Cells of Immunity – I

(Innate cells)

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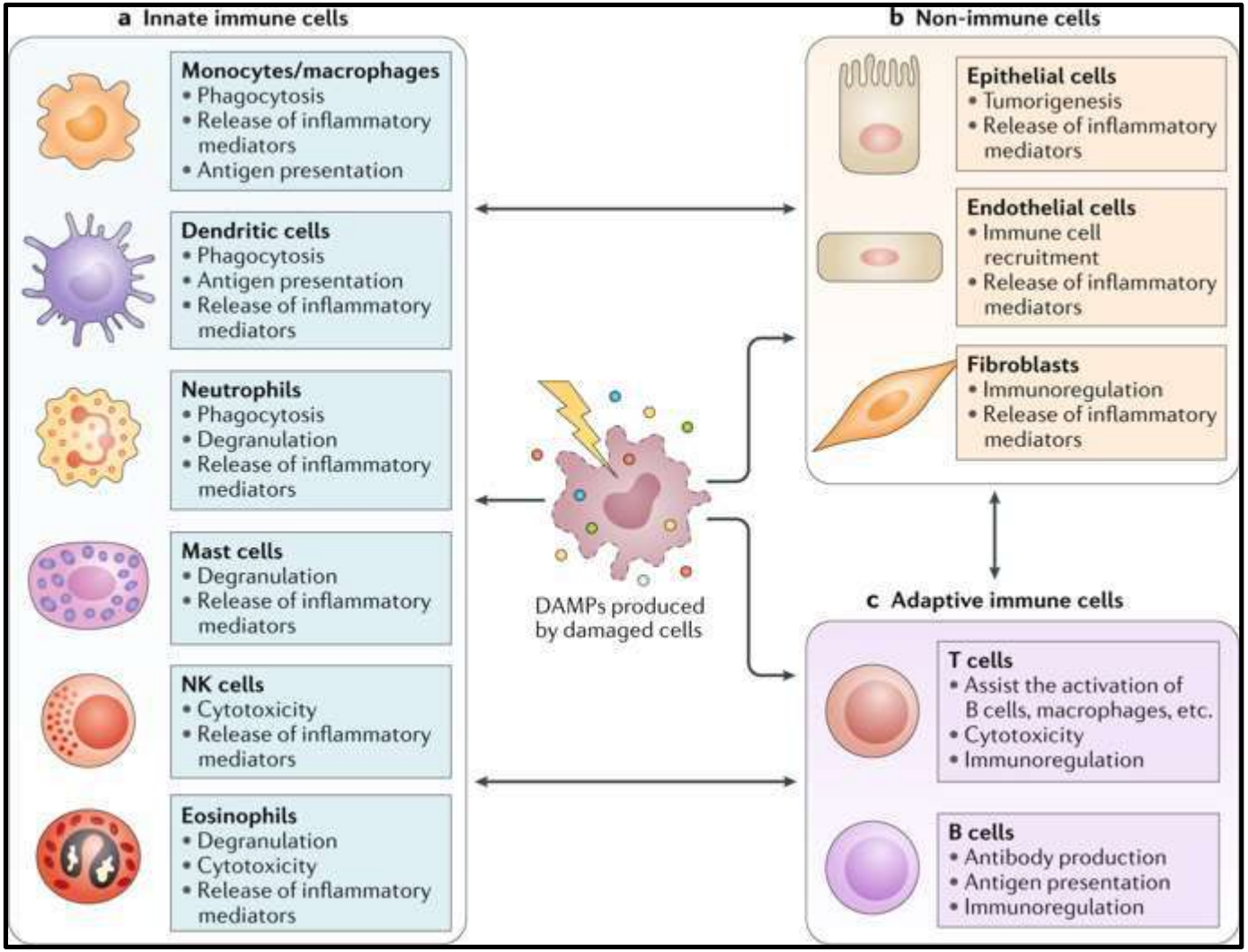
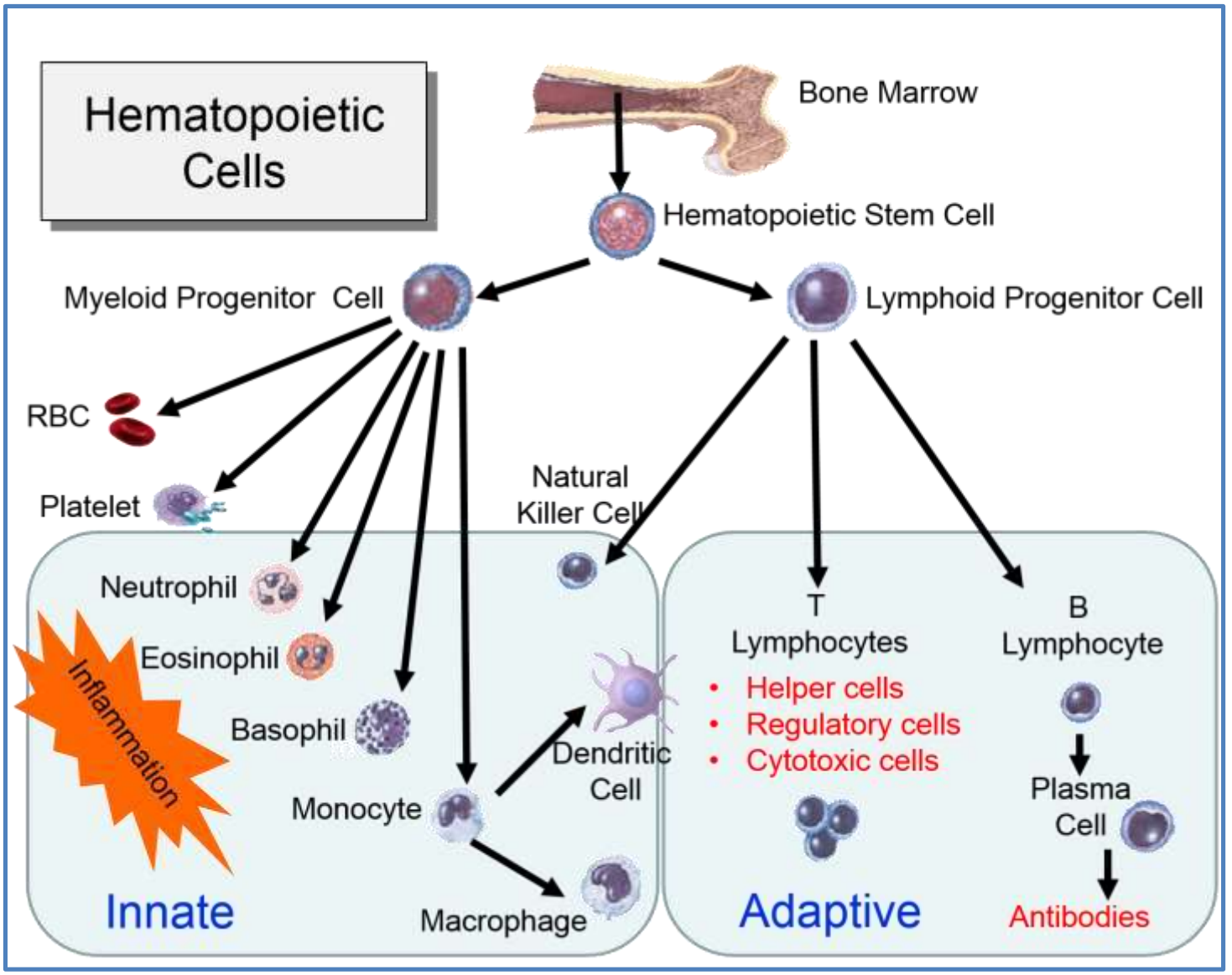
CELLS OF THE IMMUNE SYSTEM



IMMUNE CELLS



IMMUNE CELLS

Cells of the Immune System

- Lymphoid lineage:
  - Central cells of the IS
  - responsible for adaptive IR
  - Provide diversity, specificity, memory,self – nonself recognition
  - 20-40% of WBC's
  - 99% of cells in lymph

  Includes three cell types:

  B cells

  T cells

  Natural Killer cells

- **Myeloid lineage**:
  - Central cells of innate immunity
  - responsible for triggering inflammation, phagocytosis, antigen presentation, cytokine release
  - 60-80% of WBC's

  Includes:

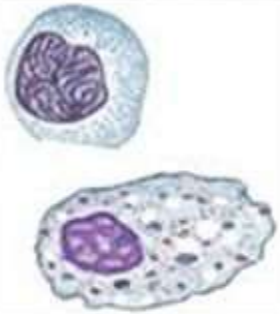
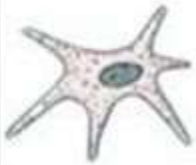
  PMN granulo's – neutrophils

  eosinophils

  basophils

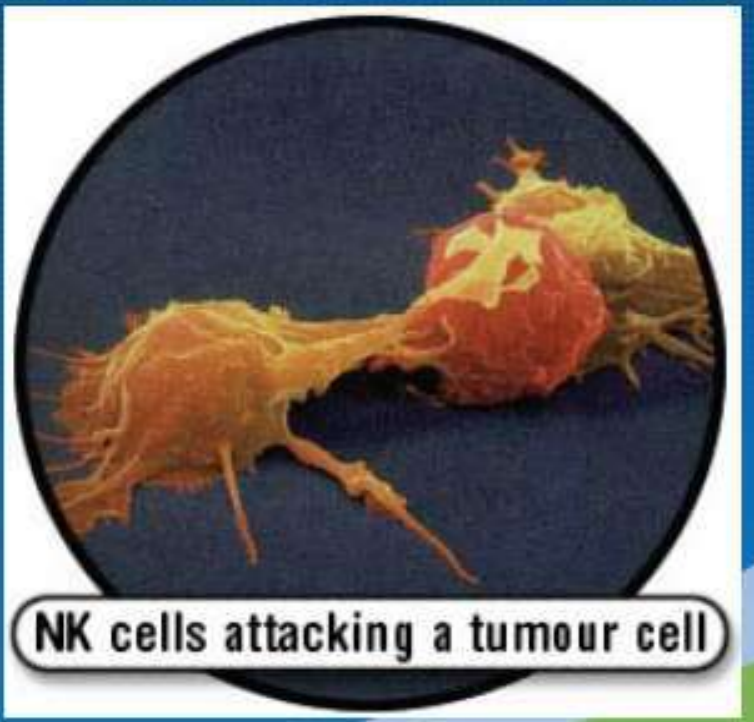
  Mononuclear agranulo's - monocytes macrophages

