What you need to know about innate immunity

Jennifer Ko MD, PhD
Cleveland Clinic
Immunology/Pathology and Laboratory Medicine

Innate Immunity

- First line of defense, immediate defense
  - Day to day protection
  - Only when innate defense bypassed, evaded or overwhelmed is adaptive immunity required
- Non-specific
- Recognize pathogens in a generic way
- Does not confer long lasting or protective immunity to host
- Evolutionarily older, found in primitive organisms

Receptor characteristic	Innate immunity	Adaptive immunity
Specificity inherited in the genome	Yes	No
Expressed by all cells of a particular type (eg, macrophages)	Yes	No
Triggers immediate response	Yes	No
Recognizes broad classes of pathogen	Yes	No
Interacts with a range of molecular structures of a given type	Yes	No
Encoded in multiple gene segments	No	Yes
Requires gene rearrangement	No	Yes
Clonal distribution	No	Yes
Able to discriminate between even closely related molecular structures	No	Yes

Figure 2-10 Immunobiology, 6/e. (© Garland Science 2005)

Innate Immunity and Inflammation

- 1) Respond rapidly to tissue damage
  - physical and chemical barrier
  - recruitment of immune cells to site of injury
- 2) Limit spread of infection
  - identification and removal of foreign substances
  - activation of the complement cascade
  - activation of coagulation cascade
- 3) Initiate adaptive immune response
  - antigen presentation and cytokine production
- 4) Initiate tissue repair

Innate Immunity – Physical/Chemical Barriers

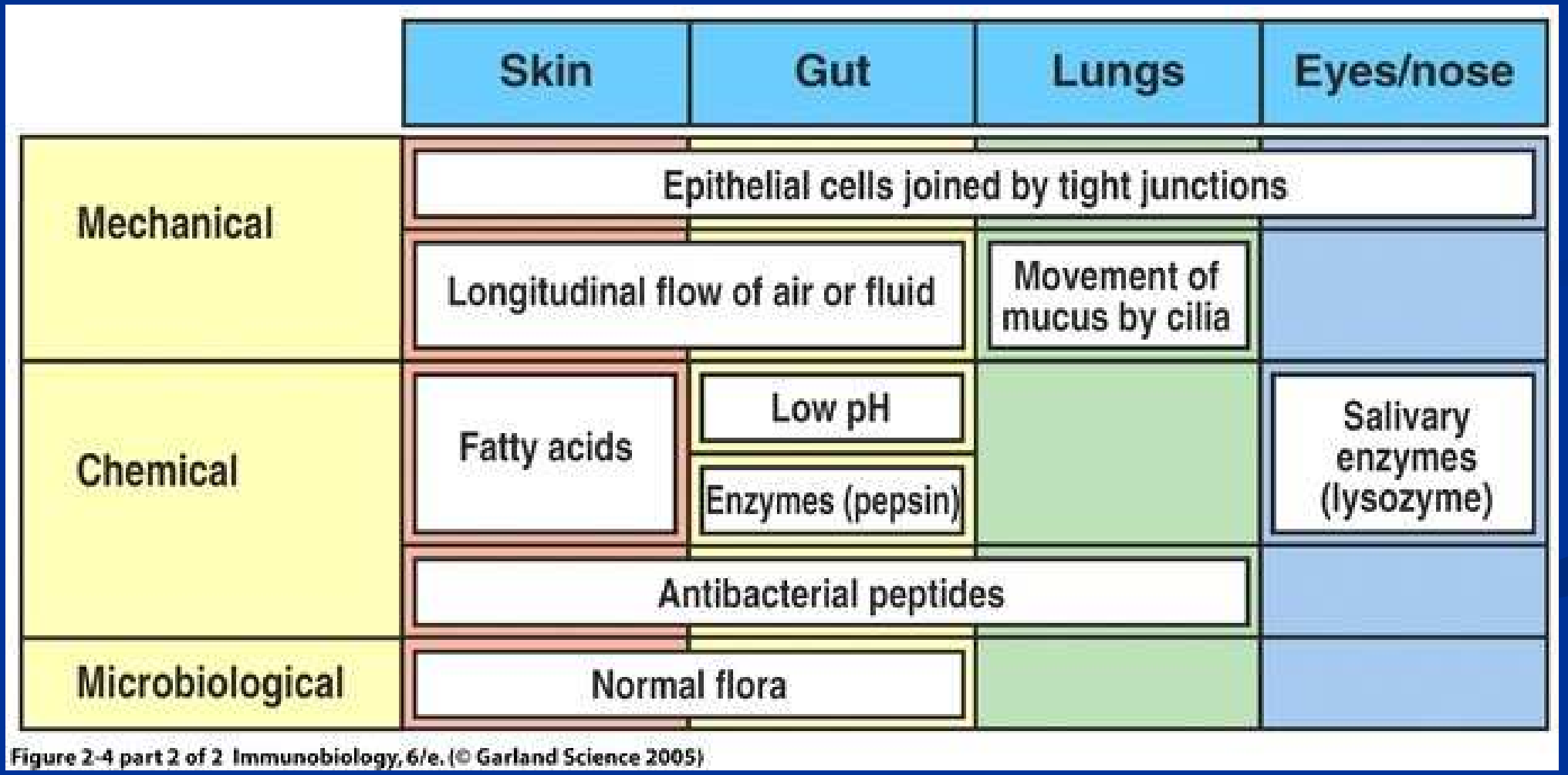


Figure 2-4 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

Breach of physical barrier →
-“resting” innate immune cells become activated to kill microbes, secrete cytokines to recruit and activate additional leukocytes, and to promote systemic killing and removal of microbes.

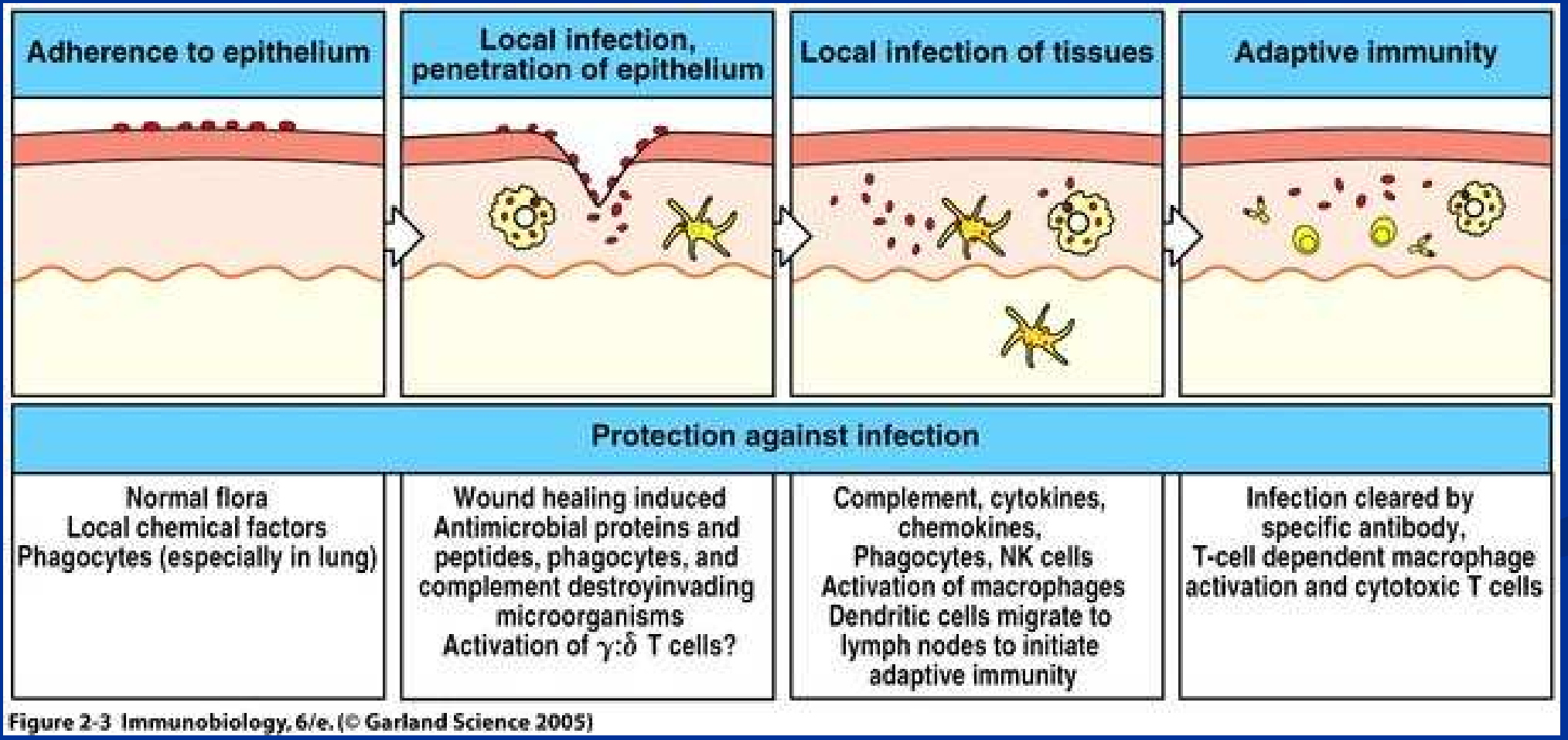
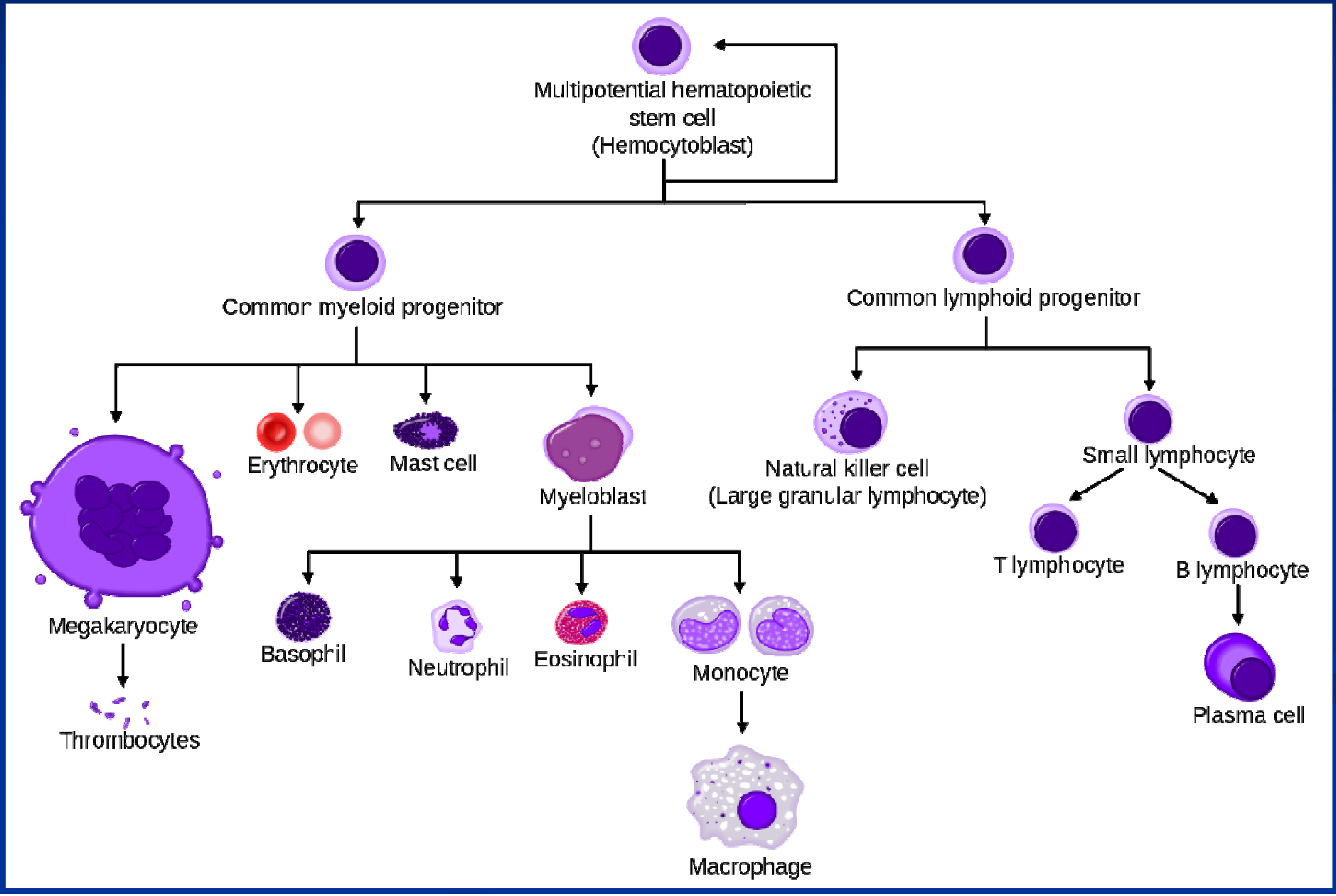
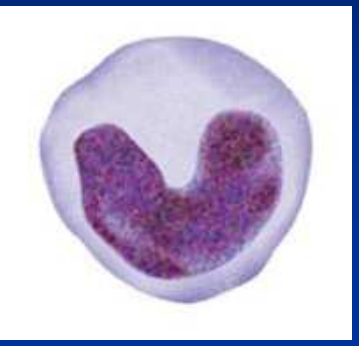


