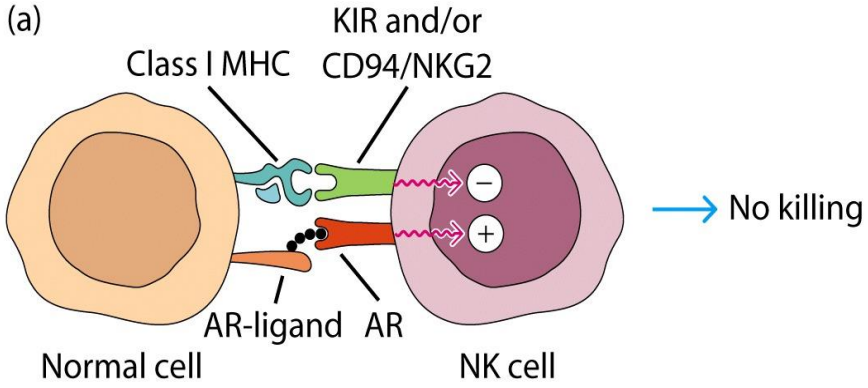

IFN-α + IFN-β
NK cells
Virus-specific CTLs
Virus titer
0 1 2 3 4 5 6 7 8 9 10 11 12
Days after viral infection

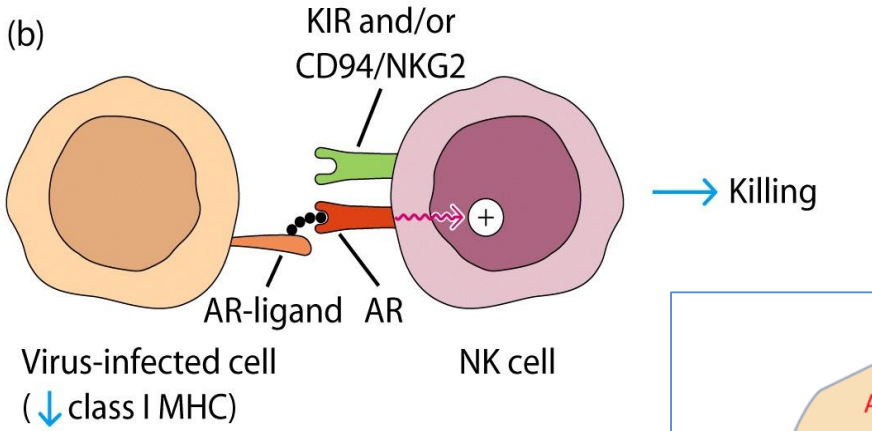
Natural Killer Cells

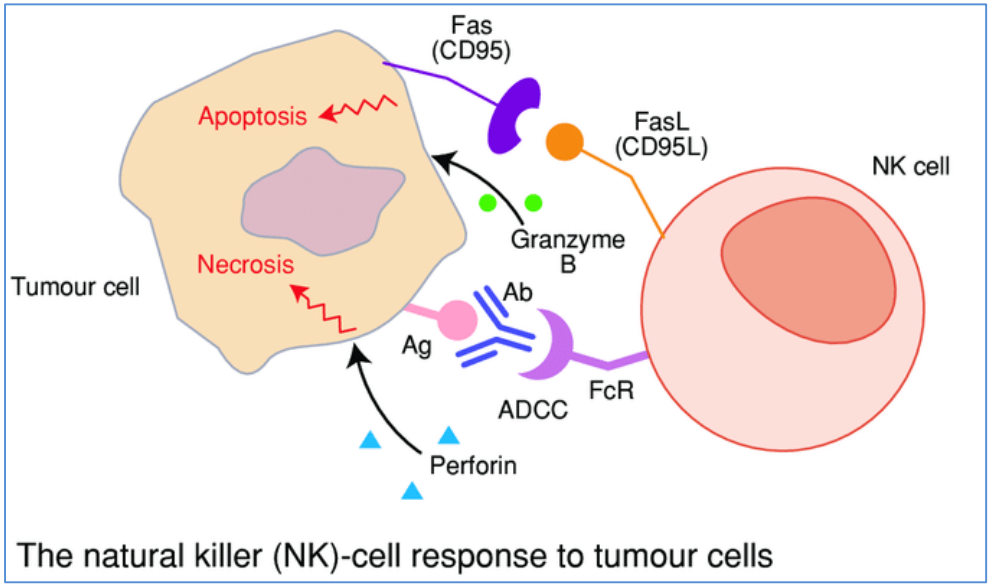
- NK eliminate target cells same way as CTLs
  - Thru perforin/granzyme and FasL/Fas
- However They Are Different From CTLs
  - No Ag Specific TCR
  - No CD3
  - No MHC restriction
  - No memory, same intensity regardless of repeated exposure
- How Do They Recognize The Target?