Cells of the Immune System

	Basophils and Mast Cells	Neutrophils	Eosinophils	Monocytes and Macrophages	Lymphocytes and Plasma Cells	Dendritic Cells
						
% of WBCs in blood	Rare	50–70%	1–3%	1–6%	20–35%	NA
Subtypes and nicknames		Called "polys" or "segs" Immature forms called "bands" or "stabs"		Called the mononuclear phagocyte system	B lymphocytes, Memory cells Plasma cells T lymphocytes Cytotoxic T cells Helper T cells Natural killer cells	Also called Langerhans cells, veiled cells
Primary function(s)	Release chemicals that mediate inflammation and allergic responses	Ingest and destroy invaders	Destoy invaders, particularly parasites	Ingest and destroy invaders Antigen presentation	Specific responses to invaders, including antibody production	Recognize pathogens and activate other immune cells by antigen presentation
		Phagocytes				
Classifications	Granulocytes					
			Cytotoxic cells		Cytotoxic cells (some types)	
					Antigen-presenting cells	

Innate Immune Effector Cells

- Epithelial Cells
- Neutrophils (PMNs)
- Monocytes and Macrophages
- Dendritic Cells
- NK Cells



NK cells attacking a tumour cell

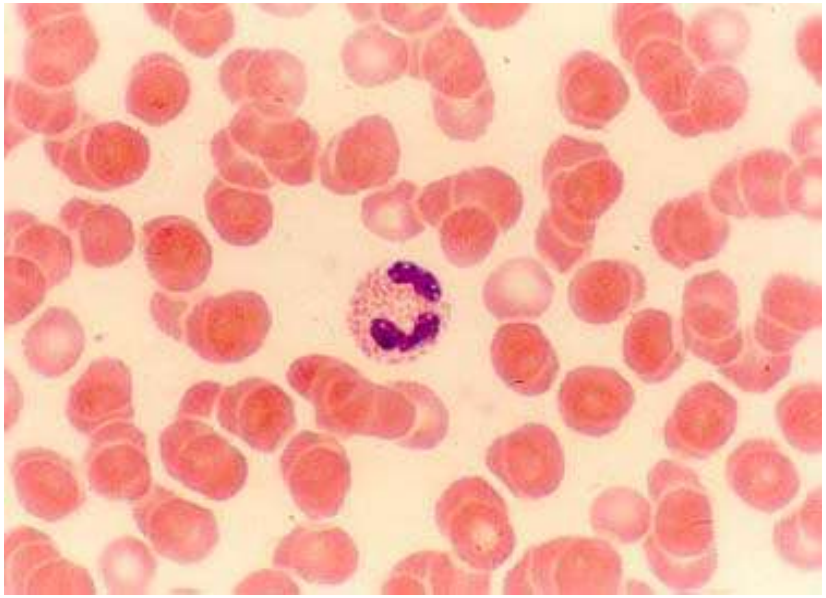
Innate defense Cells at the Surface

Epithelial cells of skin and mucous membranes

- **physical barrier**
- **desquamation**
- **keratinized tissue**
- **cilia**
- **secretion of chemicals such as sebum, mucous, sweat, antimicrobial peptides, defensins, etc.**

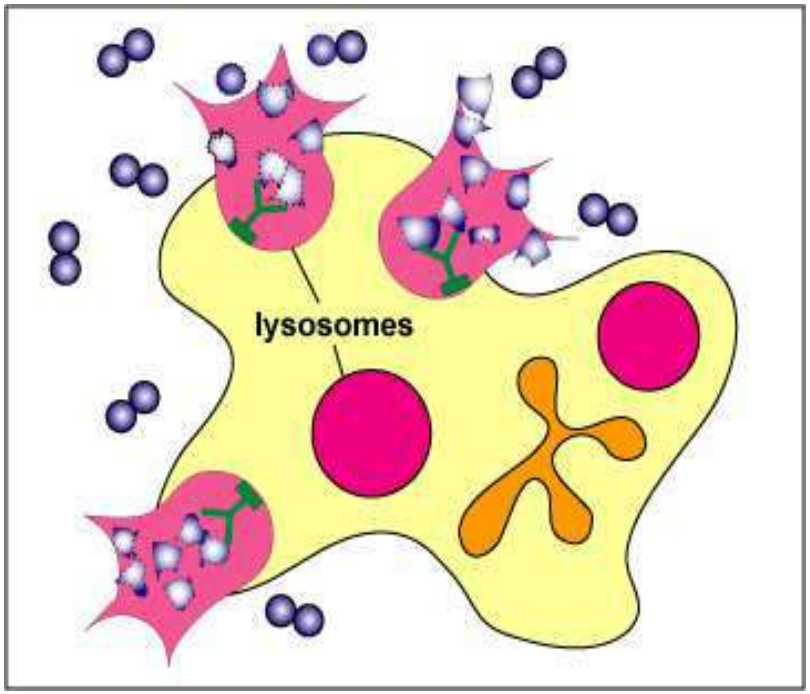
Innate defense Cells in the Blood

Eosinophil



- CD67 membrane marker
- characteristic lobed nucleus, cytoplasmic granules
- life span is 8-12 days
- phagocytosis, extra cellular killing (piece meal degranulation), inflammation and tissue damage
- anti-parasitic, allergies
- immunity against bacterial, viral, and fungal infections
- DNA-based extracellular traps (ETs)

Extracellular Destruction of Bacteria by a Phagocyte



- If the phagocyte is overwhelmed with microorganisms, the phagocyte will empty the contents of its lysosomes by a process called peace meal degranulation in order to kill the microorganisms or cell extracellularly.
- These released lysosomal contents, however, also kill surrounding host cells and tissue.
- Most tissue destruction associated with infections is a result of this process

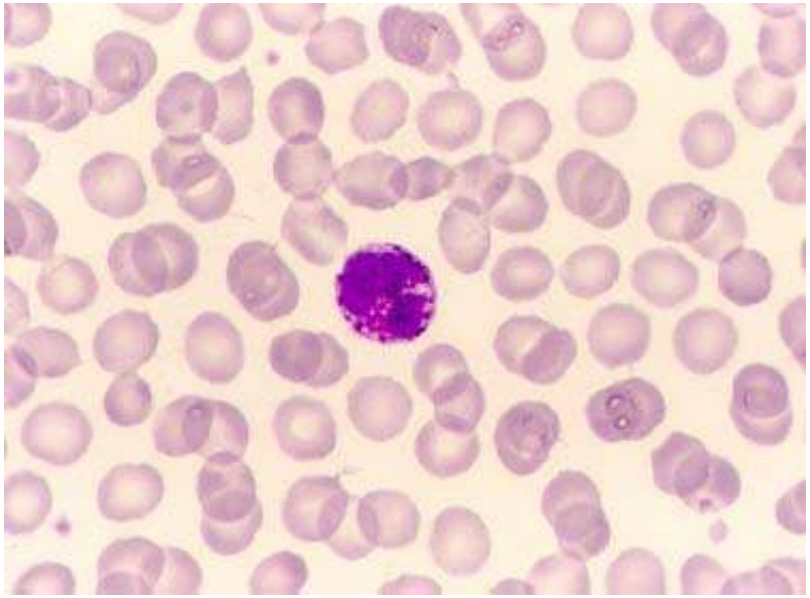
Piecemeal degranulation (PMD)

- **Degranulation** is a term used to define processes where there is release of granule proteins from viable cells, or the release of intact/ruptured granules from “dying” cells,
- Four modes of degranulation have been reported for eosinophils, namely: (i) *classical exocytosis* (ii) *compound exocytosis*; (iii) *piecemeal degranulation* (PMD); and (iv) *cytolysis*
- **PMD** is an unconventional secretory pathway characterized by vesicular transport of small packets of materials from the cytoplasmic secretory granules to the cell surface.
- **Termed piecemeal degranulation (PMD) because of a “piece by piece” release of secretory granule contents, this secretory process is now recognized as a central secretion mode during inflammatory responses**.
- In contrast to classical granule exocytosis, which involves granule fusion with the plasma membrane and release of the total granule content, piecemeal degranulation enables release of specific granule-stored proteins.
- During piecemeal degranulation in eosinophils, a distinct transport vesicle system is mobilized and enables regulated release of granule-stored proteins such as cytokines and major basic protein (MBP), eosinophil cationic protein (ECP), eosinophil-derived neurotoxin (EDN) , and eosinophil peroxidase (EPX)]

Extracellular Destruction of Bacteria by a Phagocyte (extracellular traps – Ets and ETiosis)