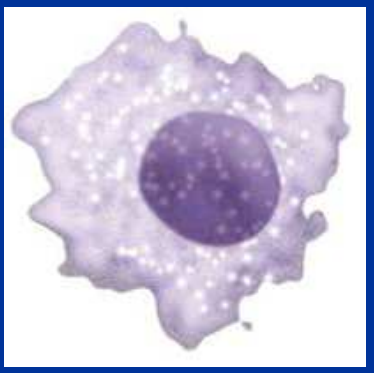
Figure 2-3 Immunobiology, 6/e. (© Garland Science 2005)

Innate Immunity- 1st responders



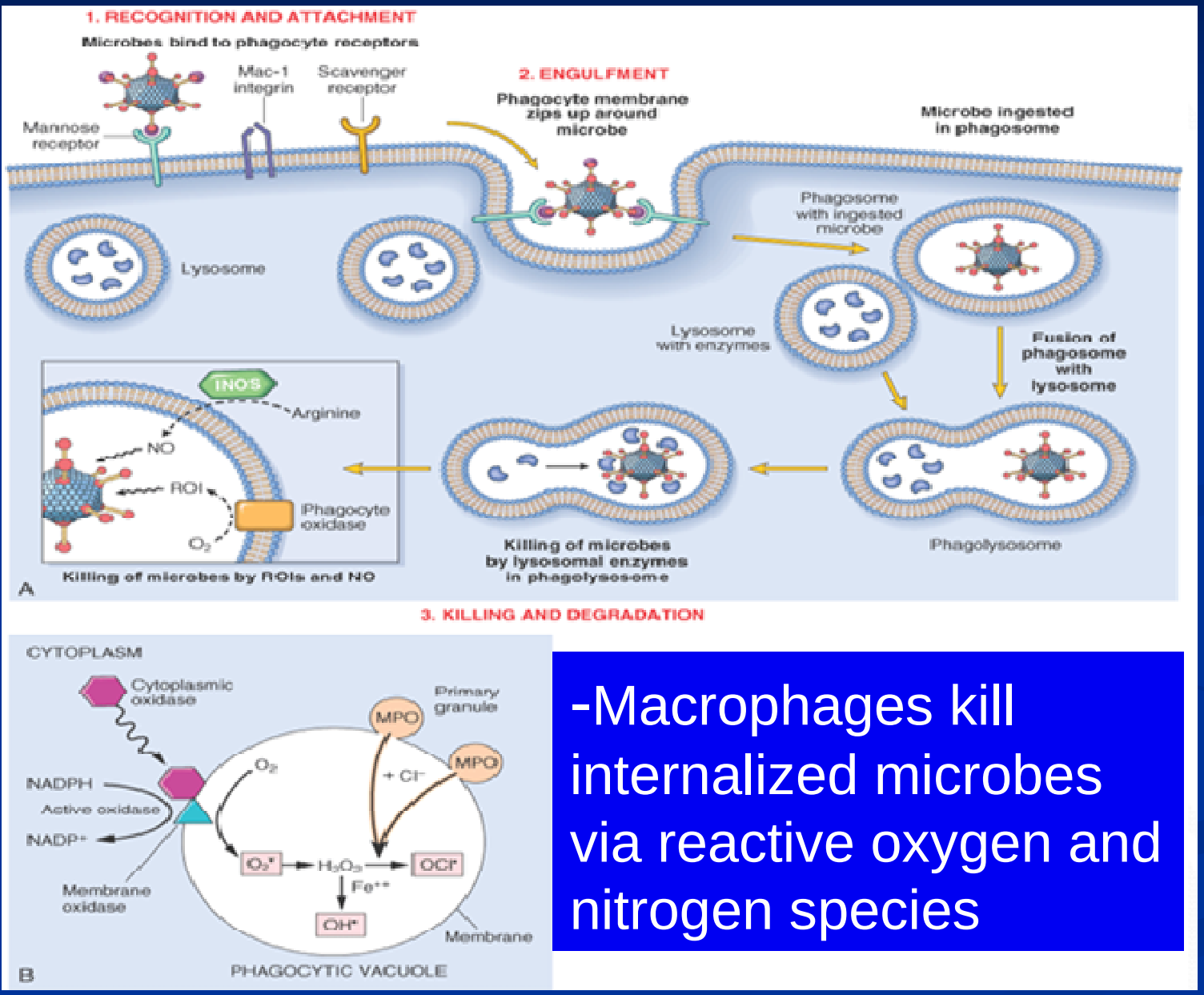
Innate Immunity- Monocytes/Macrophages





- Monocyte-derived macrophages “large eaters” or histiocytes, are present in all tissues
- “Sentinels” of immune system - survey for “foreign” invaders
- Foreign microbes are recognized via various cell surface and intracellular receptors
- Receptor ligation and cytokines causes macrophage activation
- Activated macrophages
  - Digest and present antigens from microbes
  - Produce chemokines, cytokines, other molecules to recruit other immune cells

Macrophage Phagocytosis

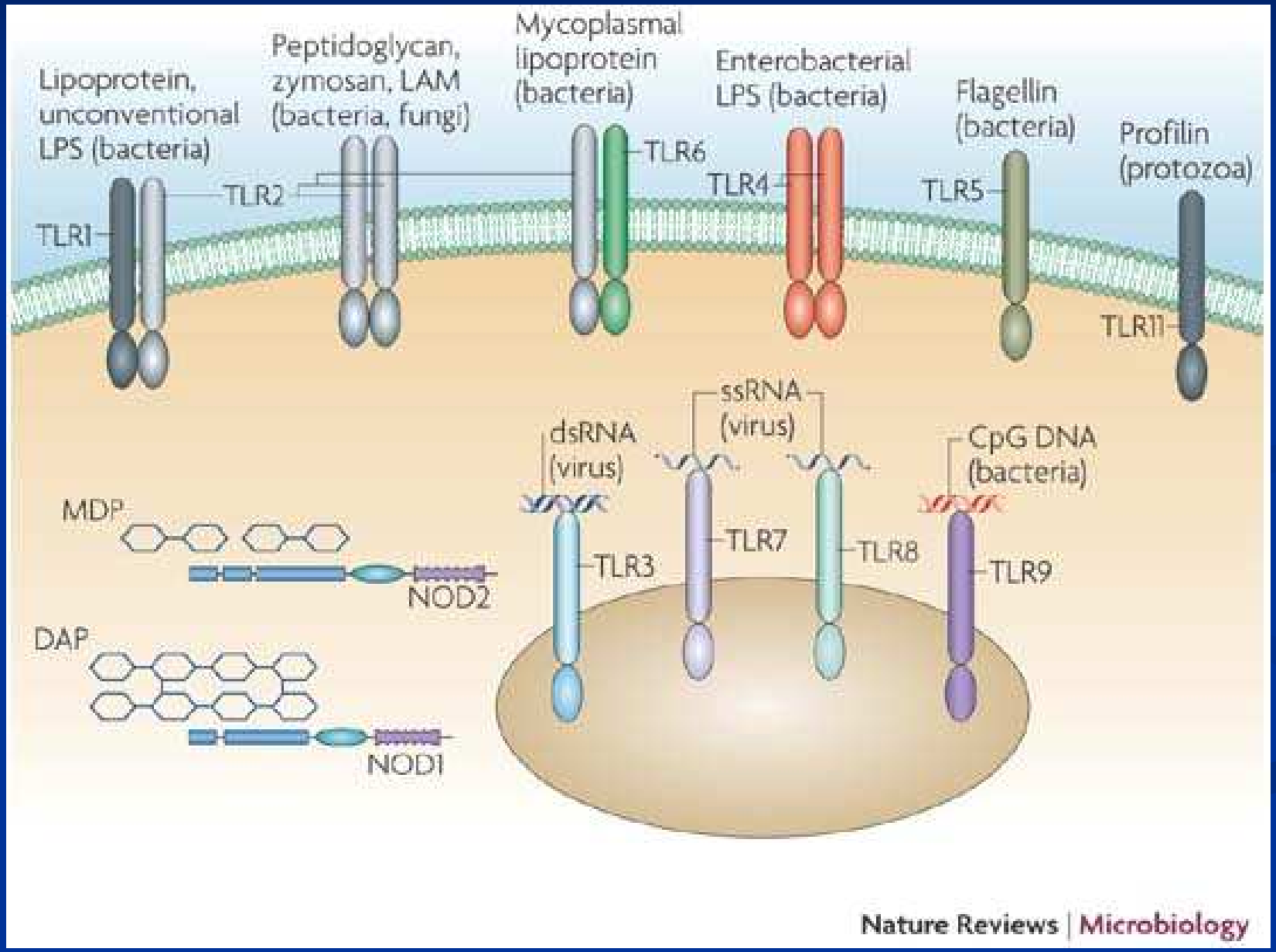


-Macrophages kill internalized microbes via reactive oxygen and nitrogen species

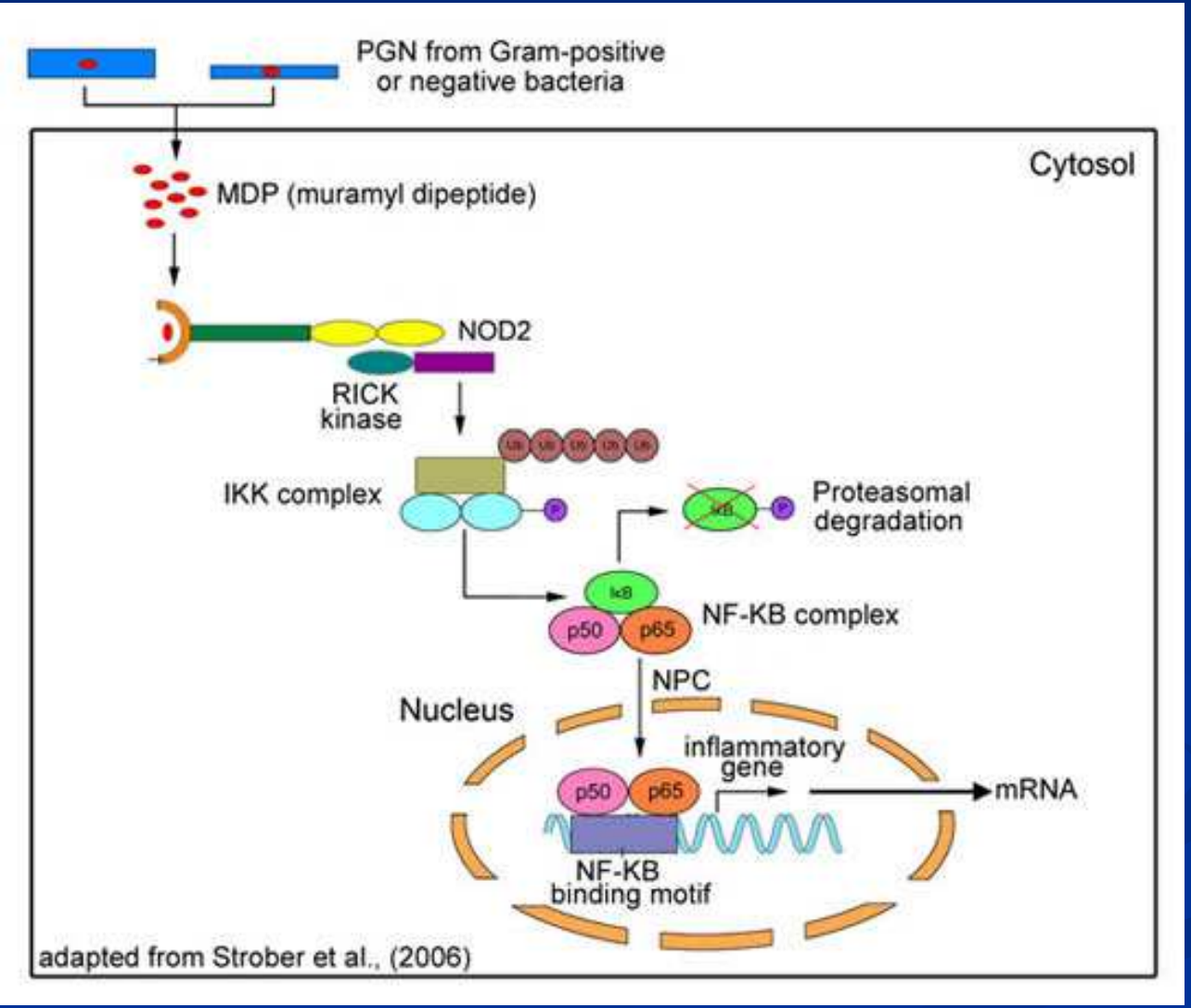
<u>Microbe killing=</u>

- **Phagosome + lysosome = phagolysosome**
- **Nitric oxide (nitric oxide synthase, iNOS2)**
- **Superoxide anion (NADPH oxidase, respiratory burst)**
- **Hydrogen peroxide (superoxide dismutase)**

Phagocyte Toll receptors are stimulated by Pathogen Associated Molecular Patterns (PAMPs)



NOD-like receptor (NLR) proteins are intracellular pattern recognition receptors (PRRs)



Induced Innate Responses mediated by cytokines secreted by stimulated sentinel cells

