I FAGOCITI. FENOTIPO E FUNZIONI

Prof Giovanni Bernardini 15-10-2024

Outline of the lesson

What are phagocytes?

What is the origin of phagocytes?

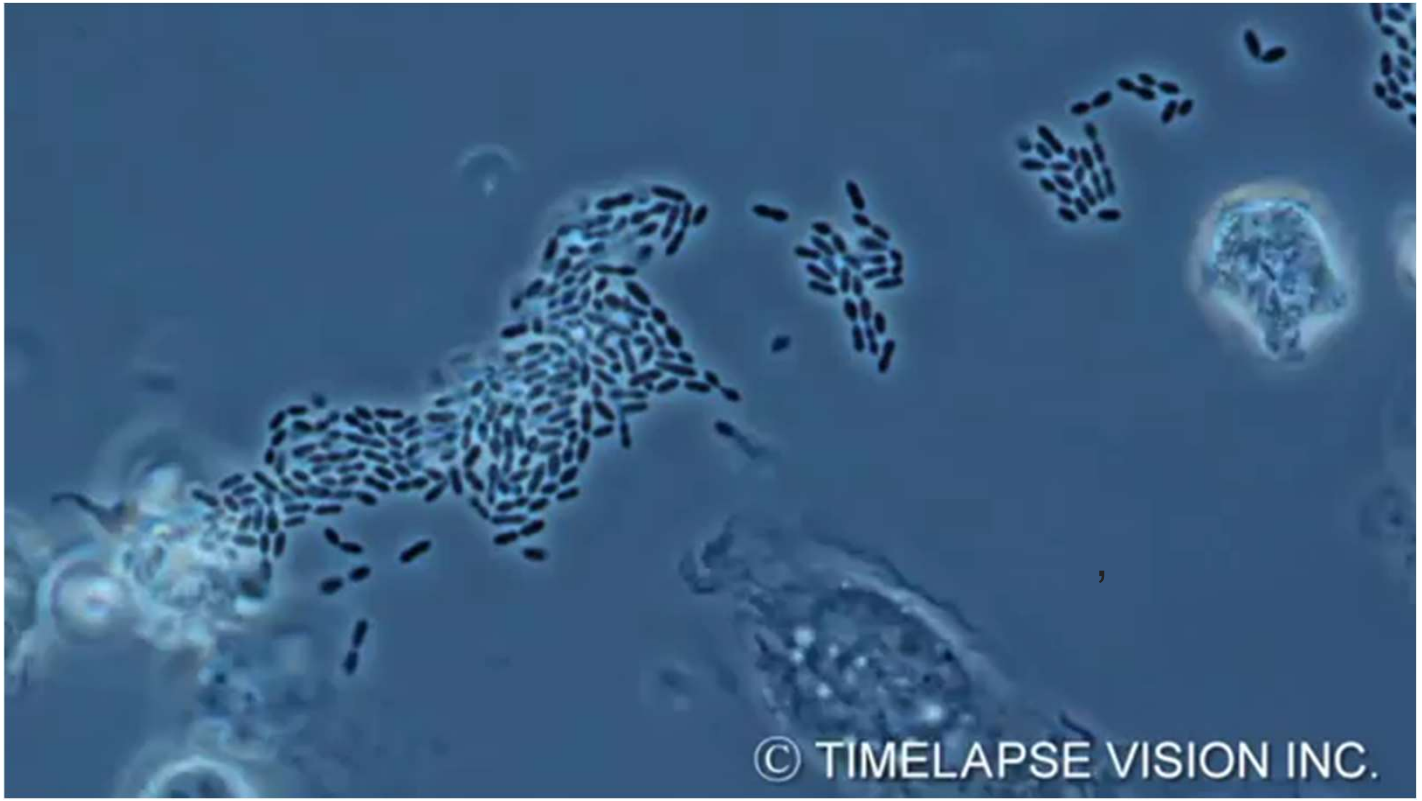
Phagocyte heterogeneity. Diversification of macrophages into subpopulations.

Phagocytosis, what else? Functions of phagocytes.