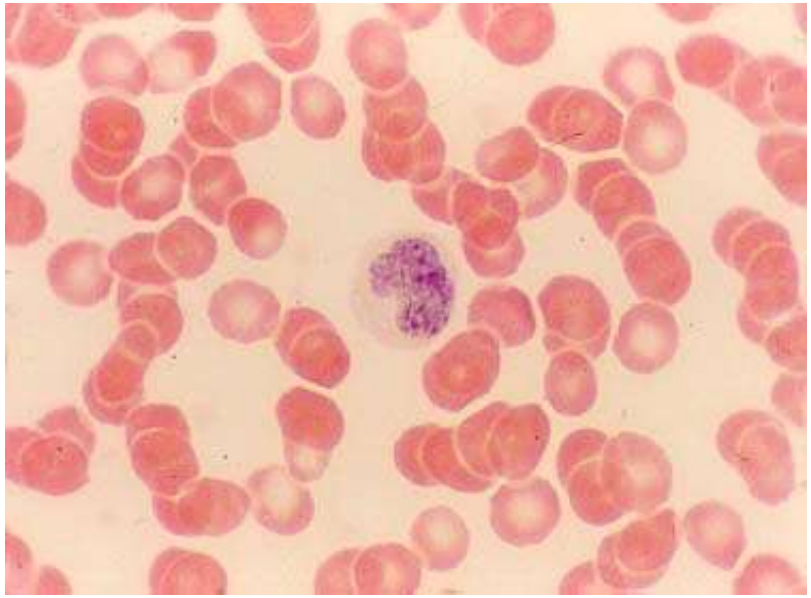
- **Extracellular traps (NETs)** are extracellular nuclear DNA deposits of phagocytic cells that are studded with high local concentrations of anti-microbial agents (peptides, proteases, reactive oxygen species).
- These are released into extracellular matrix to degrade virulence factors of and aid in control of growth and proliferation of pathogens by killing them.
- Neutrophils extracellular traps **(NETs)** were first discovered in 2004 as a phagocytosis-independent anti-microbial pathway. Similar extracellular traps **(ETs)** have subsequently been observed from other cells of the innate family members, for example, mast cells, monocytes, tissue macrophages , and eosinophils.
- The phenomenon of release of ETs from immune cells associated with rupture of the cell membrane is a novel cell death pathway (distinct from necrosis and apoptosis), and referred collectively as **ETosis**

Basophil



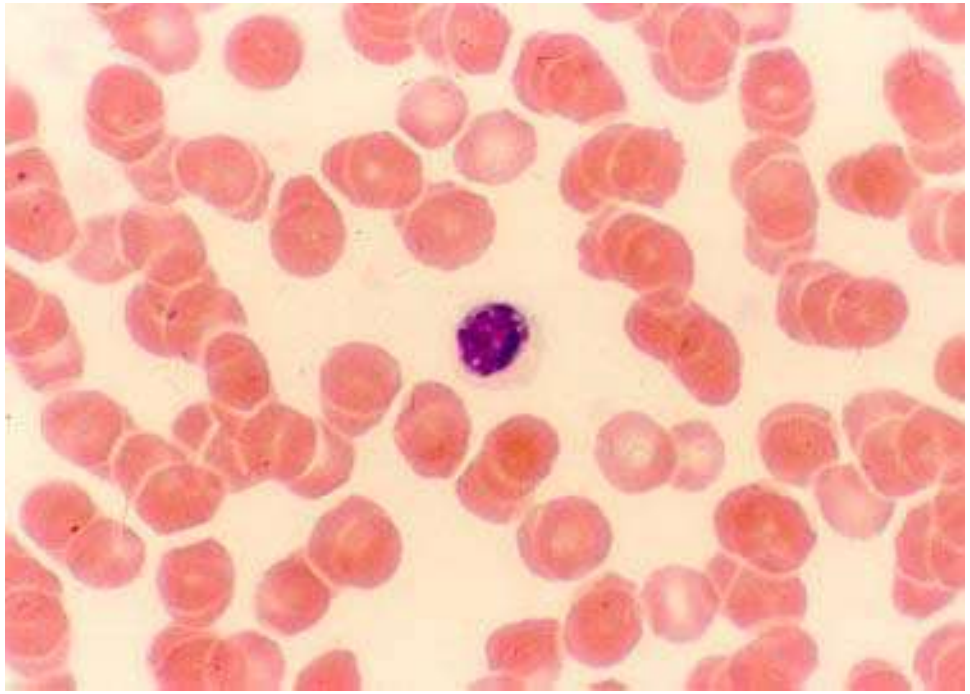
- CD123 and CCR3 membrane marker and absence of HLA-DR
- characteristic lobed nucleus, cytoplasmic granules
- release histamine , leukotrienes , and prostaglandins (vasoactive amines), non-phagocytic, inflammation, and coagulation
- involved in hypersensitivity reactions.

Monocyte



- CD14 and CD16 membrane marker
- compact nucleus, no cytoplasmic granules
- differentiate into macrophages or one of a family of phagocytic cells (dendritic cells)
- long lived (months)
- phagocytosis

Lymphocyte



- CD2 membrane marker
- compact nucleus, no cytoplasmic granules
- long lived (years)
- three types – B-cells and T-cells(adaptive), and NK cells (innate and adaptive)
- non-phagocytic, adaptive and innate immunity (ILC)

Innate Lymhpoid cells

What and who are ILC's?

They are the cells acting at the intersection of immunity.

Just like Innate Immune cells:

- They respond to infection quickly.
- They do not express Antigen-specific receptors.
- They do not undergo clonal selection and expansion.

Just like T-cells:

- They have similar functions and phenotypes.

ILCs – general characteristics

- ILCs are largely tissue-resident cells **found at barrier surfaces** – skin, lungs & intestines and also in adipose tissues and MALT
- ILCs have classical lymphoid cell lineage and morphology, characterized by the expression of **CD127 and CD161** and lack of other lineage markers
- **ILCs represent the innate version of helper and cytotoxic T cells** as part of the innate immune, but do not express the diversified antigen receptors expressed on T cells and B cells.
- **ILCs contribute to innate immune responses** to commensals and pathogens at mucosal barriers, **potentiate adaptive immunity, and regulate tissue inflammation.**
- ILCs also contribute to tissue homeostasis, morphogenesis, metabolism, repair, regeneration, and dialog with the nervous system
- The microenvironments of local tissue influence the plasticity and function of ILCs, i.e., ILC subsets can change their phenotype and functional capacities depending upon local cytokine profile

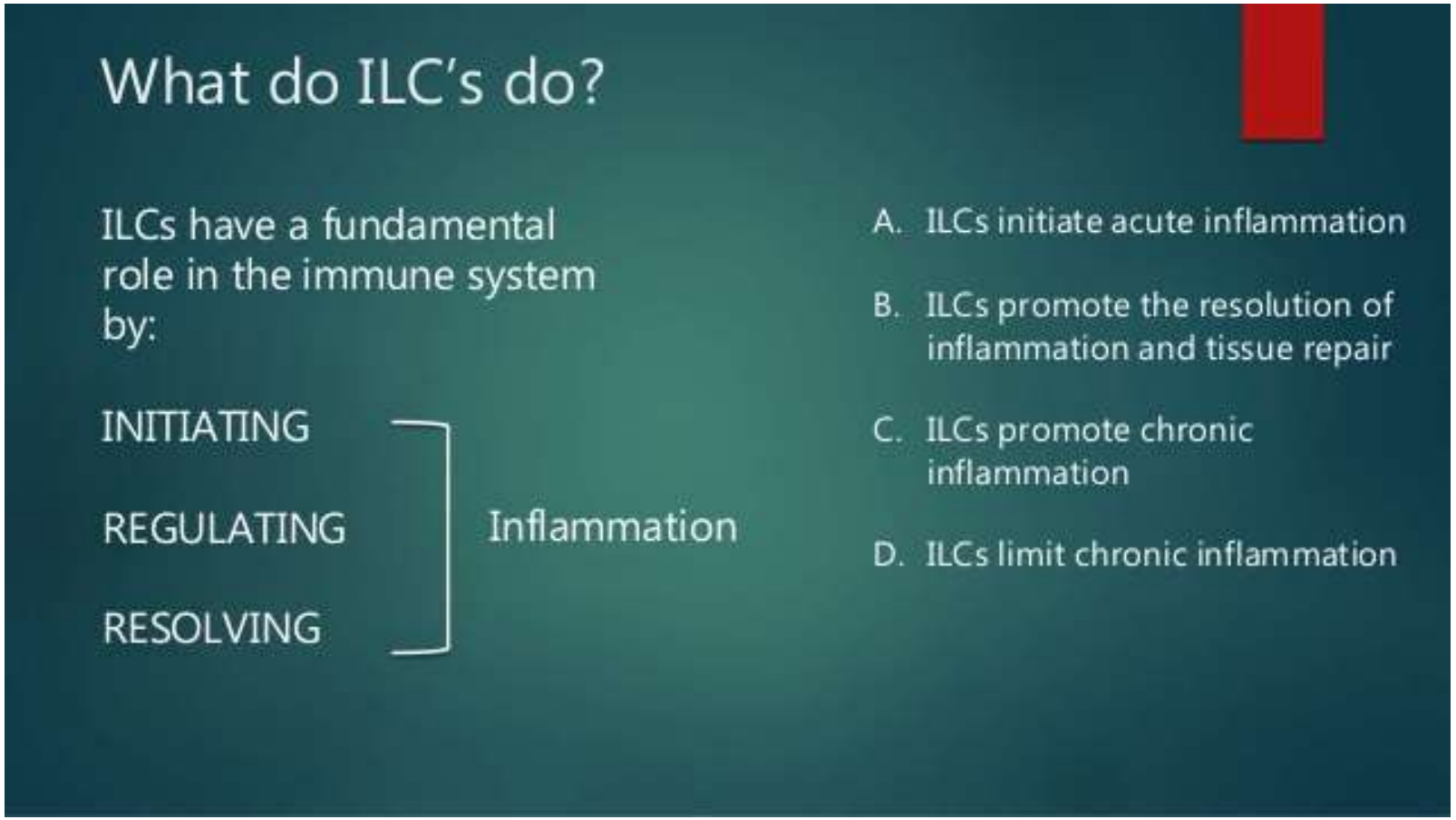
ILCs – classification / nomenclature

- Three major types- Group 1,2 and 3
- Group 1 ILC produce IFN-γ and comprise of NK cells and ILC1s
- Group 2 ILC produce IL-4, IL-5 and IL-13. Group 2 ILC contains a single subset, ILC2s
- Group 3 ILC produce IL-17 and/or IL-22 and IFN-γ. Group 3 ILCs include ILC3s and LTi cells.
- The International Union of Immunological Societies (IUIS), has proposed a new nomenclature classifying ILCs into five subsets—NK cells, ILC1s, ILC2s, ILC3s, and LTi cells—based on their development and functions