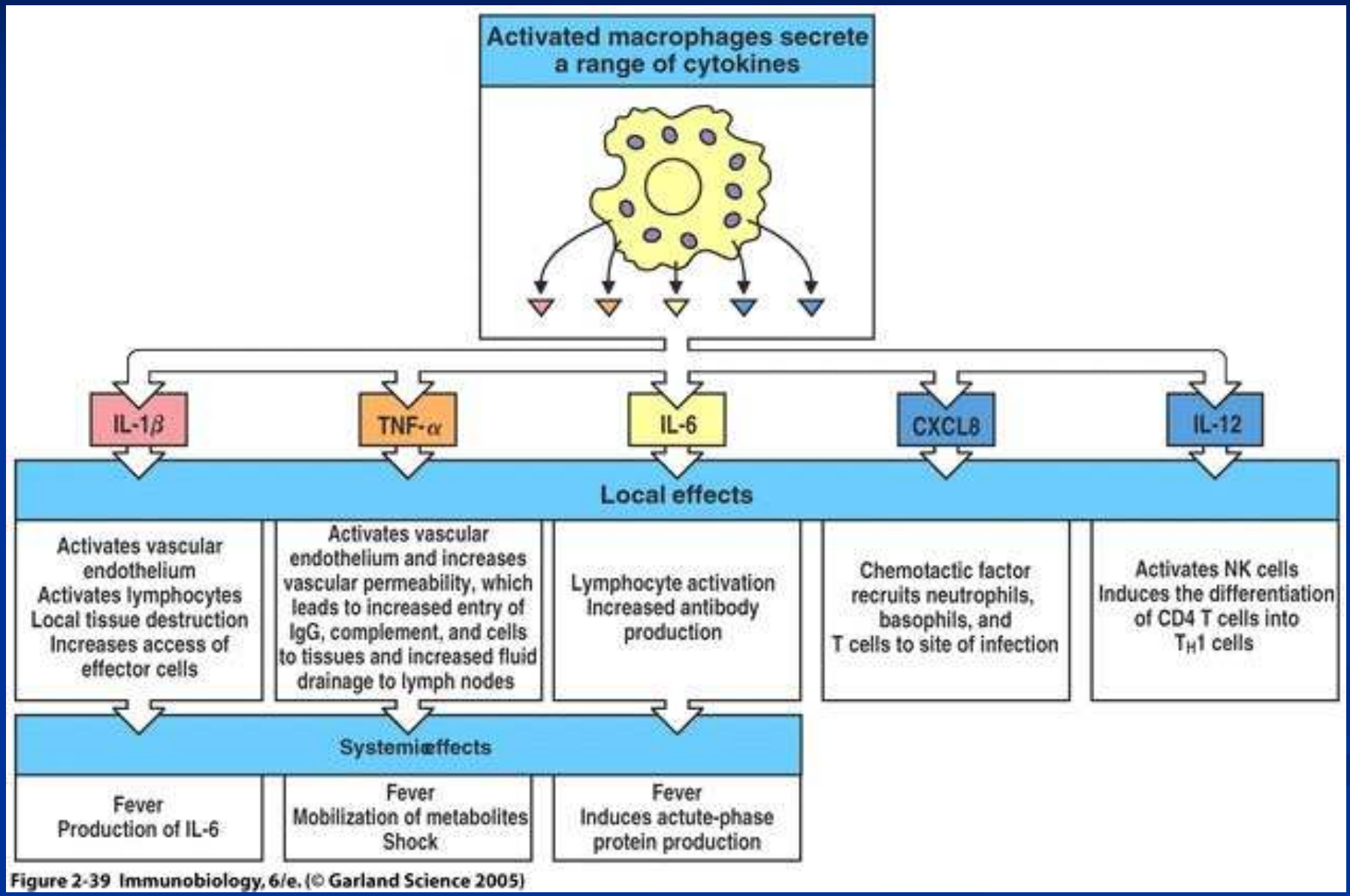


Figure 2-39 Immunobiology, 6/e. (© Garland Science 2005)

Chemokines secreted by stimulated sentinel cells recruit additional immune cells

Class	Chemokine	Produced by	Receptors	Cells attracted	Major effects
CXC	CXCL8 (IL-8)	Monocytes Macrophages Fibroblasts Keratinocytes Endothelial cells	CXCR1 CXCR2	Neutrophils Naive T cells	Mobilizes, activates and degranulates neutrophils Angiogenesis
	CXCL7 (PBP, β-TG NAP-2)	Platelets	CXCR2	Neutrophils	Activates neutrophils Clot resorption Angiogenesis
	CXCL1 (GROα) CXCL2 (GROβ) CXCL3 (GROγ)	Monocytes Fibroblasts Endothelium	CXCR2	Neutrophils Naive T cells Fibroblasts	Activates neutrophils Fibroplasia Angiogenesis
	CXCL10 (IP-10)	Keratinocytes Monocytes T cells Fibroblasts Endothelium	CXCR3	Resting T cells NK cells Monocytes	Immunostimulant Antiangiogenic Promotes T_H1 immunity
	CXCL12 (SDF-1)	Stromal cells	CXCR4	Naive T cells Progenitor ($CD34^+$) B cells	B-cell development Lymphocyte homing Competes with HIV-1
	CXCL13 (BLC)	Stromal cells	CXCR5	B cells	Lymphocyte homing

Figure 2-41 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

Class	Chemokine	Produced by	Receptors	Cells attracted	Major effects
CC	CCL3 (MIP-1α)	Monocytes T cells Mast cells Fibroblasts	CCR1, 3, 5	Monocytes NK and T cells Basophils Dendritic cells	Competes with HIV-1 Antiviral defense Promotes T_H1 immunity
	CCL4 (MIP-1β)	Monocytes Macrophages Neutrophils Endothelium	CCR1, 3, 5	Monocytes NK and T cells Dendritic cells	Competes with HIV-1
	CCL2 (MCP-1)	Monocytes Macrophages Fibroblasts Keratinocytes	CCR2B	Monocytes NK and T cells Basophils Dendritic cells	Activates macrophages Basophil histamine release Promotes T_H2 immunity
	CCL5 (RANTES)	T cells Endothelium Platelets	CCR1, 3, 5	Monocytes NK and T cells Basophils Eosinophils Dendritic cells	Degranulates basophils Activates T cells Chronic inflammation
	CCL11 (Eotaxin)	Endothelium Monocytes Epithelium T cells	CCR3	Eosinophils Monocytes T cells	Role in allergy
	CCL18 (DC-CK)	Dendritic cells	?	Naive T cells	Role in activating naive T cells
C	XCL1 (Lymphotactin)	CD8>CD4 T cells	CXCR1	Thymocytes Dendritic cells NK cells	Lymphocyte trafficking and development
CXXXC (CX_3C)	CX3CL1 (Fractalkine)	Monocytes Endothelium Microglial cells	CX_3CR1	Monocytes T cells	Leukocyte–endothelial adhesion Brain inflammation

Figure 2-41 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

Innate Immunity-Macrophage

Systemic Inflammatory Response

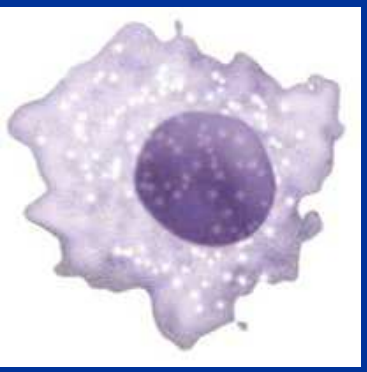
•MBL

Complement Cascade

•TNFα

•IL-6

•IL-1



•Pattern Recognition Receptors

•Toll-like receptors

•NLRs

•Macrophage mannose receptor

•Scavenger receptors

Phagocytosis (clearance of microbes)

•Macrophage chemokines/cytokines

•Co-stimulatory molecules

•CD80/86

•Neutrophils

•TNFα

•IL-8

Recruitment of other cells

Cytokine mediated vascular dilation and vascular permeability facilitate neutrophil extravasation into infected tissues

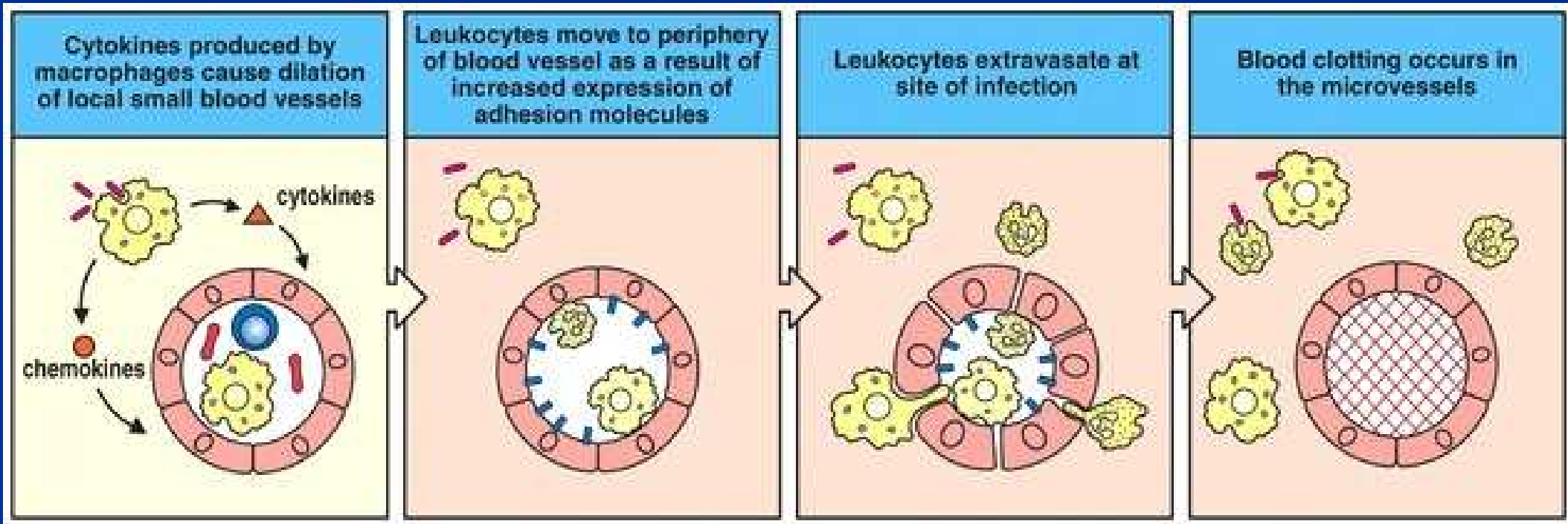
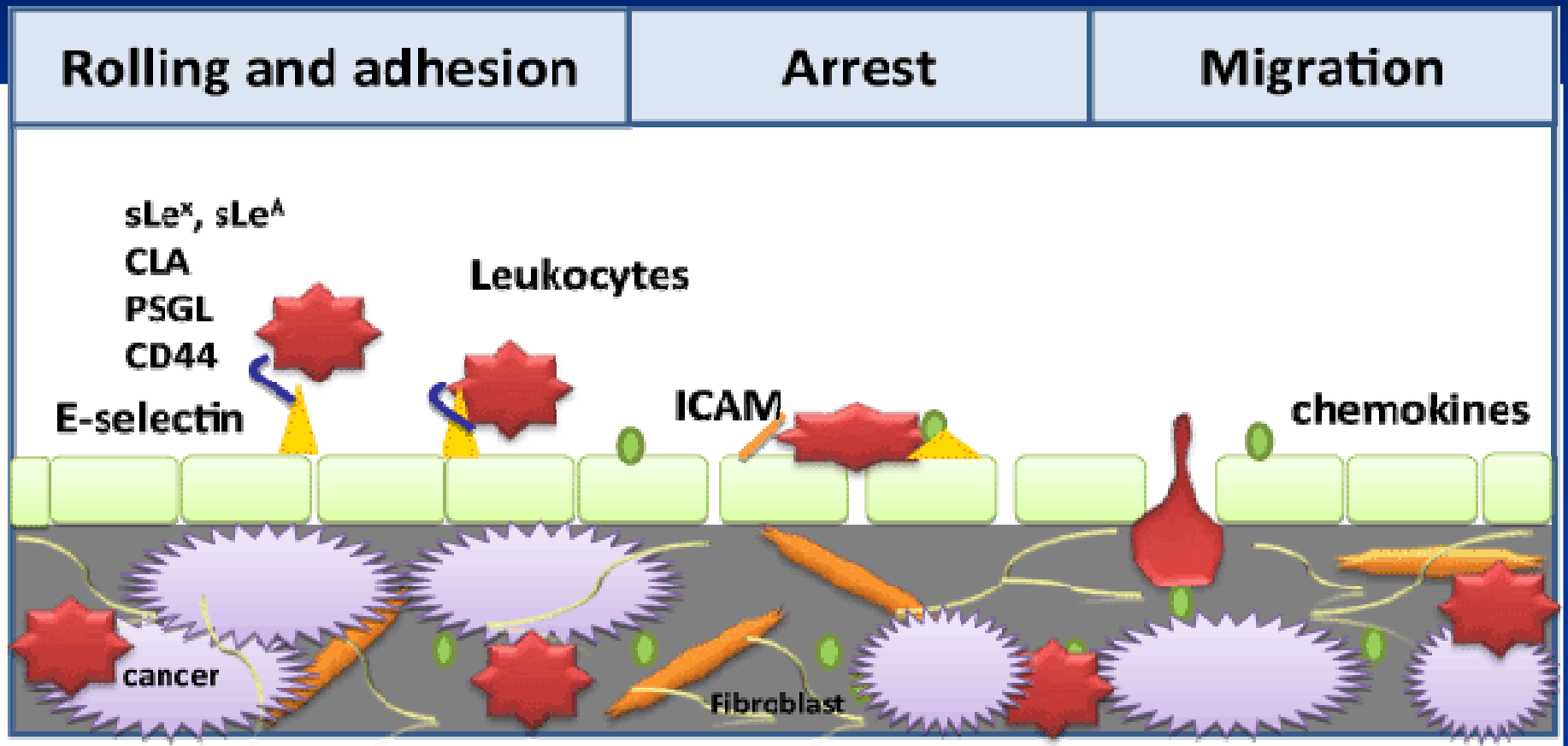