Target Recognition

- Balance Between Activating and Inhibiting Molecules Allows NK Cells To Differentiate Normal From Altered
- Still Not Clear What The Activating Receptors Are
  - C-type lectins Are Candidates
    - NKR-P1c [Natural killer cell receptor protein]
    - CD2 (Receptor for adhesion molecule LFA-3)
    - CD16 (FcγRIII, Involved In Antibody Mediated Recognition)
    - NKp30, NKp44 and NKp46
- Inhibitory Receptors
  - MHC Molecules
  - CD94/NKG2 Recognize HLA-E
  - If HLA-E is present –ve signal, no killing
  - No HLA-E (during viral infection), no –ve signal, killing
  - KIRs recognize specific MHC molecules, -ve signal, no killing



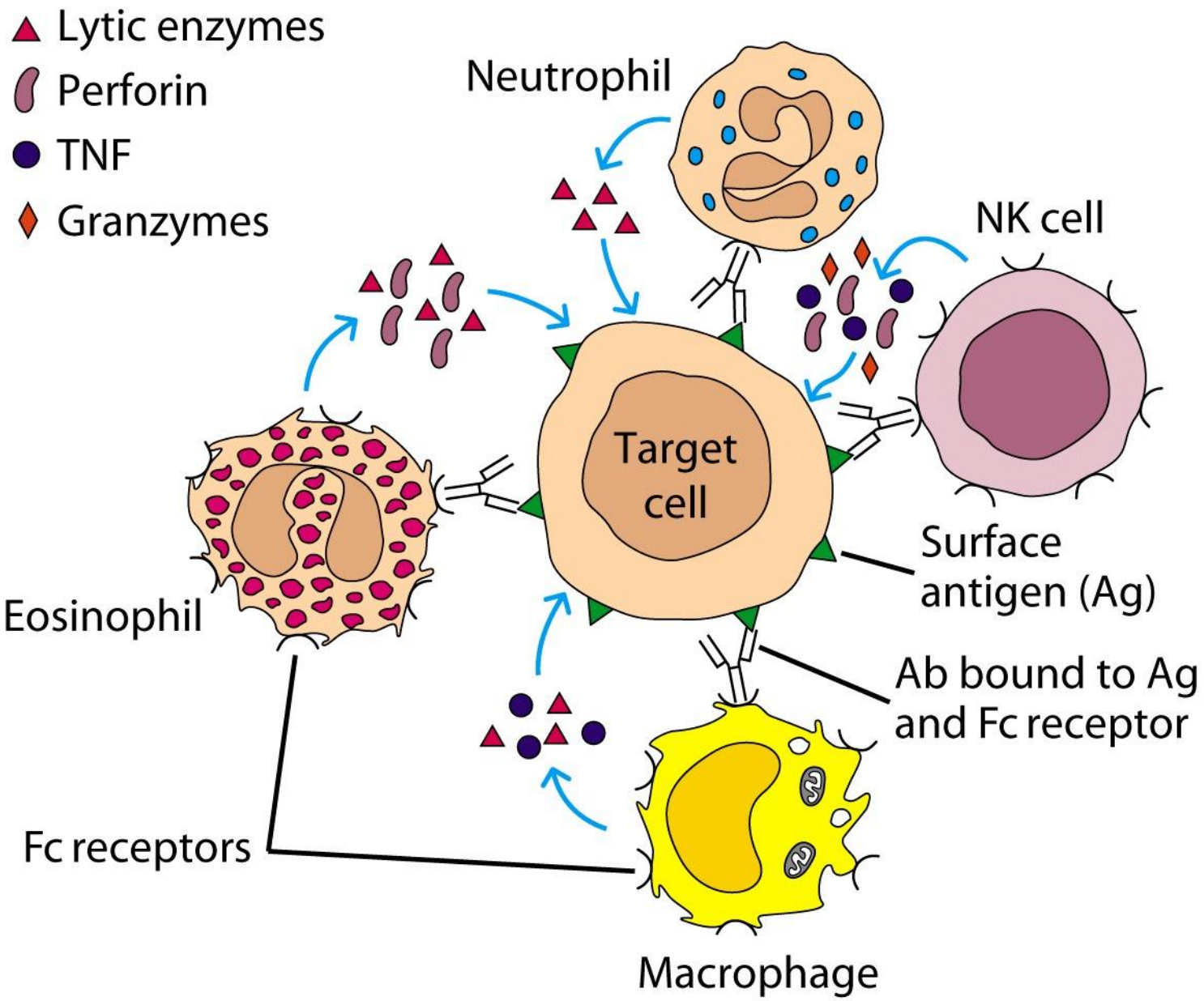




The natural killer (NK)-cell response to tumour cells