Types of ILCs - location

- ILC1s are tissue-resident cells, whereas NK cells circulate in the bloodstream
- ILC2s are tissue-resident cells and in peripheral blood
- ILC3s are abundant at mucosal sites

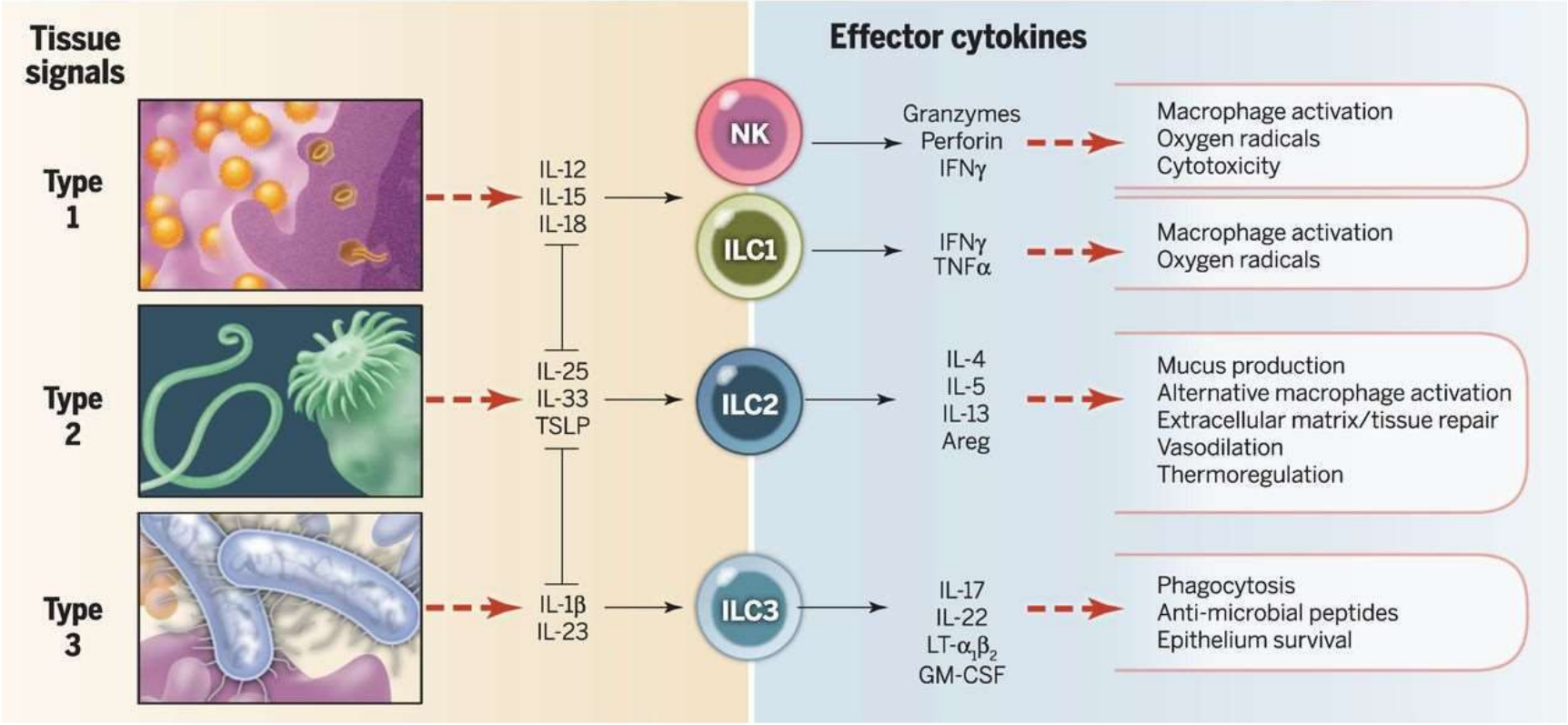
Functions of ILCs



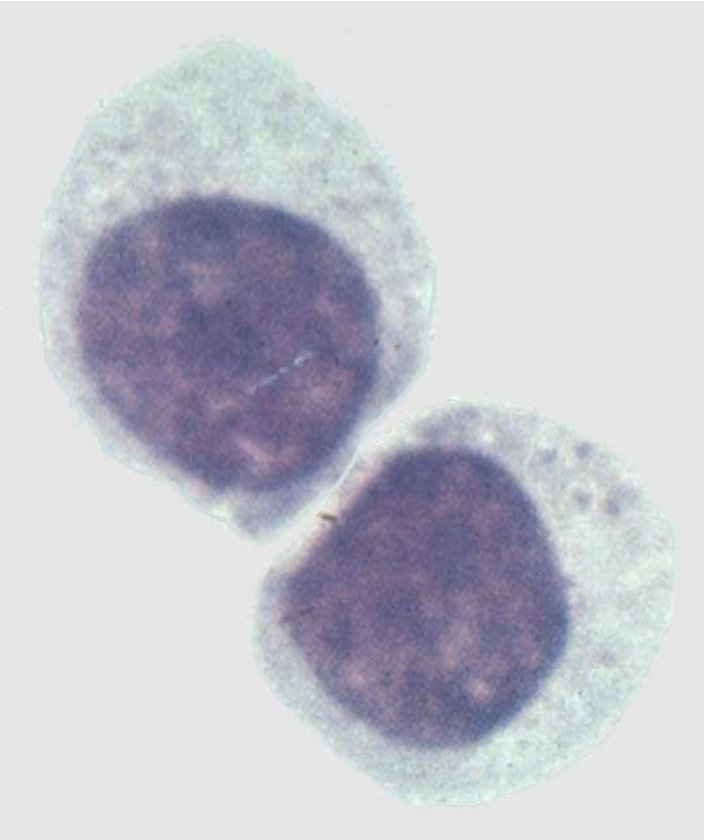
Types of ILCs - functions

- ILC1s, ILC2s, and ILC3s mirror $CD4^+$ Th1, Th2, and Th17 cells, respectively, in terms of function, whereas NK cells mirror the functions of $CD8^+$ Tc cells.
- ILC1s react to intracellular pathogens, such as viruses, and to tumors (~Th1)
- ILC2s respond to large extracellular parasites and allergens (~Th2).
- ILC3s are involved in the innate immune response to extracellular microbes, such as bacteria and fungi (~Th17), and the containment of intestinal commensals
- ILCs act early in the immune response, whereas the T cell response takes several days.
- A few days after the initiation of an immune reaction, both ILCs and T cells are active, and they cross-regulate each other.
- The lymphoid tissue-inducer cells (LTi cells) induce the development of most of the secondary lymphoid organs.
- ILCs play a key role in homeostasis - regulates symbiotic microbiota, thermogensis.
- ILCs are also involved in tissue tolerance (to irradiation, toxins, etc) and regeneration of damaged tissue.

Functions of ILCs



Lymphocyte – NK cells



- compact nucleus, no cytoplasmic granules
- are identified by the presence of CD56 & CD16 and absence of CD3 and sIG
- long lived (years)
- non-phagocytic
- immunosurveillance

NK cells

- Large granular lymphocytes, components of innate immunity
- 10-15% of peripheral blood lymphocytes
- Kill virus-infected and tumor cells
- Secrete cytokines and chemokines CCL3, CCL4 & CCL5