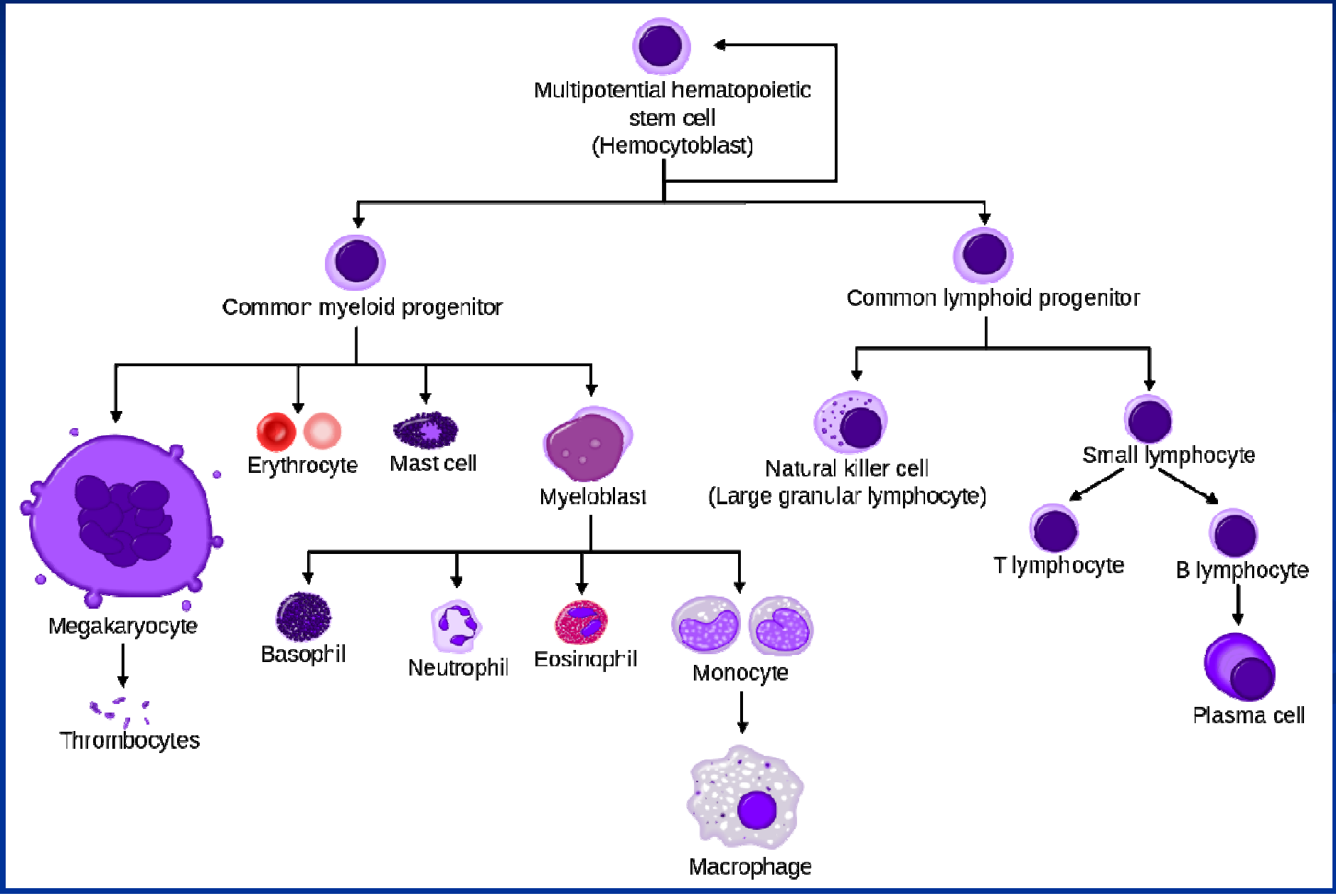
Figure 2-8 Immunobiology, 6/e. (© Garland Science 2005)

-Macrophage activation causes degradation of membrane phospholipids and rapid production of prostaglandins, leukotrienes and platelet-activating factor which act with cytokines directly on smooth muscle and endothelial cells

Cytokine induced adhesion→ cytokines and chemokines mediate neutrophil weak and firm adhesion to vascular endothelium



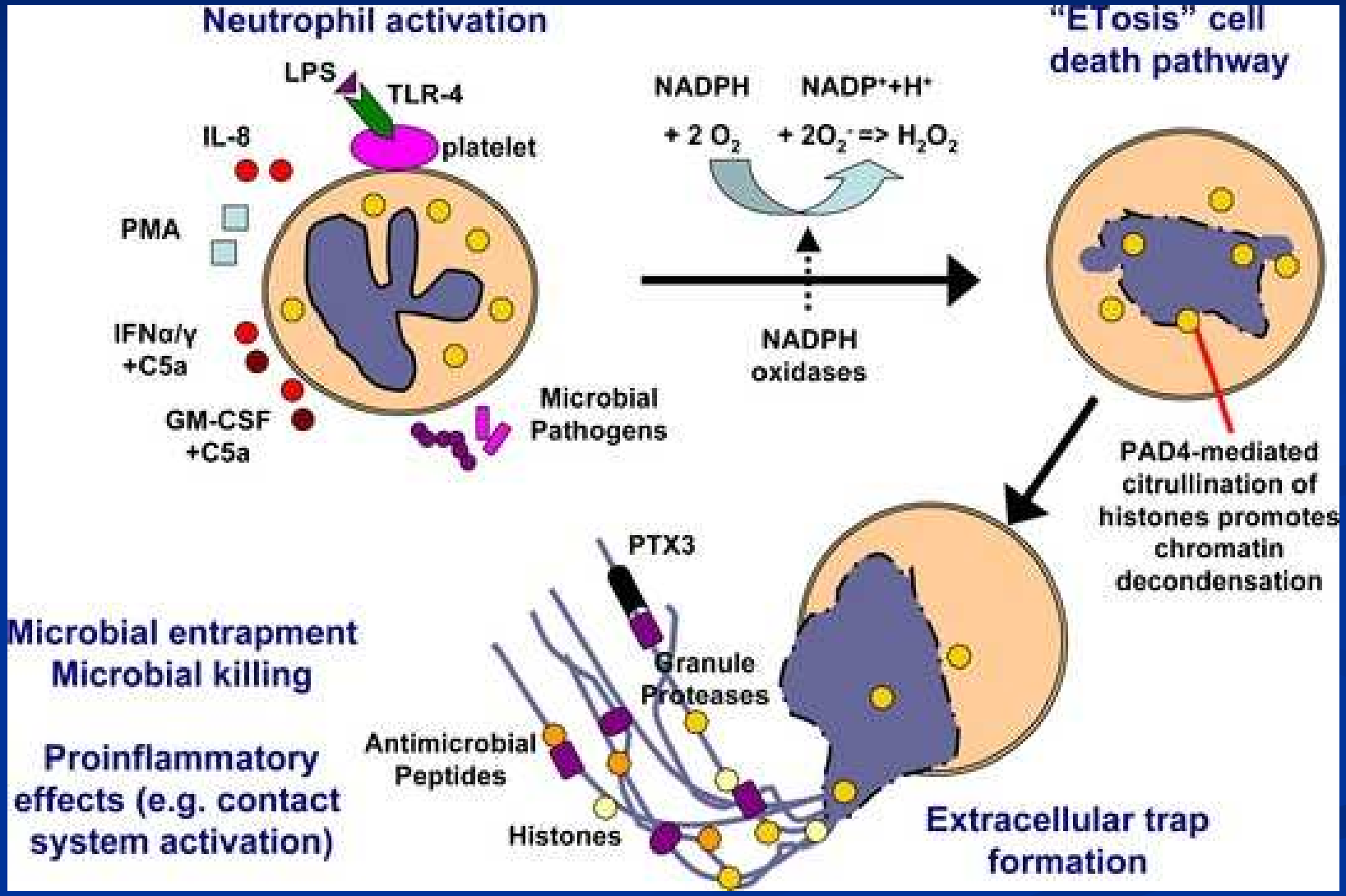
Innate Immunity- 1st responders



Neutrophils

- Essential to innate immunity, hallmark of acute inflammation
- Most prevalent WBC in blood with 50-100 billion produced per day
  - 55% bone marrow weight dedicated
- Migrate in response to IL-8, C5a, leukotrienes, fMLP via chemotaxis
- Circulate 5.4 days, live in tissue 1-2 days
  - Limit propagation of certain pathogens
  - Limit host damage due to inflammation
  - Phagocytosed by macrophages after pathogen digestion

Neutrophil NET formation



Innate Immunity-Macrophage

Systemic Inflammatory Response

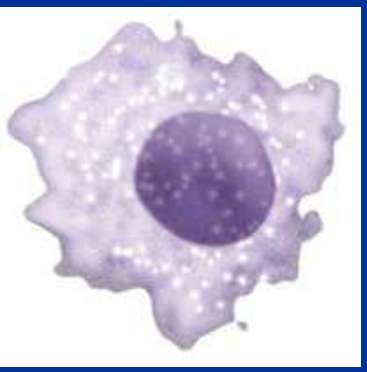
•MBL

Complement Cascade

•TNFα

•IL-6

•IL-1



•Pattern Recognition Receptors

•Toll-like receptors

•NLRs

•Macrophage mannose receptor

•Scavenger receptors

Phagocytosis (clearance of microbes)

•Macrophage chemokines/cytokines

•Co-stimulatory molecules

•CD80/86

•TNFα

•IL-8

Recruitment of other cells

•Neutrophils

NETosis

Cytokines from PAMP-stimulated sentinel cells stimulate the production of acute phase proteins, which opsonize a large spectrum of pathogens bearing common pathogen associated molecular patterns

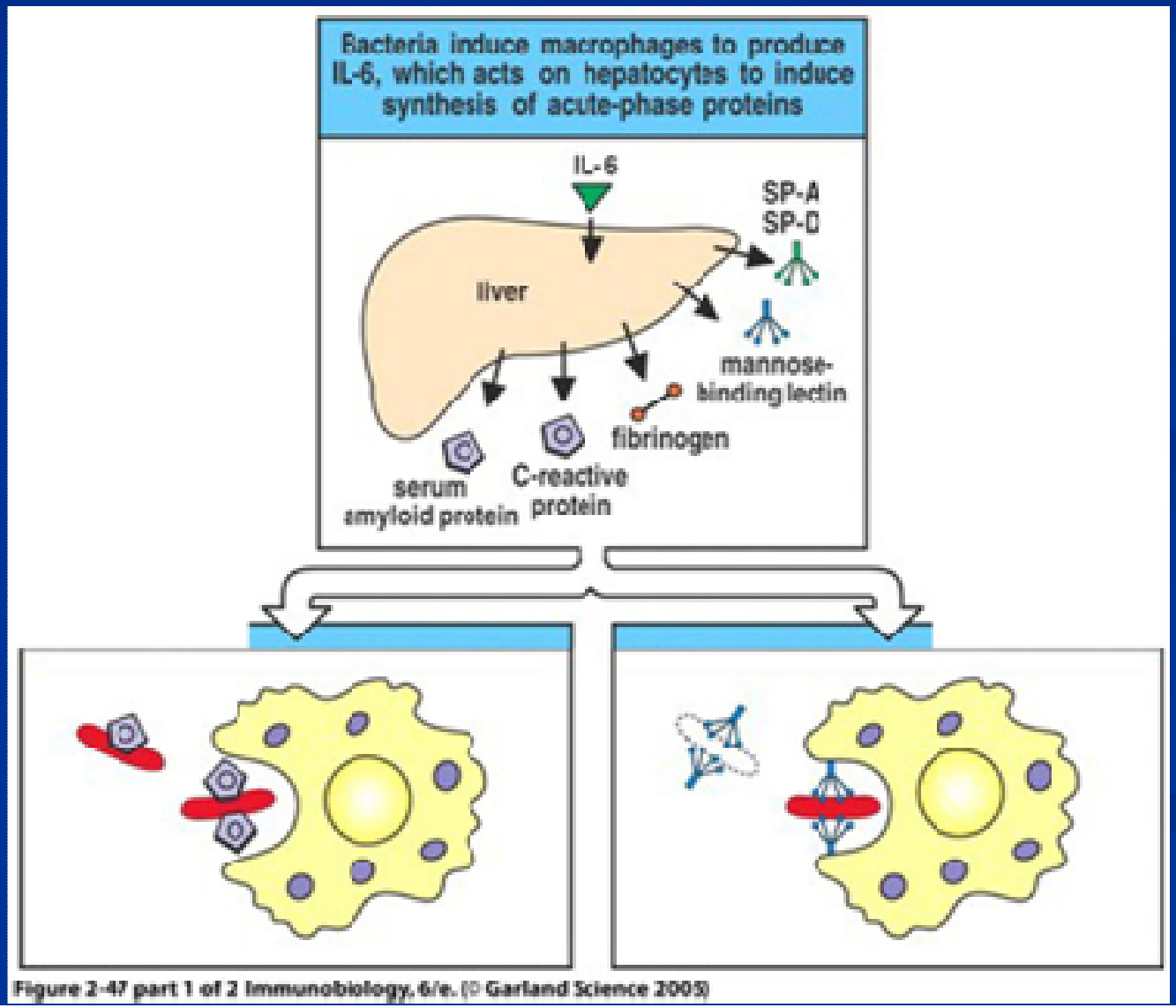


Figure 2-47 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

There are two innate mechanisms by which complement can be activated: the lectin pathway and the alternative pathway