Antibody Dependent Cell Mediated Cytotoxicity (ADCC)

- Cells capable of cytotoxicity express Fc Receptors
- Antibody Binds Target Cell, Cytotoxic Cells Bind Fc Portion Of Ab
- Antibody provides the specificity
- Examples of cells capable of ADCC
  - MΦ, NK, Neutrophils, eosinophils
- Killing of target is accomplished
  - Thru perforin, granzyme (NK, Eosinophils)
  - TNF (MΦ, NK)
  - Lytic enzymes (MΦ, Neutrophils, Eosinophils, NK)


Lytic enzymes
Perforin
TNF
Granzymes
Neutrophil
NK cell
Target cell
Surface antigen (Ag)
Eosinophil
Ab bound to Ag and Fc receptor
Fc receptors
Macrophage

B-lymphocytes - ontogenesis, surface markers, function.

B-lymphocytes

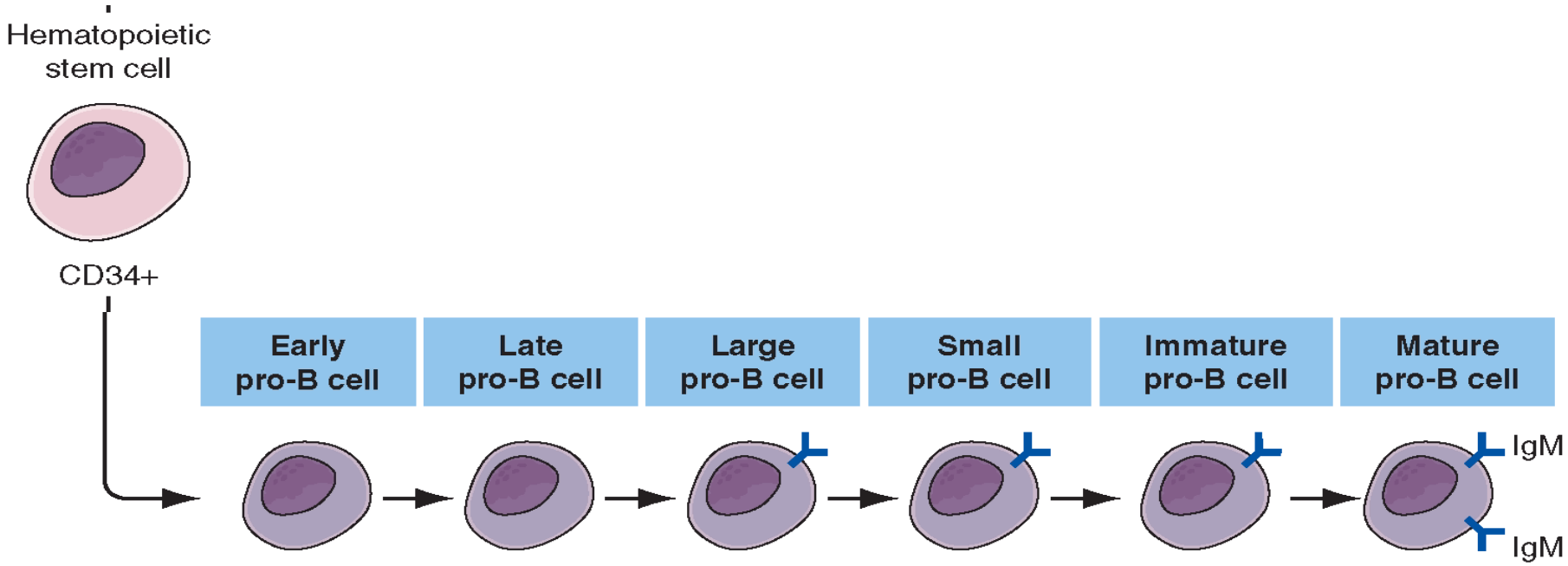
are an essential component of the innate immune system