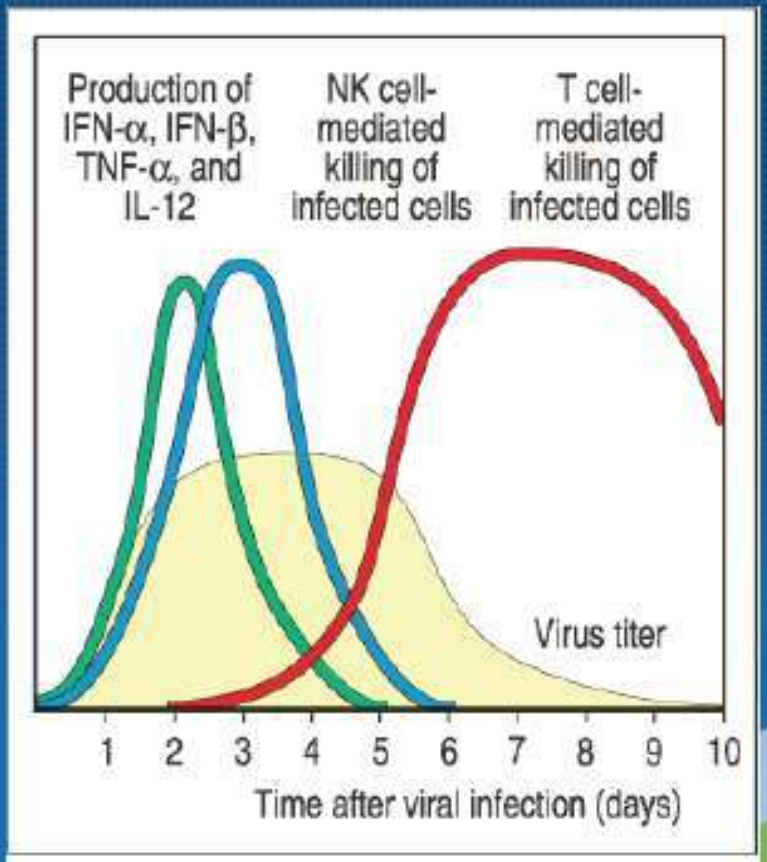
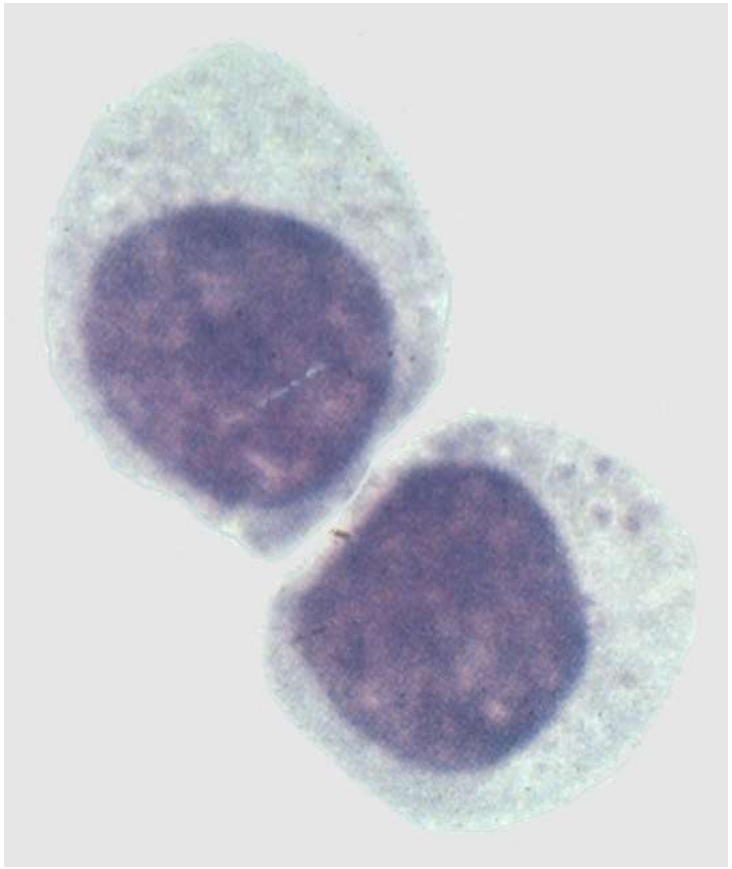


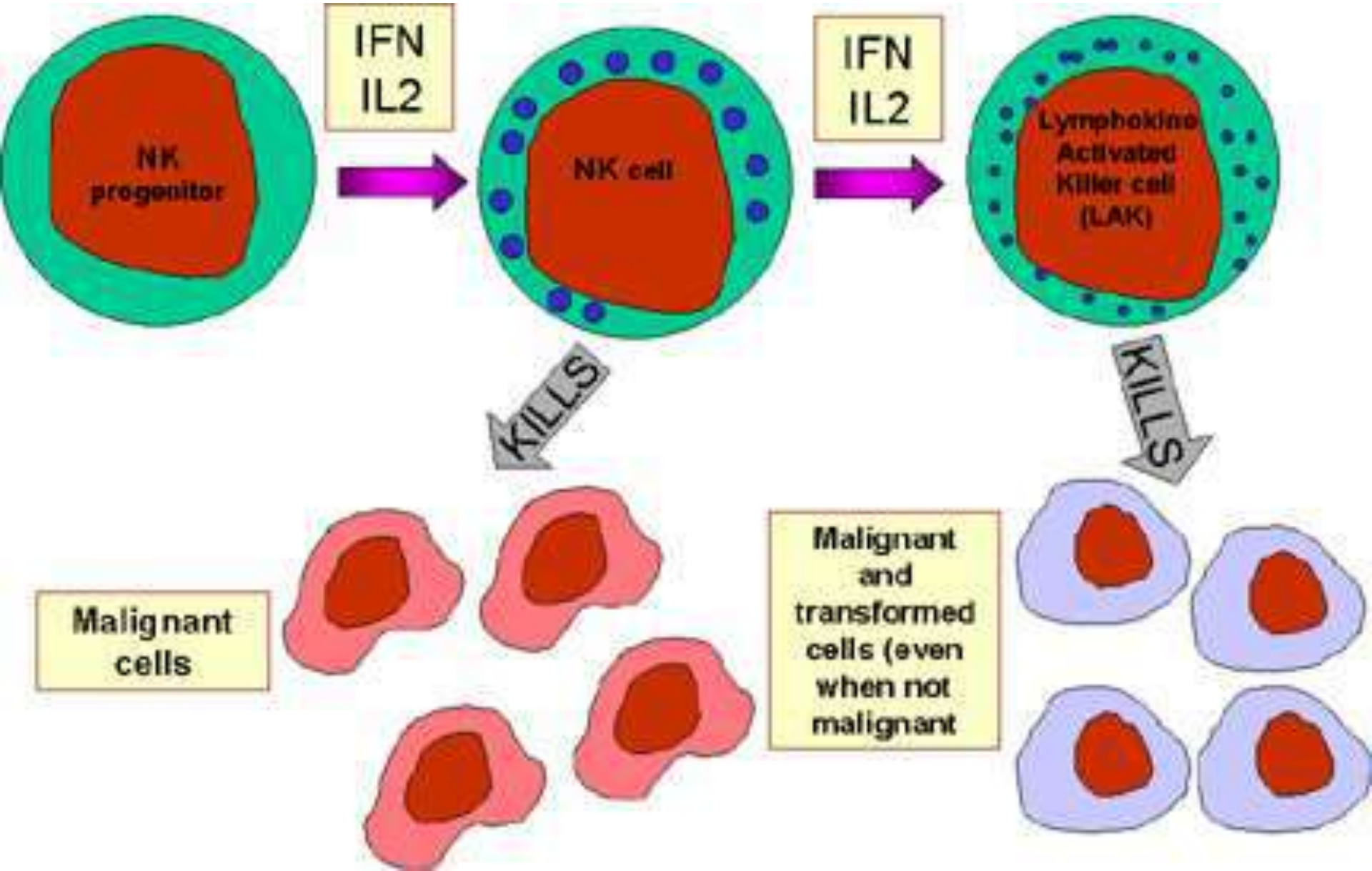
Fig 2.41 © 2001 Garland Science

NK cells

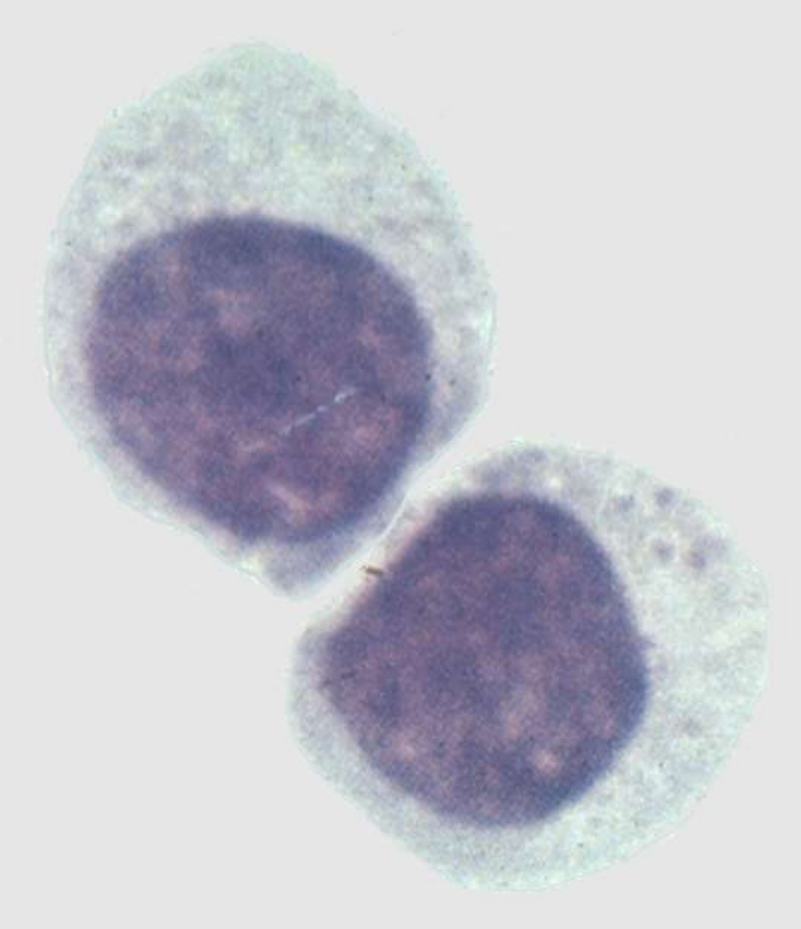


- granules contain cytolytic proteins such as perforin and granyzymes
- recognize the difference between normal and abnormal cells in a non-specific manner via sugar-lectin interaction and kill them following intimate contact
- activated by IL2 and IFN-γ to become LAK cells