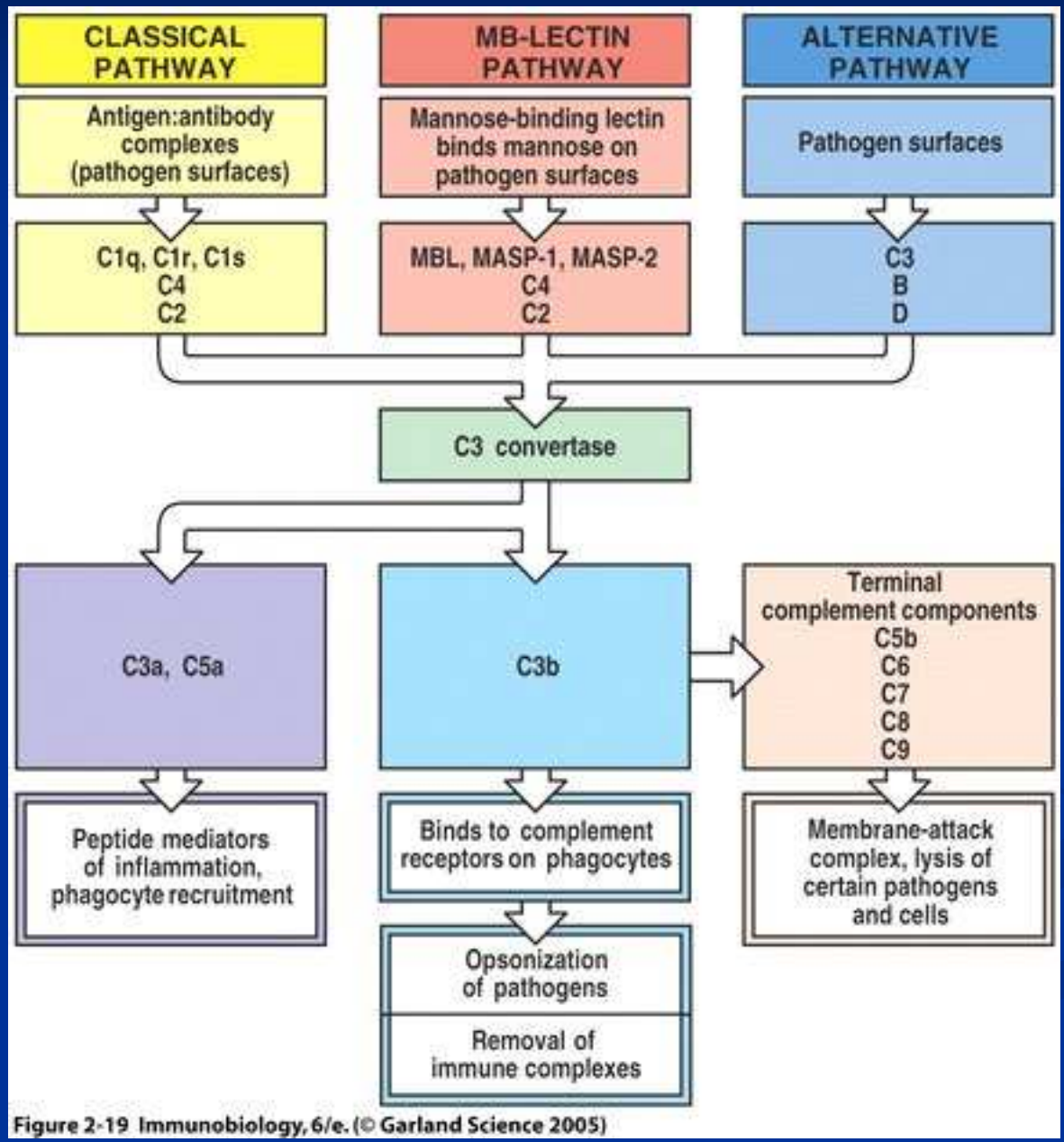


Figure 2-19 Immunobiology, 6/e. (© Garland Science 2005)

Complement feeds forward to activate and increase macrophage and neutrophil phagocytosis

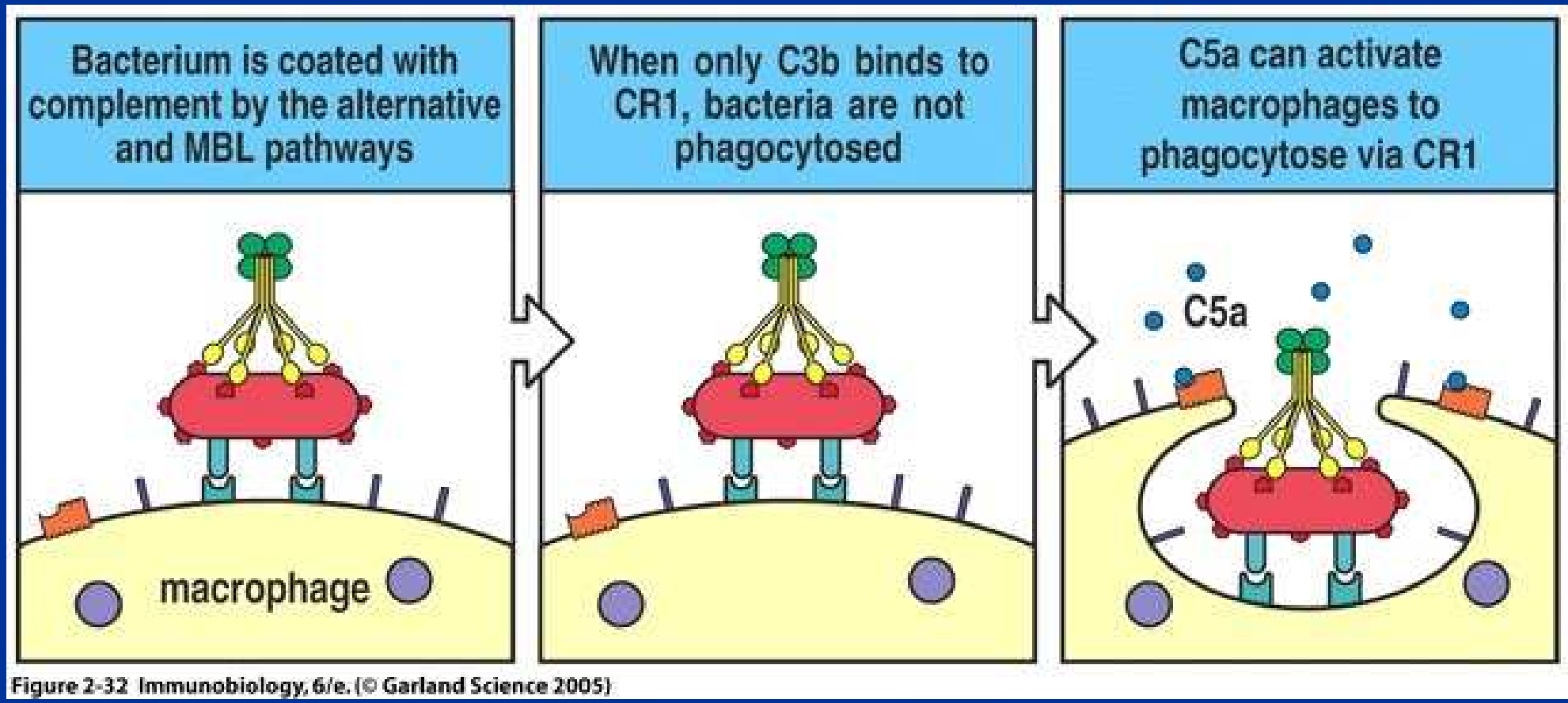


Figure 2-32 Immunobiology, 6/e. (© Garland Science 2005)

Complement can form the membrane attack complex, which leads to the lysis of target pathogens

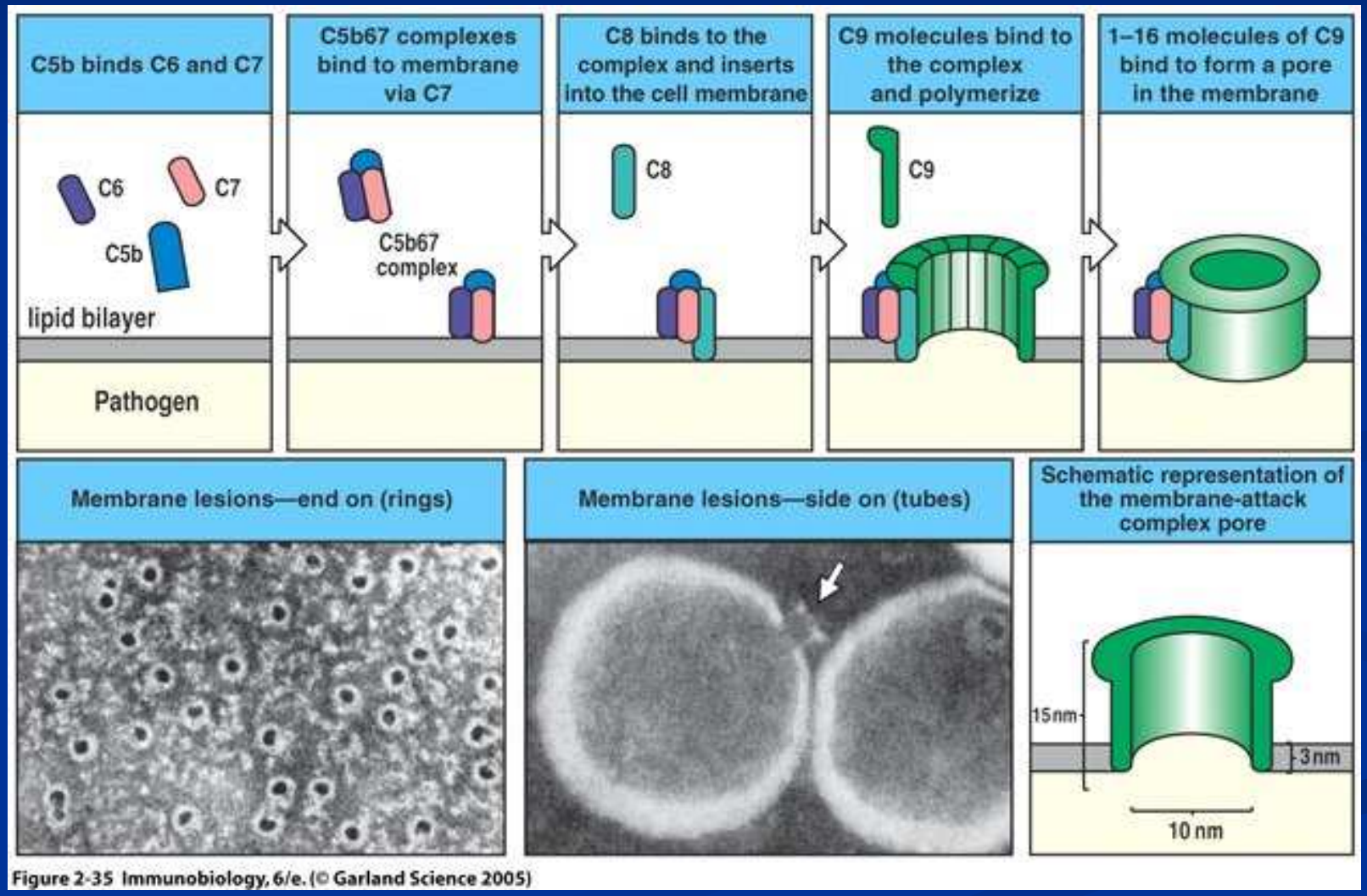


Figure 2-35 Immunobiology, 6/e. (© Garland Science 2005)