- Maturation of B cells course in the **BM**
- B cells ordinate from stem cells and need to be in touch with the stromal cells in the bone marrow
- **Stromal cells** produce **SCF** (stem cell factor) needed for development at early period, **IL-7** needed at later period of maturation
- Ig gene rearrangements and the appearance of surface markers identify the stage of B-cell development

Development of B lymphocytes

- Lymphoid progenitor gives rise to precursors of B cells = pro- B cells
- During maturation from the pro-B cells into the pre-B cells – Ig genes of the heavy chain recombine; pre-B cells express **pre-BCR**
- During maturation from the pre-B cells into the

  B cells – Ig genes of the light chain recombine
- Immature B cells express membrane IgM
- Mature B cells express membrane IgM and IgD = **BCR** and are able to respond to antigen in peripheral lymphoid tissues



	Early pro-B cell	Late pro-B cell	Large pro-B cell	Small pro-B cell	Immature pro-B cell	Mature pro-B cell
High-chain genes	DJ rearranged	VDJ rearranged	VDJ rearranged	VDJ rearranged	VDJ rearranged	VDJ rearranged
Low-chain genes	Germ line	Germ line	Germ line	VDJ rearranged	VDJ rearranged	VDJ rearranged
Surface Ig	Absent	Absent	μ H-chain at surface as part of pre-β receptor	μ H-chain in cytoplasm and at surface	IgM expressed on cell surface	IgD and IgM made from alternatively spliced H-chain transcripts
Surface marker proteins	CD34 CD10 CD19 CD38	CD10 CD19 CD20 CD38 CD40	CD19 CD20 CD38 CD40	CD19 CD20 CD38 CD40	CD19 CD20 CD40	CD19 CD20 CD21 CD40

Negative selection

- If an immature B cell **binds** an antigen in the bone marrow with **high affinity**- further maturation is stopped and B cell dies by **apoptosis**
- **Negative selection** eliminates potentially dangerous cells that can **recognize** and **react** against **self antigens**
- B cells that survive this selection process leave the bone marrow through efferent blood vessels

B-lymphocytes – surface markers

- **CD 10** - immature B cells, malignant cells
- **CD 35 -** receptor for the C3b of the complement
- **CD 19 -** a characteristic marker of B cells
- **CD 20 -** a typical surface antigen of Ig-positive B lymphocytes
- **IgM, IgD** - antigen receptors = BCR
- **MHC class II** - antigen-presenting molecules