Activation of NK cells



NK cells

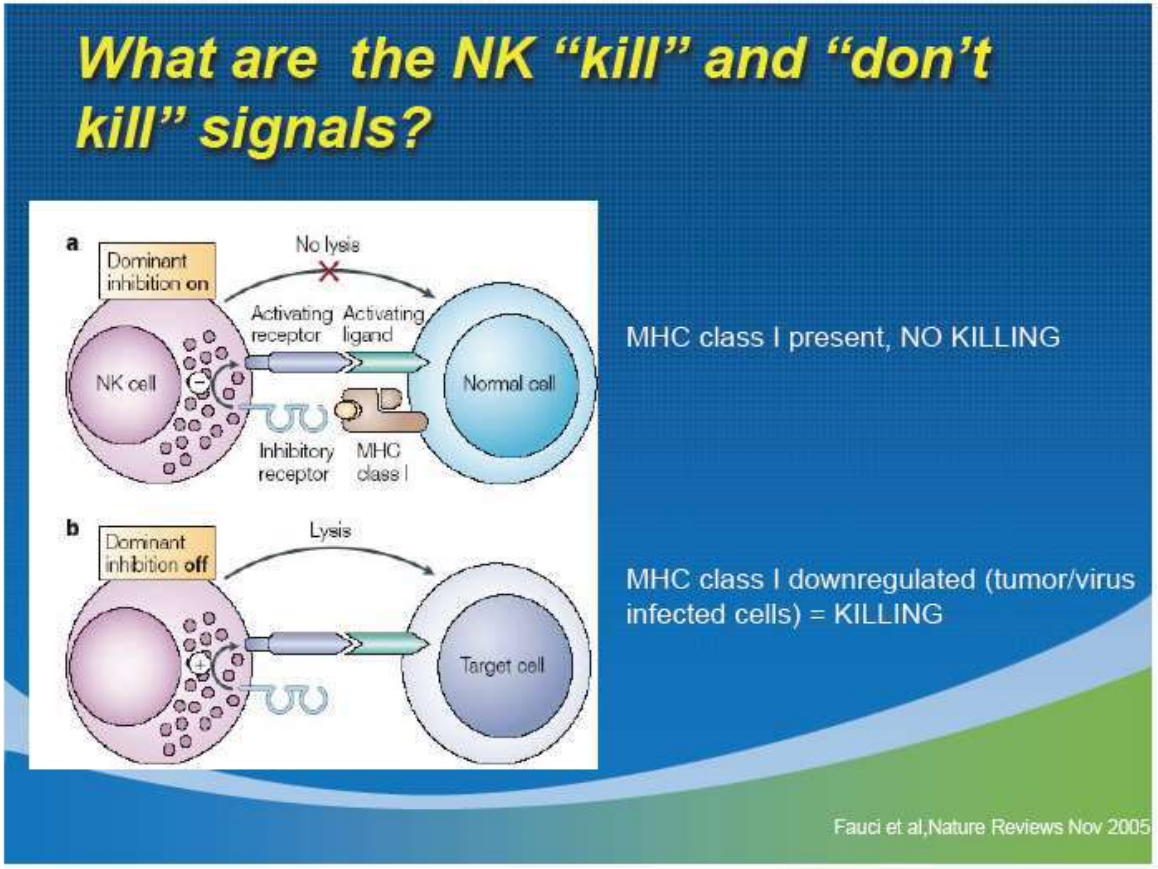


- The activating and inhibitory receptor signaling regulates the natural killer (NK) cells activation.
- After activation, NK cells secrete several cytokines such as IFN-γ, TNF-α, GM-CSF, and chemokines (CCL1, CCL2, CCL3, CCL4, CCL5, and CXCL8) that can modulate the function of other innate and adaptive immune cells.

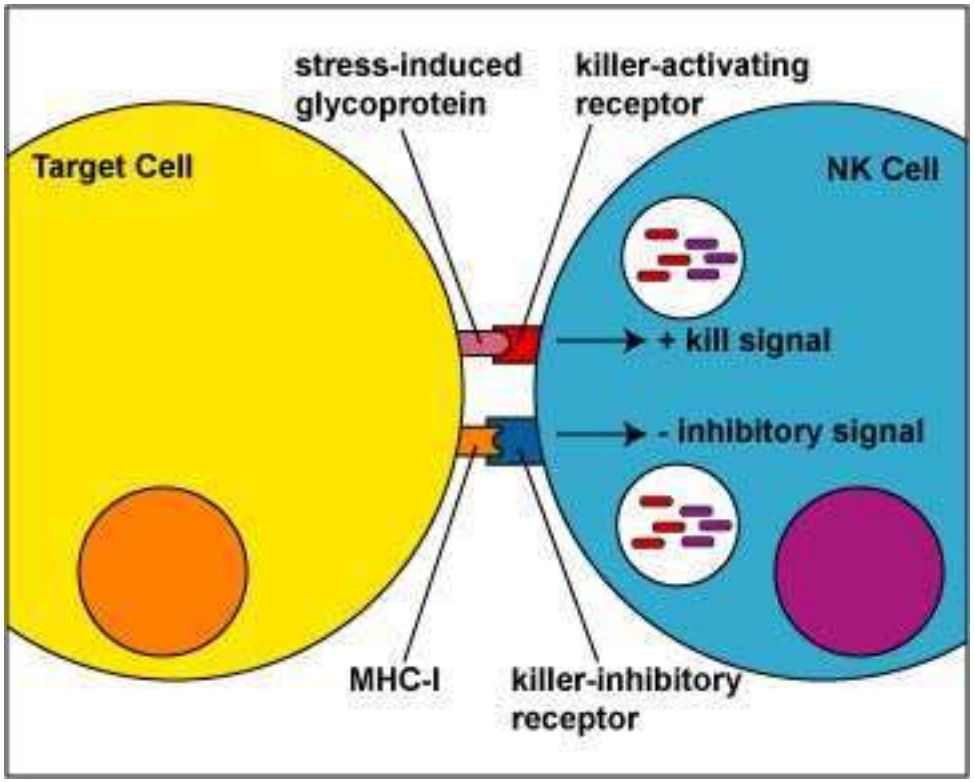
How NK cells act?

Immunosurveillance by NK cells
(an innate immunity mechanism)

NK cells have intrinsic capacity to non-speicifically recognize non- or altered-self antigens on surface of abnormal cells by KAR vs KIR pathway and subsequently destroy target cell by inducing apoptosis



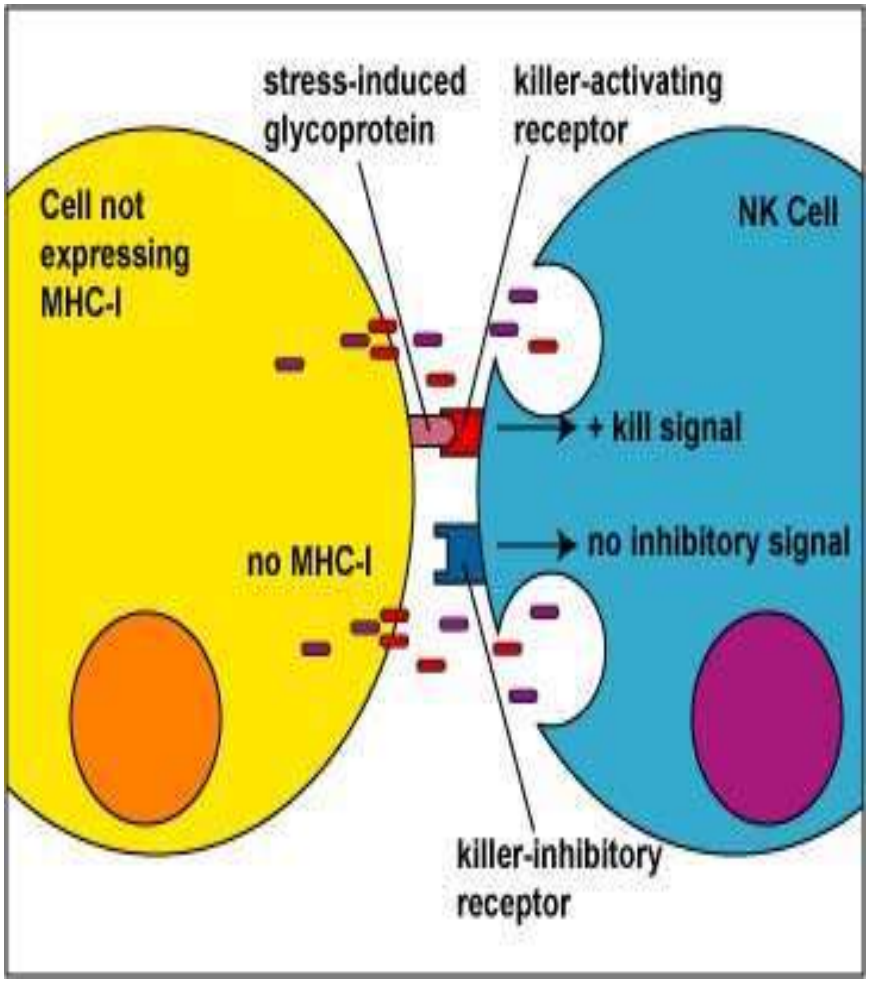
NK Cell Interacting with a Normal Body Cell



- KAR versus KIR (killer cell immunoglobulin-like receptor)
- KAR binds to ligand on a cell to activate killing (C-type lectin ligand binding)
- KIR (e.g.) binds to MHC-I molecule, if present, on same cell and inhibit killing action