The spread of viruses is limited by the interferon response

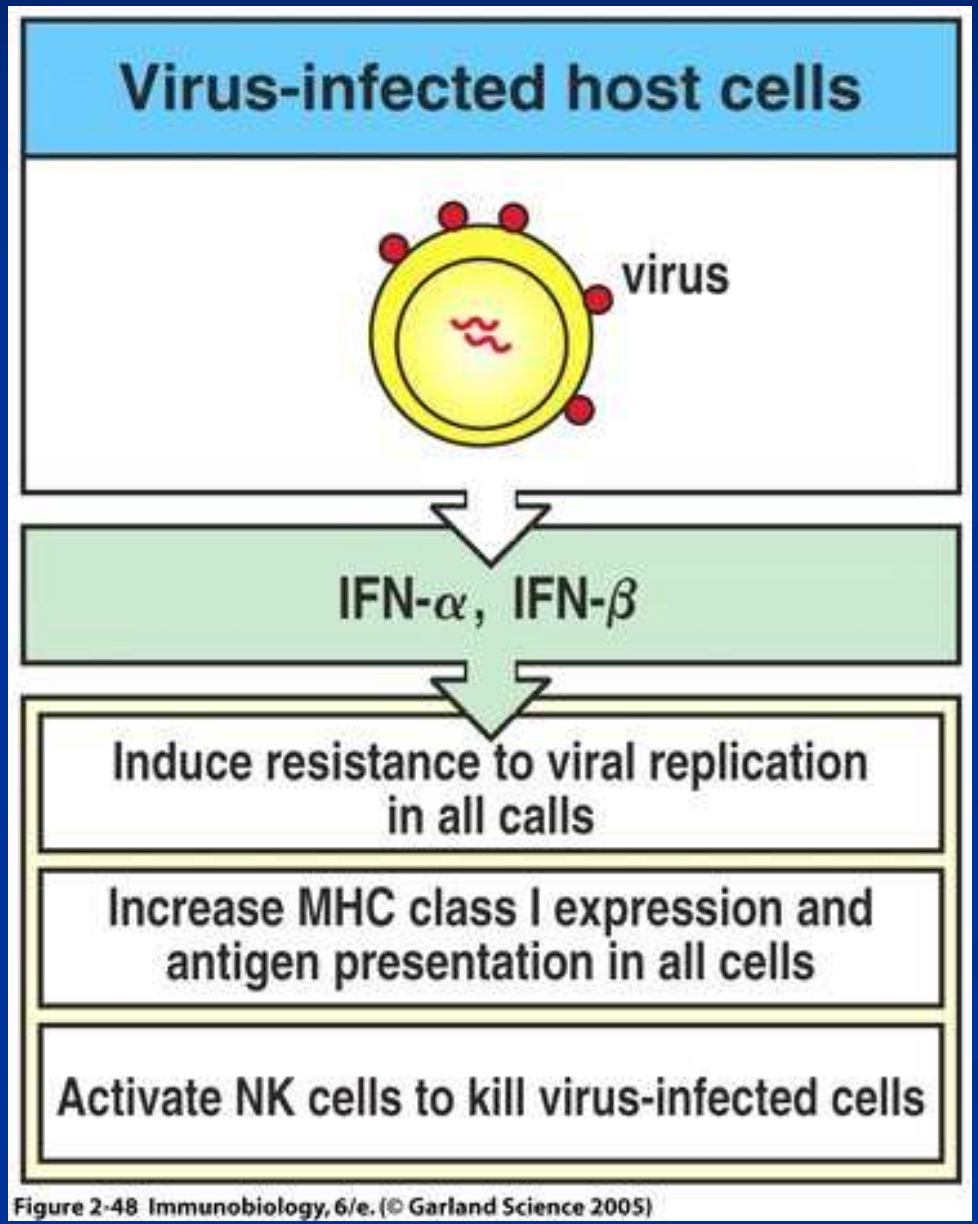


Figure 2-48 Immunobiology, 6/e. (© Garland Science 2005)

Viral nucleic acids stimulate the production of interferon by the infected cell, which mediates both autocrine and paracrine protective responses

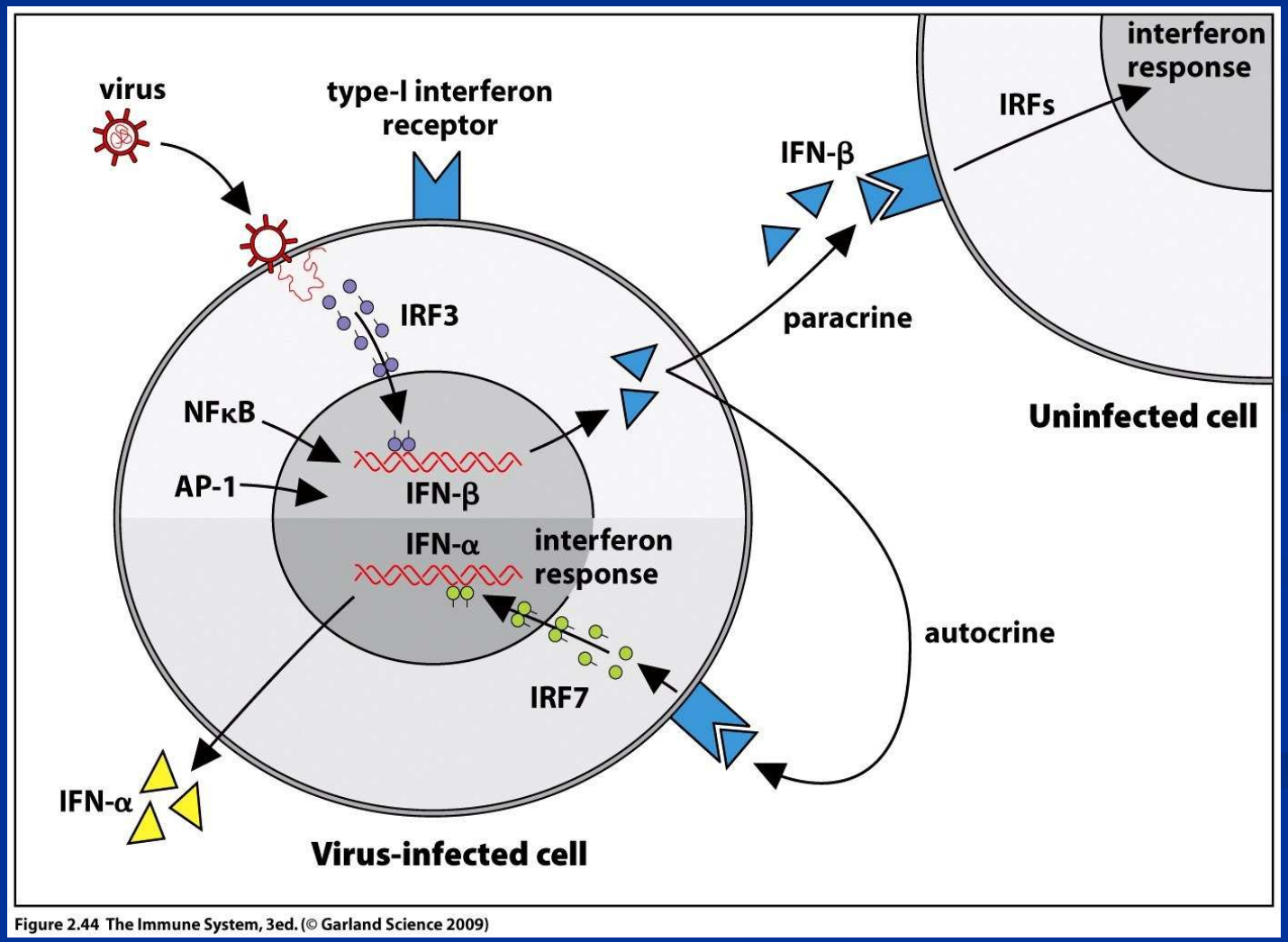
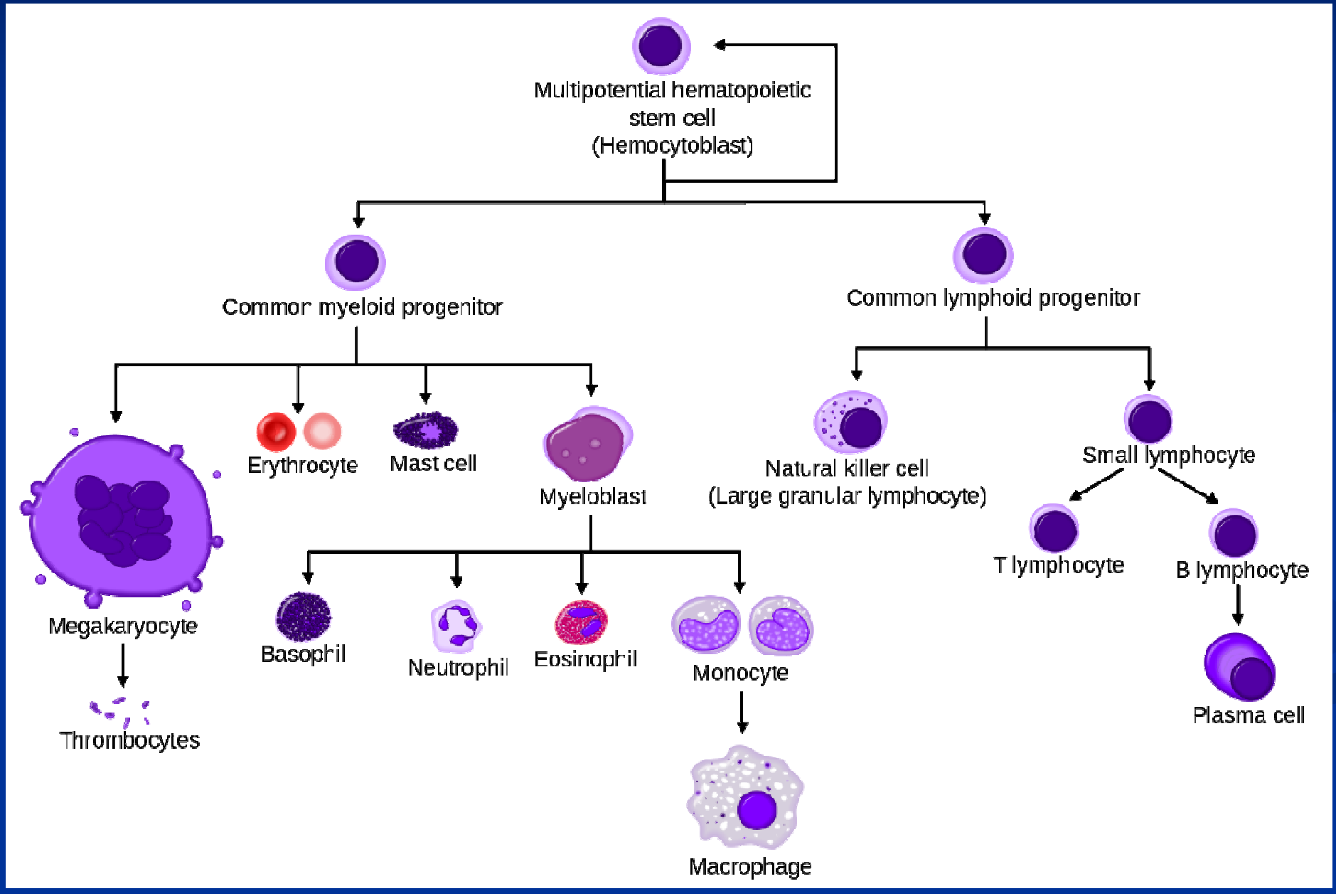


Figure 2.44 The Immune System, 3ed. (© Garland Science 2009)

Innate Immunity- 1st responders



NK cells

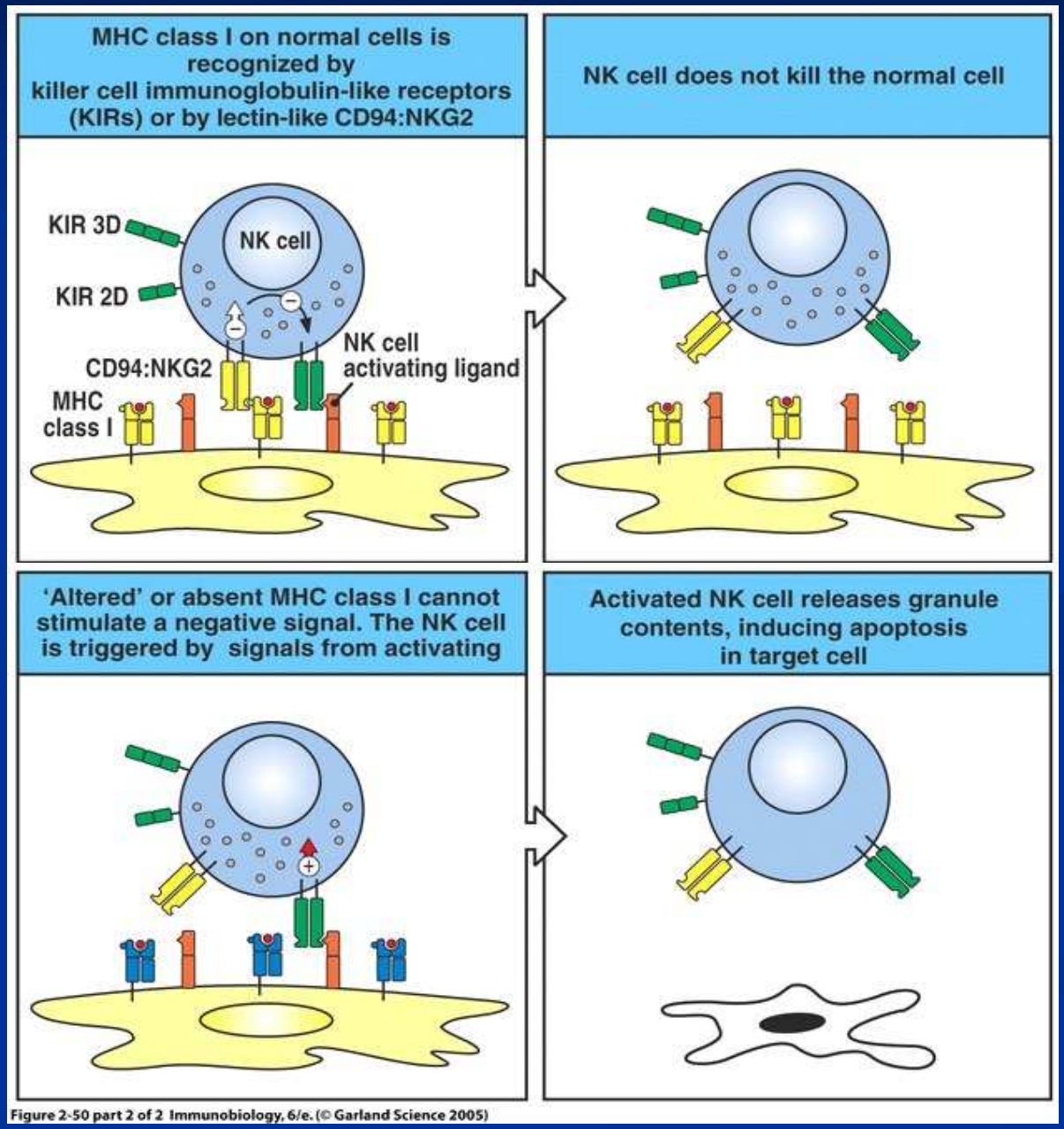


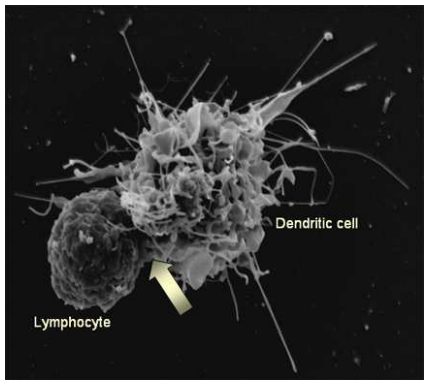
Figure 2-50 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