B-lymphocytes – functions

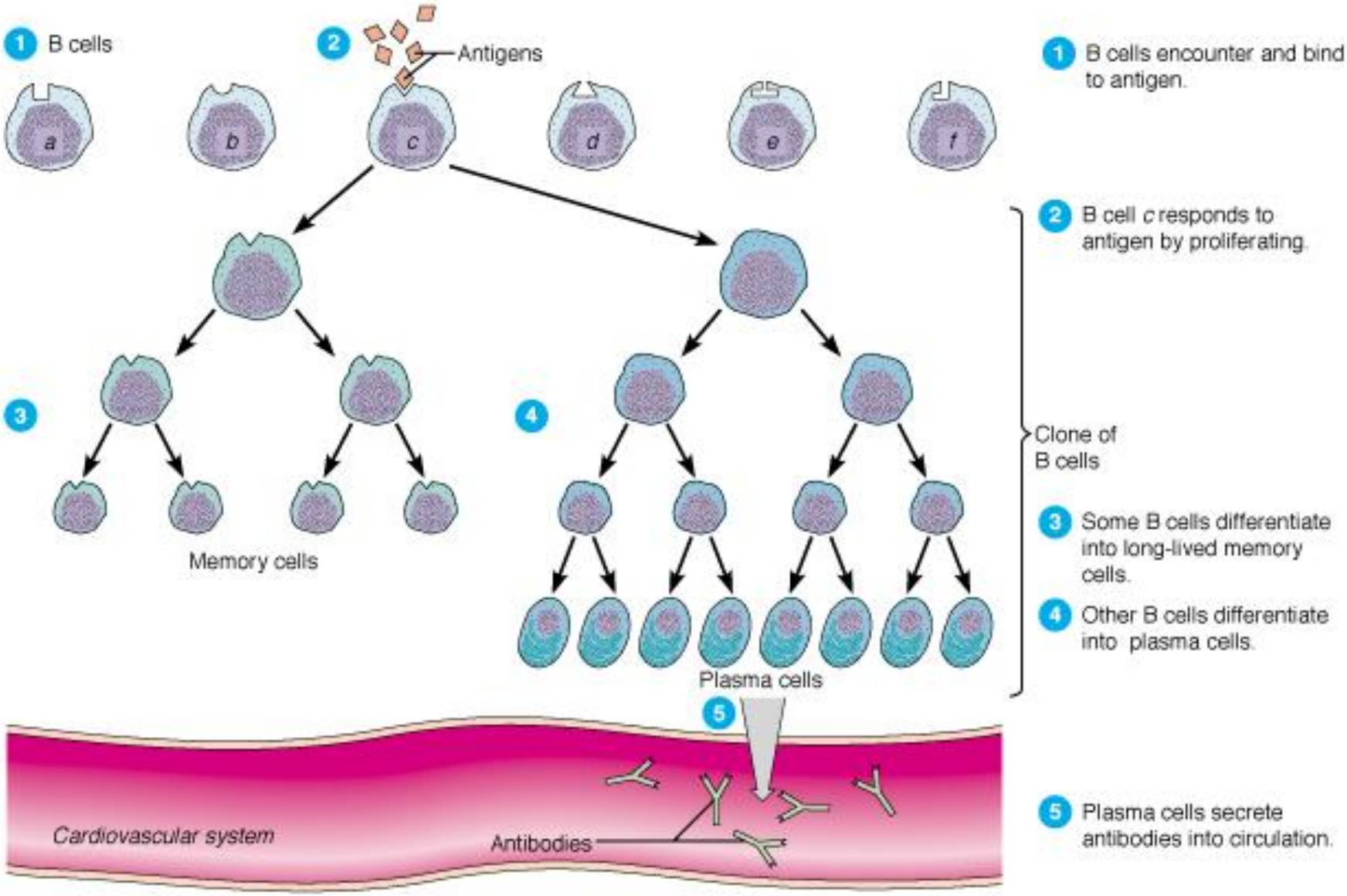
- After stimulation B lymfocytes convert into the **plasma cells** and **produce antibodies** against soluble antigens

- Other functions are :