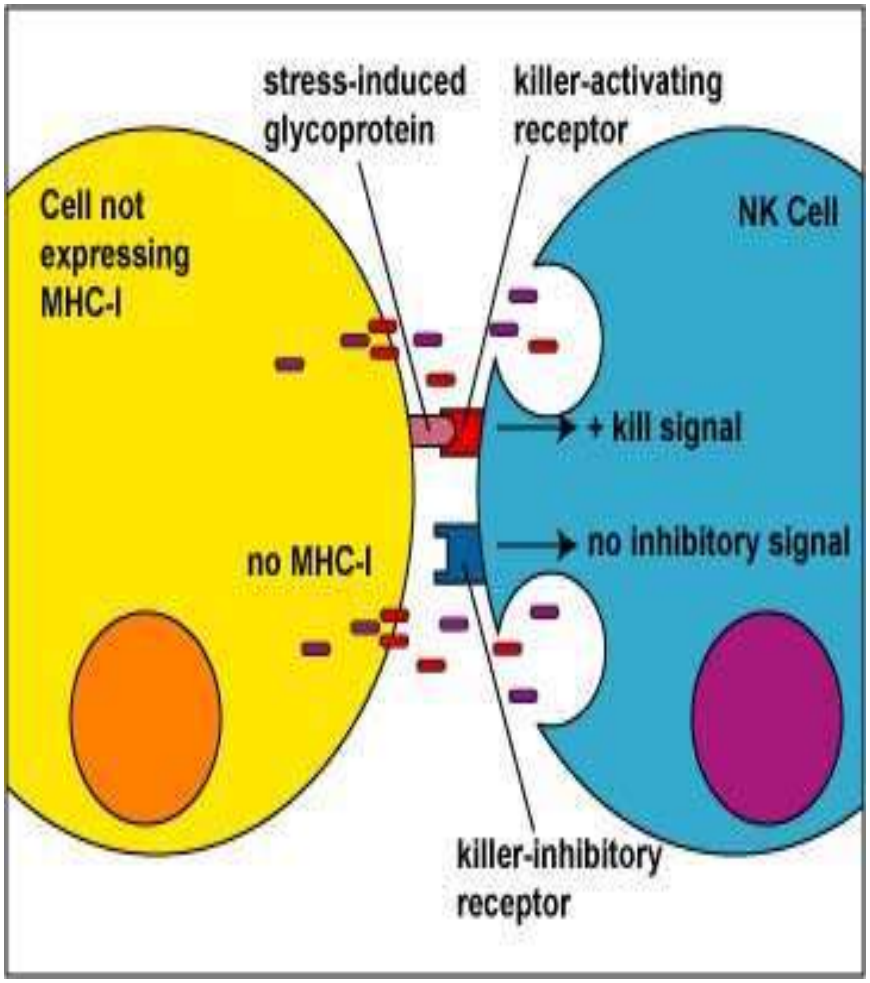
Example of KIR - NKG_2a:CD94
Example of KAR - NKG_2d:DAP10

NK Cell Interacting with a Target Cell



- Cells undergoing stress such as tumor cells lose their MHC class I molecules, a ligand for inhibitory receptors on NK cells. At the same time, they acquire stress-associated molecules which act as ligands for activating receptors. Thus, the lack of inhibitory signaling coupled with induction of activating signaling shifts the balance toward NK cell activation, leading to secretion of cytokines and killing of tumor cells.

NK Cell Interacting with a Target Cell

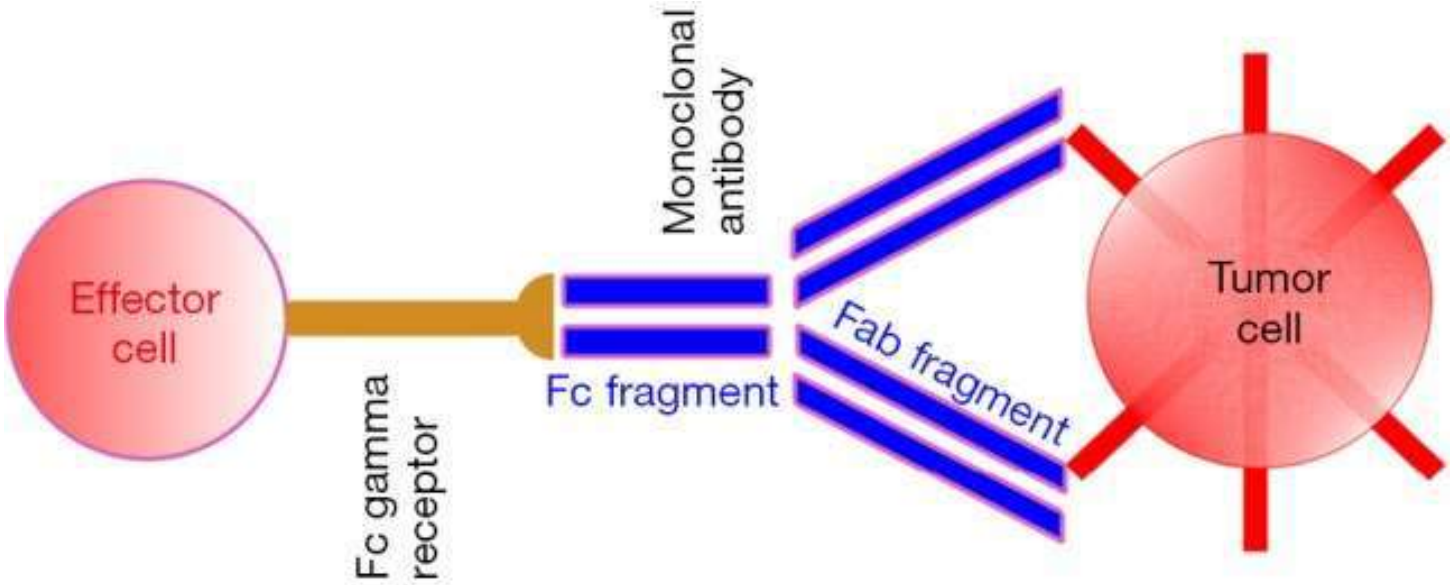


- Down regulation of MHC-I molecules on target cell (virus infected or cancer cell)
- KIR not activated
- KAR activated following binding with stress induced glycoproteins (danger signals)
- NK cell releases pore-forming proteins called perforins, proteolytic enzymes called granzymes, and chemokines

Antibody dependent cell cytotoxicity (ADCC) (an adaptive immunity mechanism of NK cells)

- **ADCC is a mechanism where effector cells (NK cells in this case) secrete cytotoxic molecules and lyse antibody-coated target cells.**
- ADCC depends on the bifunctional structure of IgG molecules.
- The fragment antigen-binding (Fab) of the IgG molecule bind to its specific viral or TAA associated on the surface tumor or the target cell.
- The fragment Fc (stem portion) of IgG bind with FcγRIII (CD16) present on surface of NK cell.
- On engagement of both, cell antigen and an activating FcγR, by the Fab and Fc portions of the Ab respectively, ADCC is initiated, since this creates a bridge from the tumor/target cell to the effector NK cell.
- The recognition of target cells is then combined to a lytic attack on the target cell mounted by effector cells.
- ADCC does not depend on the immune complement system in which targets are also lysed but no other cell is required.
- ADCC requires an effector cell, mainly NK cells (due to the lack of inhibitory FcγR IIb), that typically interact with IgG antibodies

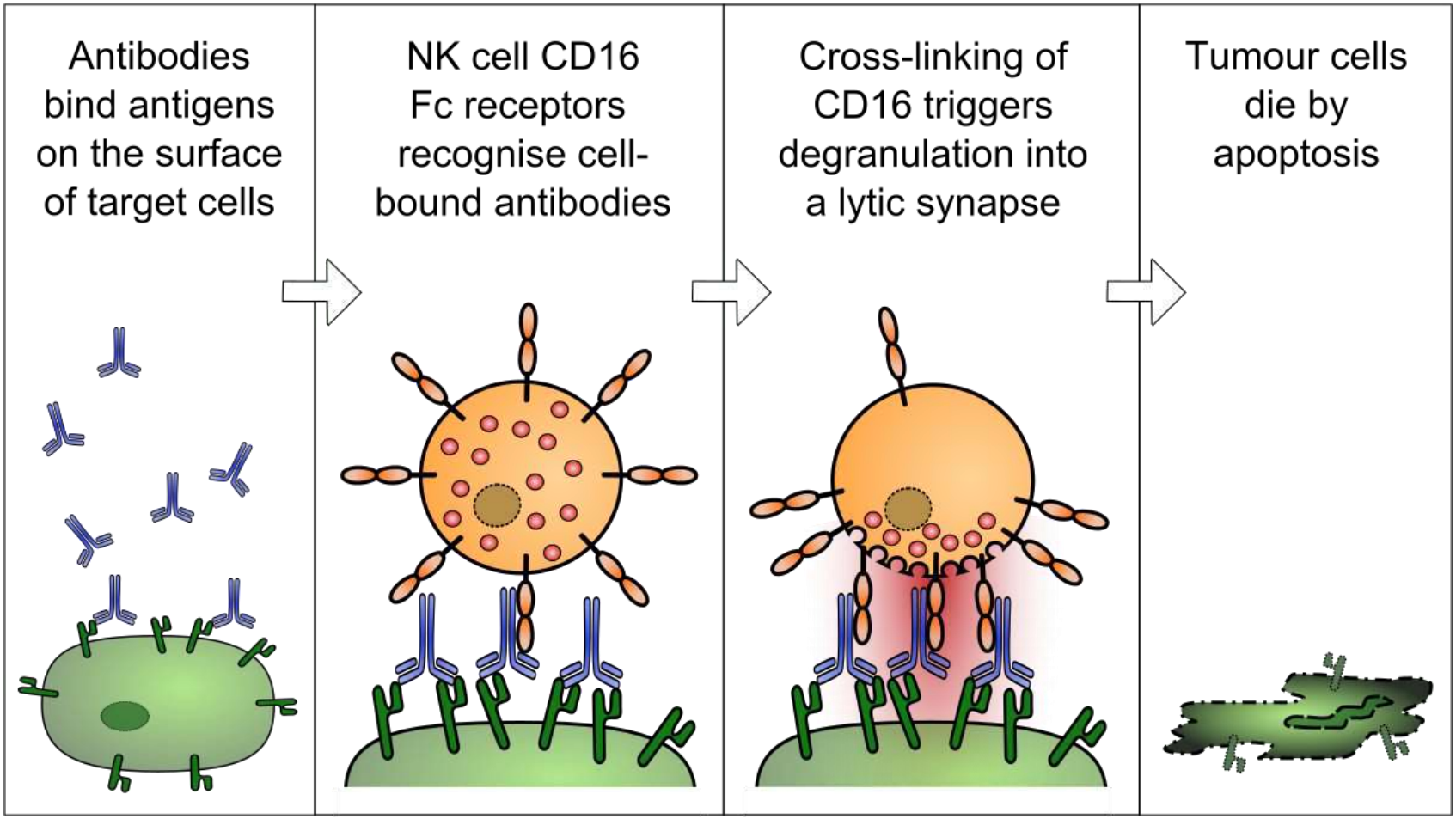
ADCC


Monoclonal antibody
Effector cell
Fc gamma receptor
Fc fragment
Fab fragment
Tumor cell