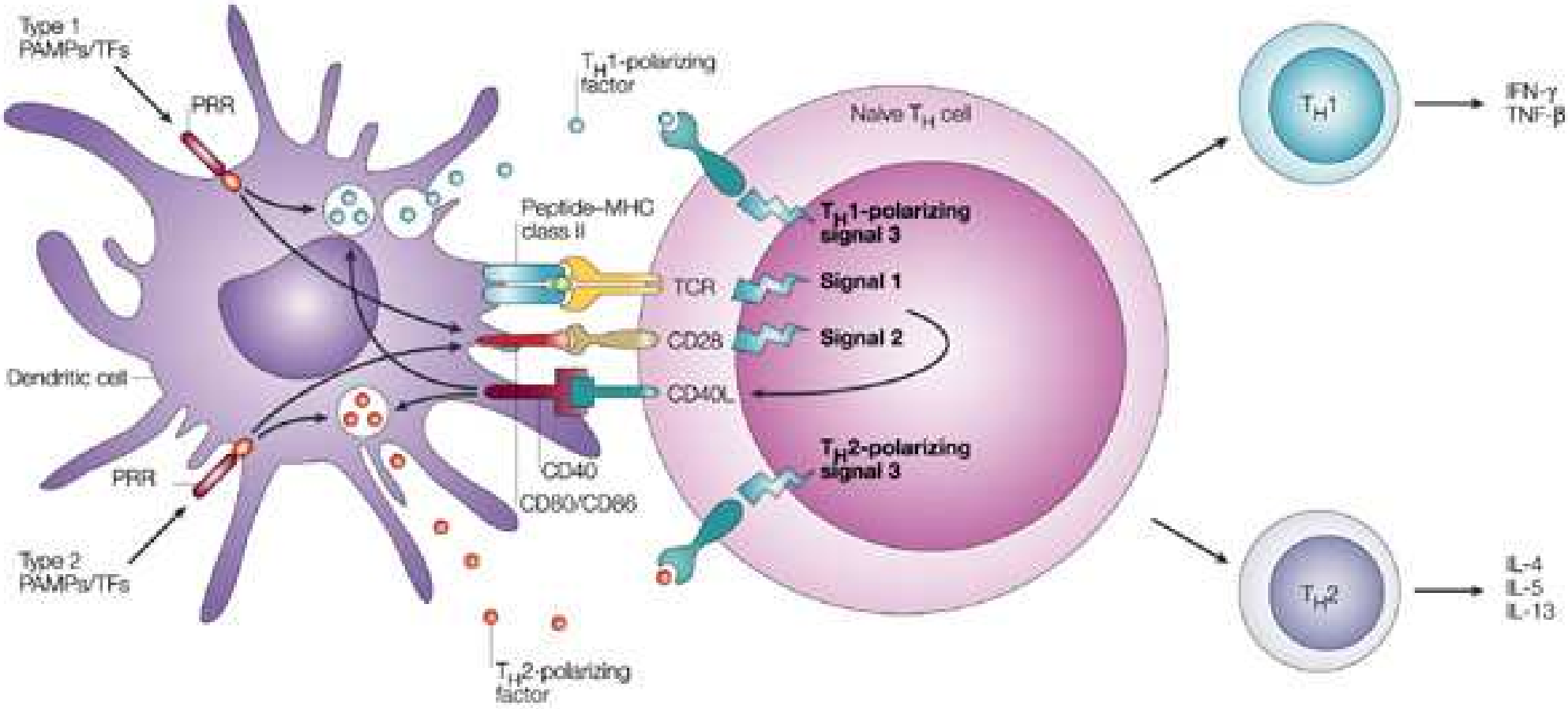
- Overlap innate and adaptive immunity
- Stimulated by Type I interferons
- Kill cells with down-regulated MHC I expression
  - Down-regulated by viruses and tumors trying to avoid CD8+ T-cell killing
- Kill "non-self" via mostly shared mechanisms with cytotoxic T-cells
  - TRAIL
  - GranzymeB
  - Perforin

Innate Immunity and Inflammation

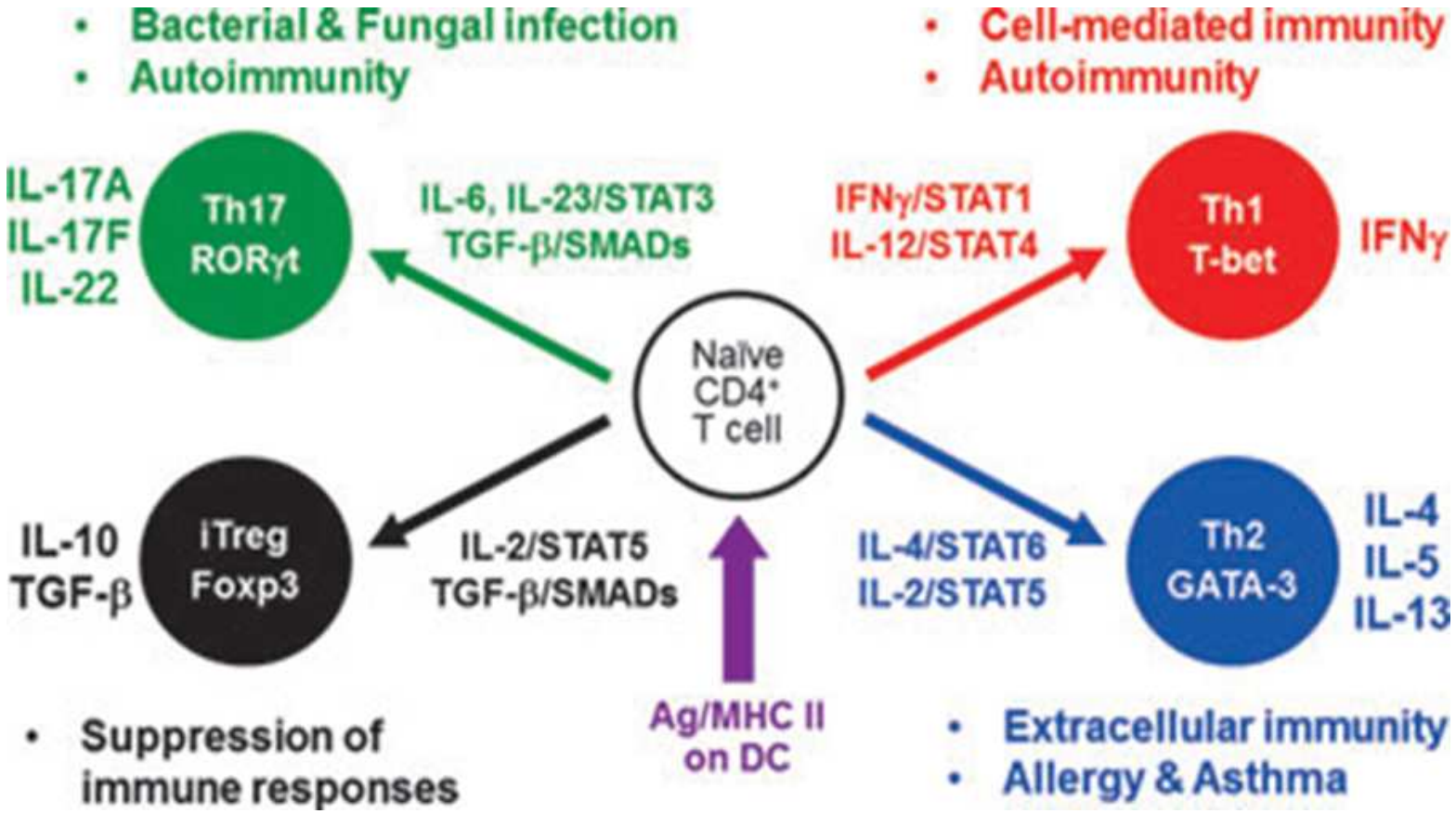
- 1) Respond rapidly to tissue damage
  - physical and chemical barrier
  - recruitment of immune cells to site of injury
- 2) Limit spread of infection
  - identification and removal of foreign substances
  - activation of the complement cascade
  - activation of coagulation cascade
- 3) Initiate adaptive immune response
  - antigen presentation and cytokine production
- 4) Initiate tissue repair

Adaptive response initiation: Antigen presentation to T cells





The context in which macrophage-derived dendritic cells present antigen to T-cells determines the type of adaptive T cell response that follows



Cell

Leading Edge
Review

Hallmarks of Cancer: The Next Generation

Douglas Hanahan[1,2,*] and Robert A. Weinberg[3,*]
[1]The Swiss Institute for Experimental Cancer Research (ISREC), School of Life Sciences, EPFL, Lausanne CH-1015, Switzerland
[2]The Department of Biochemistry & Biophysics, UCSF, San Francisco, CA 94158, USA
[3]Whitehead Institute for Biomedical Research, Ludwig/MIT Center for Molecular Oncology, and MIT Department of Biology, Cambridge, MA 02142, USA
*Correspondence: dh@epfl.ch (D.H.), weinberg@wi.mit.edu (R.A.W.)
DOI 10.1016/j.cell.2011.02.013

An Emerging Hallmark: Evading Immune Destruction

An Enabling Characteristic: Tumor-Promoting Inflammation

- Immune surveillance in cancer
  - Carcinogen-induced tumors arise more frequently and quickly in immunodeficient mice
  - Cancer cells that arise in immunodeficient mice are inefficient at initiating secondary tumors in syngeneic immunocompetent mice

- Immune surveillance in cancer
  - Heavy CTL and NK cell infiltration predicts improved outcome in several human tumors
  - Immunosuppressed organ transplant recipients develop donor-derived cancer from ostensibly tumor-free donors

- Immune escape in cancer
  - Clinically apparent tumors variably suppress the anti-tumor immune response
  - Tumor-associated inflammation can enhance tumor progression

Immunoediting

Normal Cells

•Inflammation/radiation/carcinogens/viruses

Transformed Cells

•Tumor antigens/peptide-MHC I/ MHCII cross-presentation/NKG2D

Protection

1) Elimination/Immunosurveillance

•IFN γ and IFN α / β
•Perforin
•Trail
•Th1 and T_c T cells, NKT, NK, $\gamma\delta$ T cells

2) Equilibrium/Tumor Persistence

•Genetic instability and immune selection
•Tumor variants
•Immune exhaustion/inhibition

•Tumors may remain dormant

3) Escape/Tumor Progression

•T regulatory cells – IL-10 and TGF β

•Tumor-derived cytokines, growth factors, chemokines

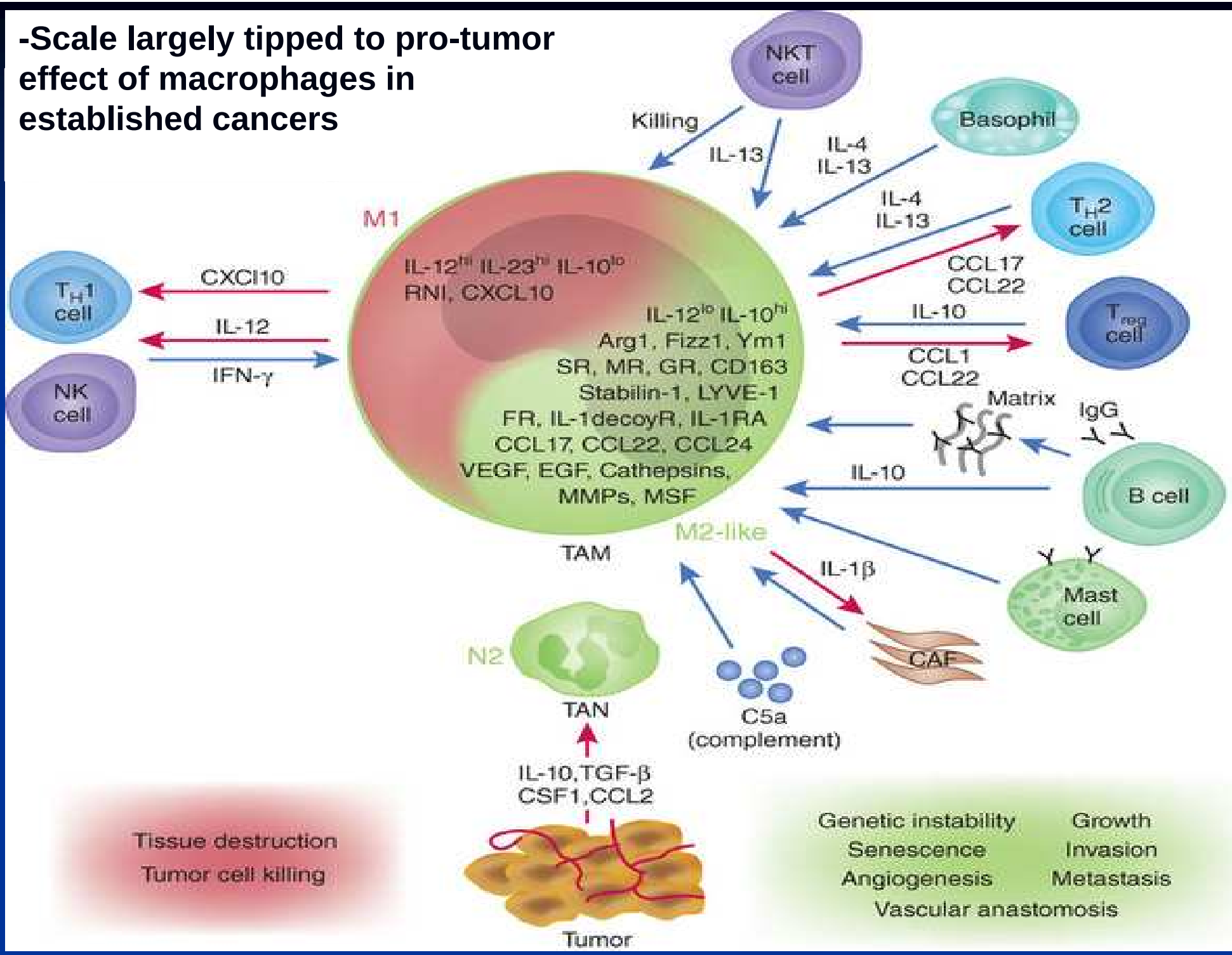
•Myeloid-derived suppressor cells (MDSC)

Adapted from Dunn G et al, Nat Rev Immunol 6, 836-48 (2006).