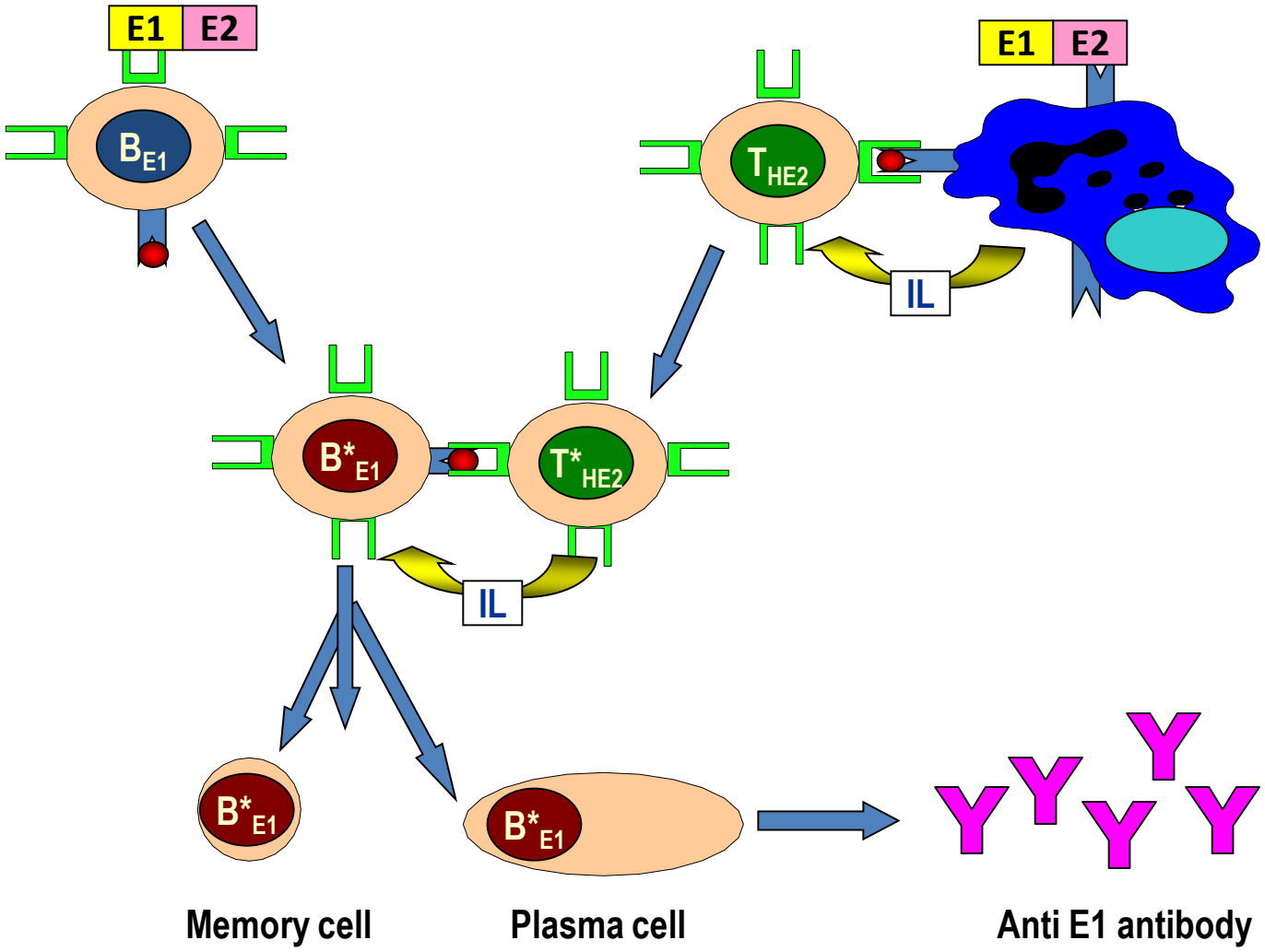
  antigen presentation

  cooperation with complement

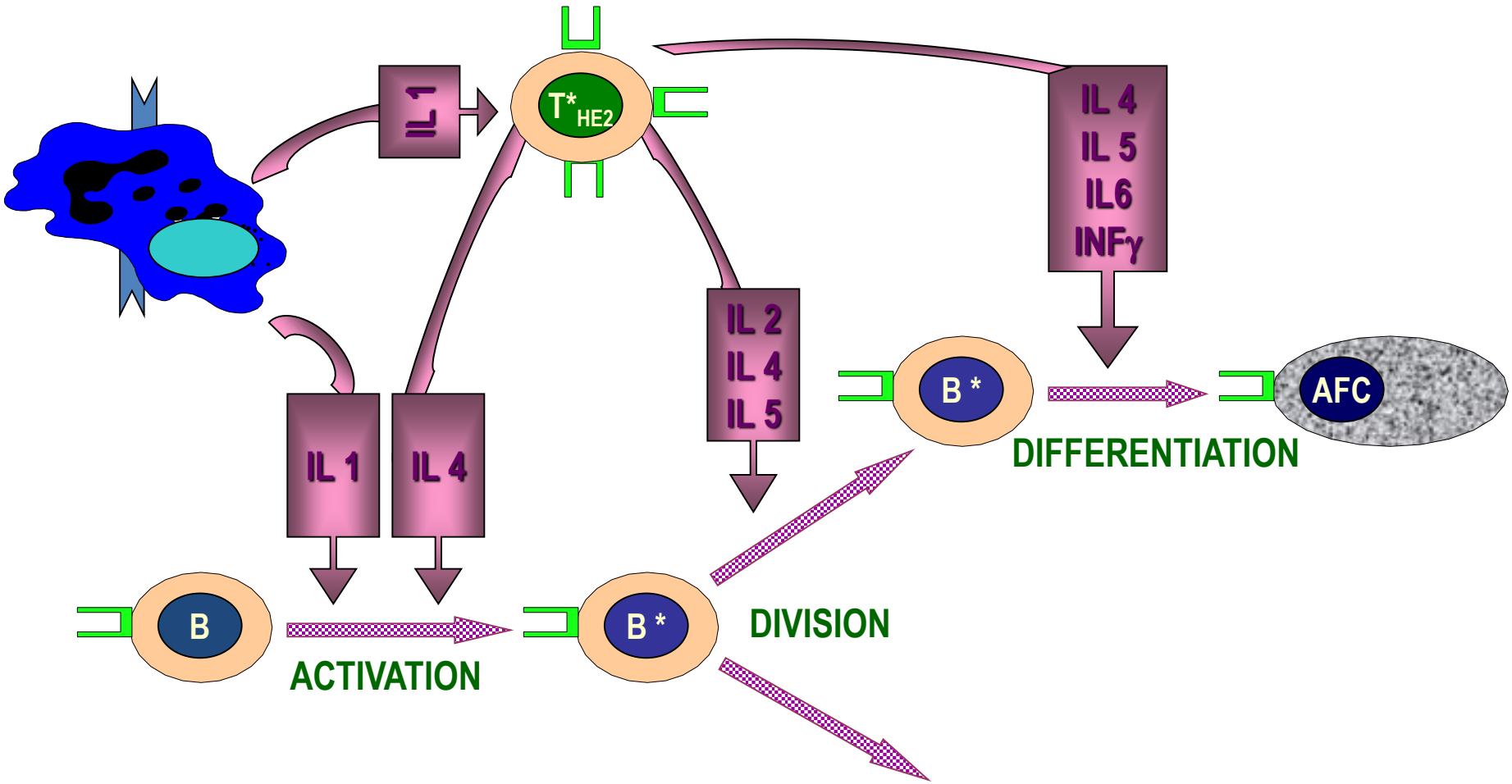
Humoral Immunity



Cellular interaction for Ab formation



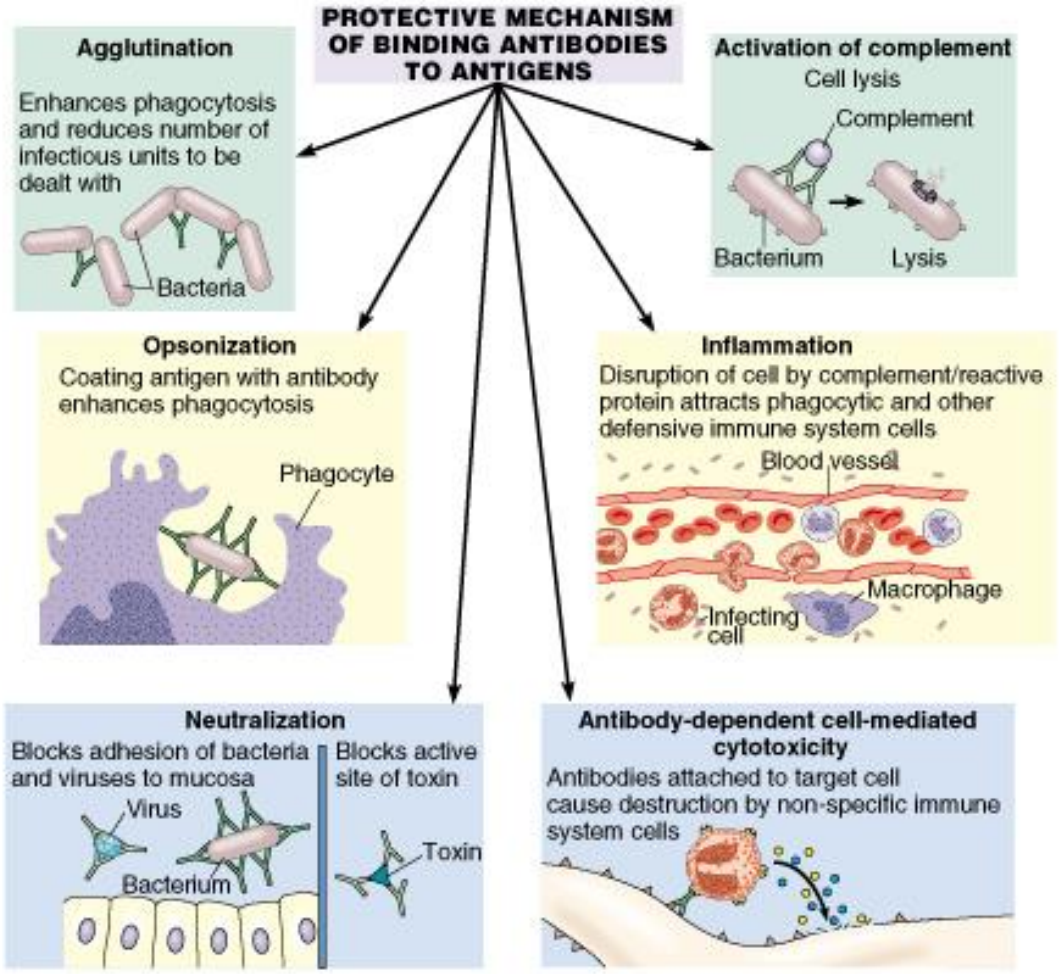
Interleukin (IL) mediated activation



Humoral Immunity

Mecanisms of Ab Protect

1. Agglutination
2. Opsonization
3. Neutralization
4. Activate Complement
5. Inflammation
6. Ab Dependent Cell-Mediated cytotoxicity



Parameters	B Cells	T Cells
Cell surface markers	B20, complement receptors, Ig	CD 2, 3, 4/8, T-cell receptor
Subsets	B1, B2, B-regulatory	TH, TC, Treg
Ag recognized	Lipids, polysaccharides, proteins	Peptides (processed)
Receptor	B cell receptor	T cell receptor
Properties of the epitope	Assemble, conformational	Linear peptide with MHC
Primary function	Produce antibody	Produce cytokines, cytotoxic, helper or suppressive activity
Hypersensitivity	Type I, II & III	Type IV