Antibody dependent cell cytotoxicity (ADCC)
(an adaptive immunity mechanism of NK cells)

- Antibody-dependent tumor killing through NK mediation takes place via various routes:
  - cytotoxic granule exocytosis releasing granzymes and perforins,
  - tumor necrosis factor (TNF) family death receptor signalling, and
  - release of a pro-inflammatory cytokine, such as interferon-gamma (IFNγ), which activate nearby immune cells to encourage antigen presentation and adaptive immune responses
- Macrophages, neutrophils and eosinophils can also mediate ADCC, such as eosinophils killing certain parasitic worms known as helminths via IgE antibodies.

ADCC

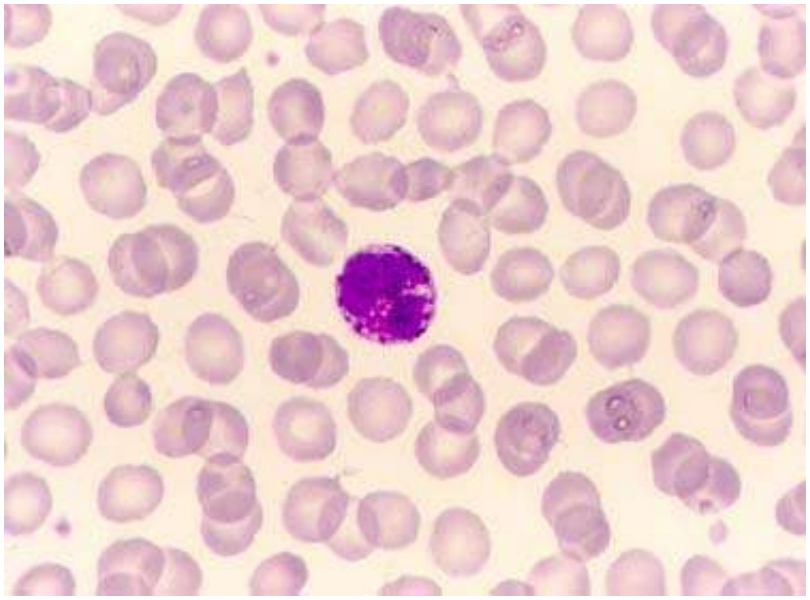

Antibodies bind antigens on the surface of target cells
NK cell CD16 Fc receptors recognise cell-bound antibodies
Cross-linking of CD16 triggers degranulation into a lytic synapse
Tumour cells die by apoptosis

Innate defense Cells in the Tissue

These include:

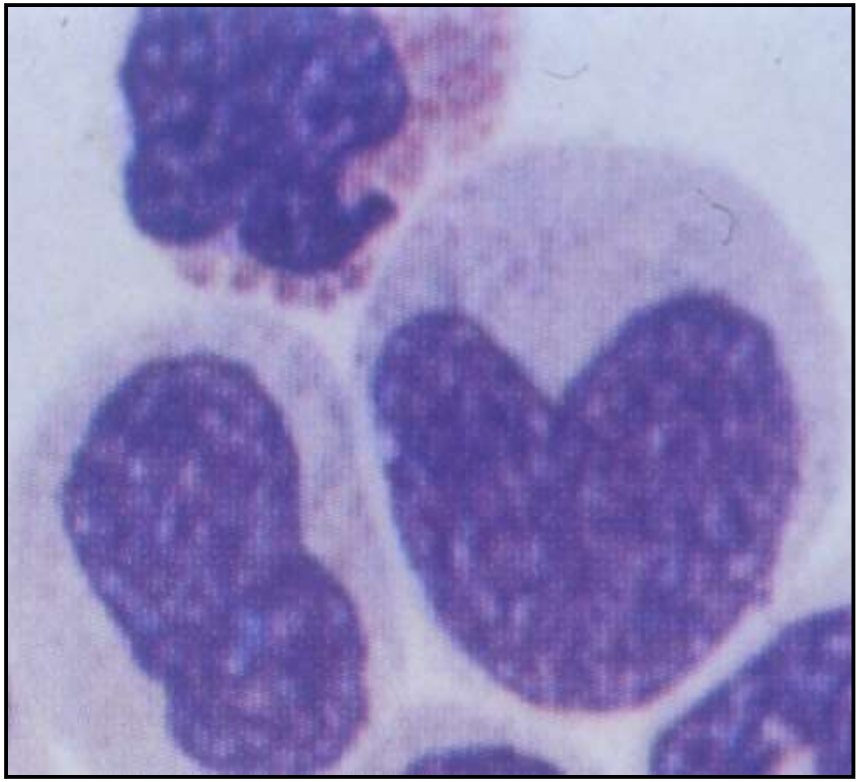
- Mast Cells
- Macrophages (cells of mononuclear macrophage system)
- Dendritic Cells

Mast Cells



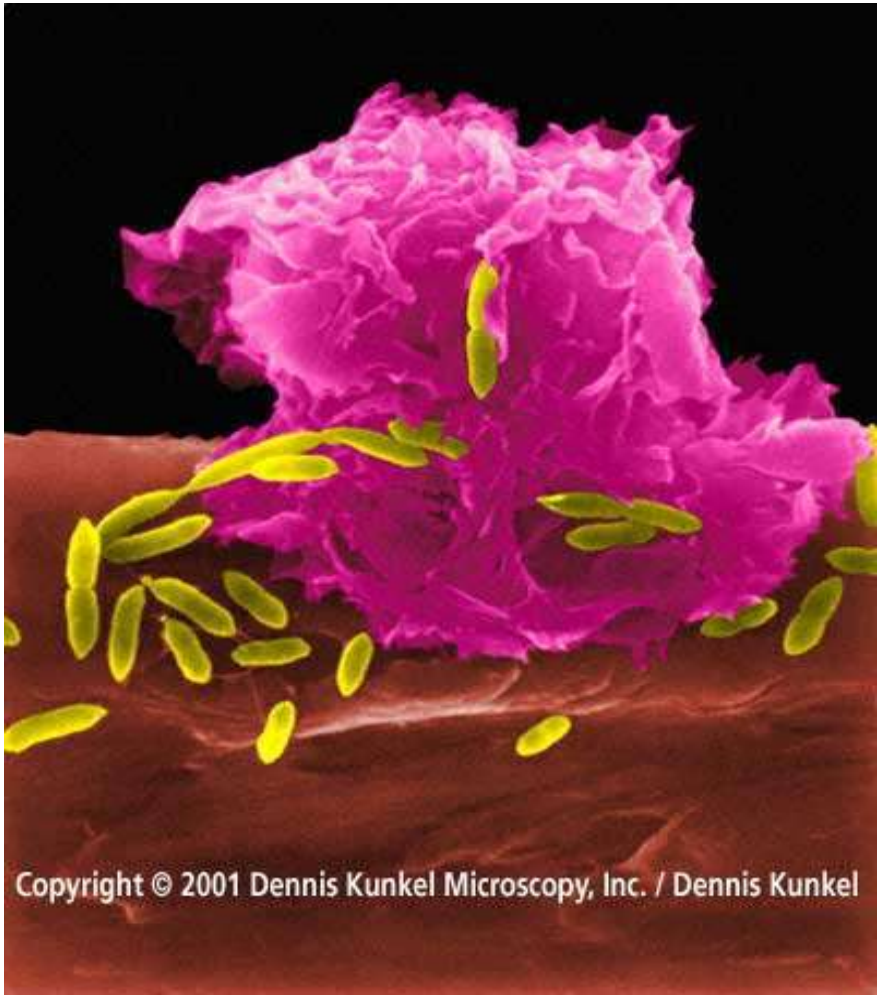
- **CD markers** include the high-affinity IgE receptor, CD117, and CD203c
- characteristic lobed nucleus, cytoplasmic granules
- release histamine , leukotrienes, and prostaglandins (vasoactive amines)
- non-phagocytic, inflammation, and coagulation, recruitment of cells of immunity
- involved in hypersensitivity reactions.

Macrophages



- monocytes in blood, cells of RES in tissues
- characteristic nucleus
- CD68 membrane marker.
- activated by cytokines
- phagocytosis, **“call for help”** by secreting cytokines, tissue repair

Electron Micrograph of a Macrophage Phagocytosing *E. coli*



Dendritic cells

- have numerous cytoplasmic processes giving ‘tree-like’ or dendritic shapes
- function as the ‘sentinels’ of the immune system
- found in every non-lymphoid tissue, example Langerhan’s cells
- induce pro-inflammatory response and activate innate lymphocytes such as natural killer (NK) cells, NK T cells, and γδ T cells.
- act as bridge between innate and adaptive immunity