Chronic Inflammation has a known role in cancer initiation

- Tobacco, asbestos → Bronchial CA
- Alcohol → Hepatocellular CA, Gastric CA, Pancreatic CA
- *Helicobacter pylori* → MALT lymphoma
- Shistosoma → bladder CA
- HCV → Hepatocellular CA
- HPV → Cervical CA
- Endogenous inflammation in inflammatory bowel disease → Colon CA
- Barrett's esophagus → Esophageal CA

-Scale largely tipped to pro-tumor effect of macrophages in established cancers



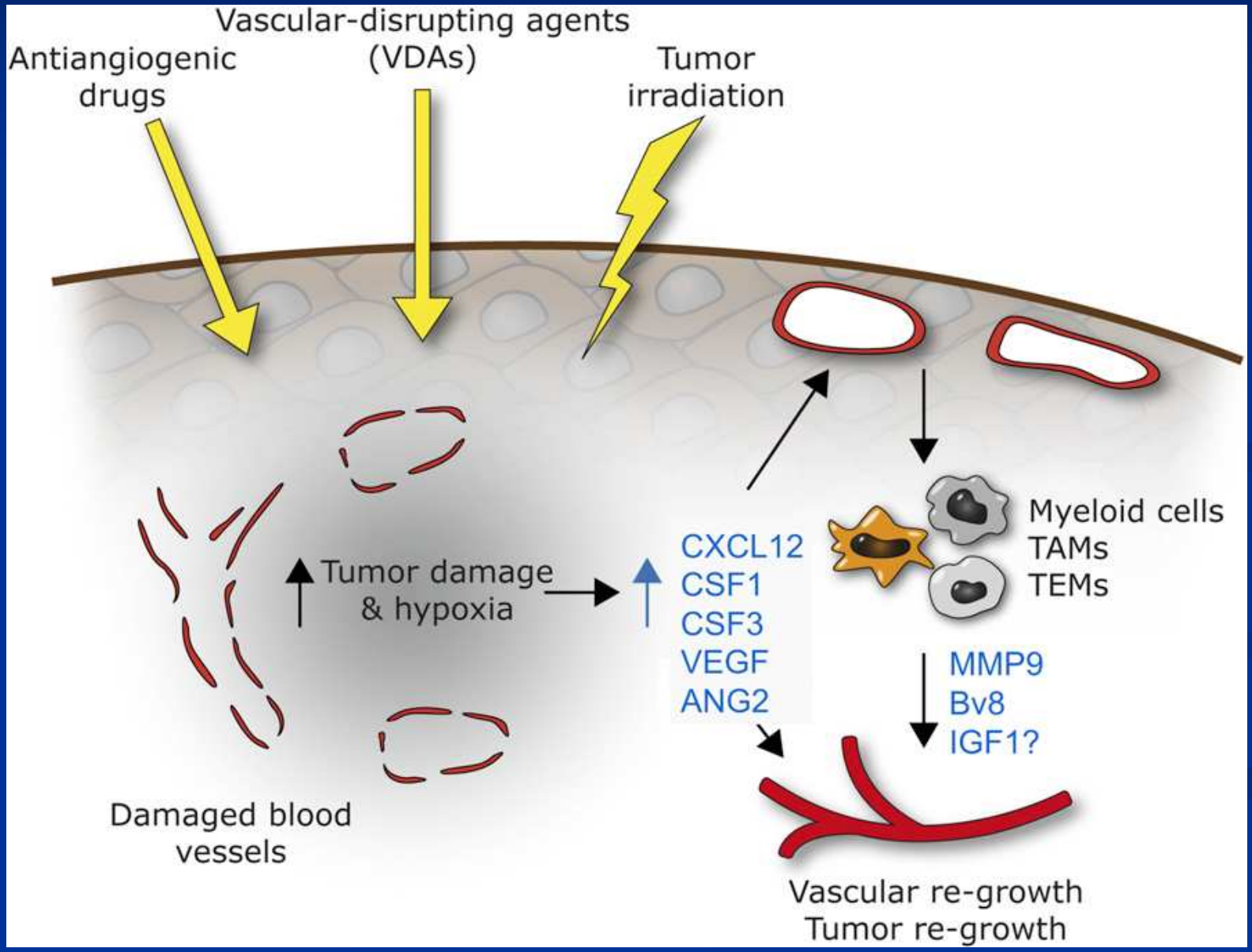
Macrophage and chemotherapy

- TAM modulate responses to chemotherapy
- Macrophages and DCs are known to mediate ‘‘immunogenic cell death’’ (ICD) which some chemotherapies induce in some tumor models
  - release of ‘‘eat-me’’ signals (e.g. ATP and high-mobility group B1 [HMGB1]) from dying tumor cells enhanced by some chemotherapies
  - monocyte activation and enhancement of their APC capacity and promotion of T cell responses against immunogenic tumors
- Antitumor activity of some cytotoxic agents may depend on their ability to reprogram pro-tumoral macrophages

Macrophage and chemotherapy

- TAM depletion (anti-CSF1 antibodies) enhances the efficacy of some combination chemotherapy
  - cyclophosphamide, methotrexate, and 5-fluorouracil in chemoresistant, human breast cancer xenografts in immunodeficient mice
  - Paclitaxel in immunocompetent mice via increased anti-tumor CD8+ T-cell responses when macrophages were depleted
- Macrophage-derived cathepsins protect cancer cells from the direct cytotoxic effects of several chemotherapeutics
  - CathepsinB → inflammasome →IL-1b → IL-17 → blunted chemo effect
- Macrophage-derived factors activate STAT3 in cancer stem cells to promote chemoresistance
- Ultimate effect may depend on tumor immunogenicity, sensitivity of macrophages to drug, and the inherent state of the macrophages in the particular tumor

Macrophages initiate a wound reparative program that enhances tumor regrowth



Neutrophil

Potential

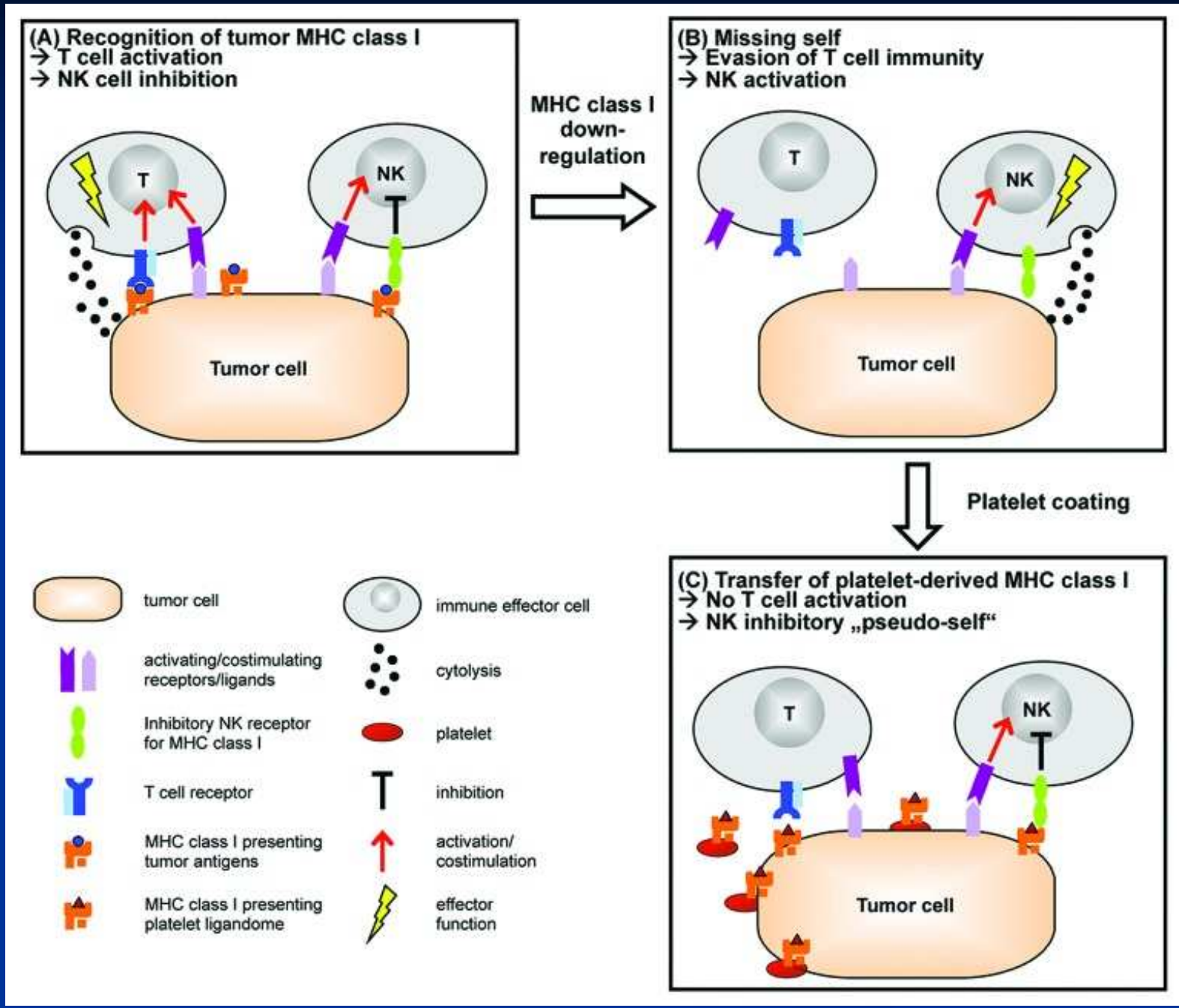
- Tumor cell killing through reactive oxygen species???

Reality

- Genotoxicity associated with neutrophil ROS may initiate cancer
- Neutrophilia, and high neutrophil:lymphocyte (N:L) ratio associated with poor outcome in multiple tumor types
- Promote angiogenesis
- Anti-tumor T-cell suppression
- Tumor cell migration, invasion and metastasis

NK cells

- Prevent tumor progression
  - Immunosurveillance → Elimination phase
  - Cell stress
  - Non-self (low or absent MHC I)
- Prevent tumor metastasis
  - Attack tumor emboli in the lungs
- NK cell tumor targeting can be inhibited by platelets
  - “Pseudo-self”



Oncoimmunology. 2012 July 1; 1(4): 557–559.

Summary

- Function of Innate Immunity
  - 1) Respond rapidly to tissue damage
  - 2) Limit spread of infection
  - 3) Initiate adaptive immune response
  - 4) Initiate tissue repair
- Innate Immunity in Cancer
  - Neutrophils
    - Promote tumor initiation
    - Promote tumor spread
  - Macrophages
    - Pro and anti- tumor effects in tumor initiation
    - Promote tumor spread
  - NK cells
    - Inhibit tumor initiation
    - Inhibit tumor spread

Questions?

1) What is the importance of innate immunity in cancer?

- A) Innate immunity initiates malignant transformation via neutrophil-derived genotoxic stress
- B) Innate immunity eradicates transformed tumor cells via NK recognition of non-self or cell stress
- C) Innate immunity promotes tumor spread via myeloid cell mediated CD8+ T cell inhibition
- D) Innate immunity promotes tumor spread via angiogenesis upregulation
- E) All of the above

2) Which of the following is not a function of Macrophages?

- A) Phagocytosis and presentation of microbes or tumor-associated antigens to T-cells
- B) Upregulation of acute phase protein production and systemic inflammatory response
- C) Extrude a web of fibers composed of chromatin and serine proteases that trap and kill microbes extracellularly
- D) Promote wound healing through increased angiogenesis
- E) All of the above are functions

3) Complement cascade can be activated by the lectin pathway and the alternative pathway?

- A) True
- B) False

4) Tumor associated macrophages are mostly M2-polarized, which is desirable for tumor eradication?

- A) True
- B) False

5)Tumor infiltrating neutrophils promote tumor cell invasion and metastasis via matrix metalloproteinases ?

- A) True
- B) False