Phagocytes are cells able to perform phagocytosis

Phagocytosis (from Ancient Greek «*phago*» , **meaning** 'to eat', and «*kytos*» , **meaning** 'cell') Greek *–ōsis* meaning a process (often associated to a disease)

In summary phagocytosis is the cell uptake of particulate material (leading to cell disruption)



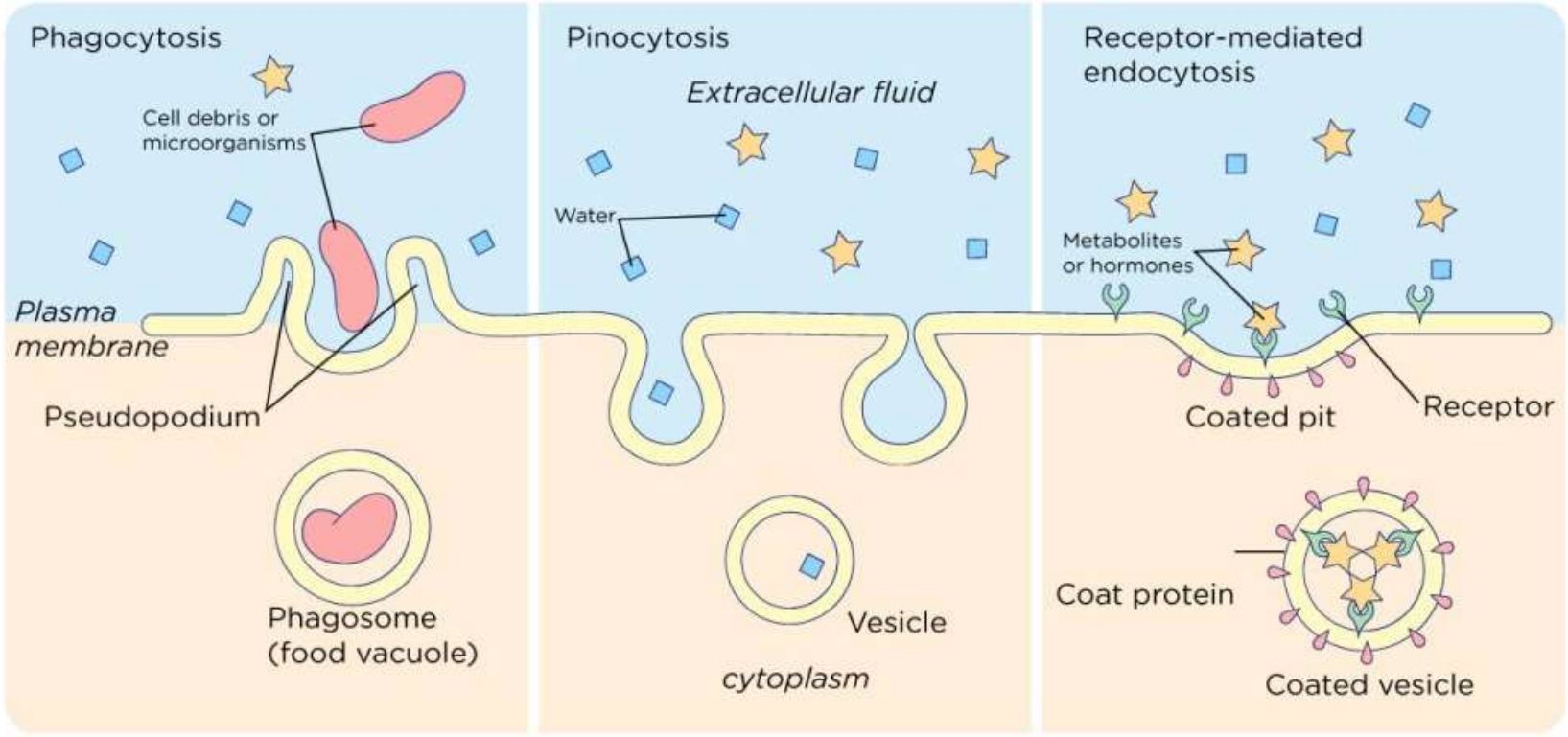
Phagocytosis

It is central to innate and adaptive immunity against pathogens.

Cell death by phagocytosis mediates physiological turnover of erythrocytes and of leucocytes.

Overall, it is the most abundant form of cell death in the mammalian body

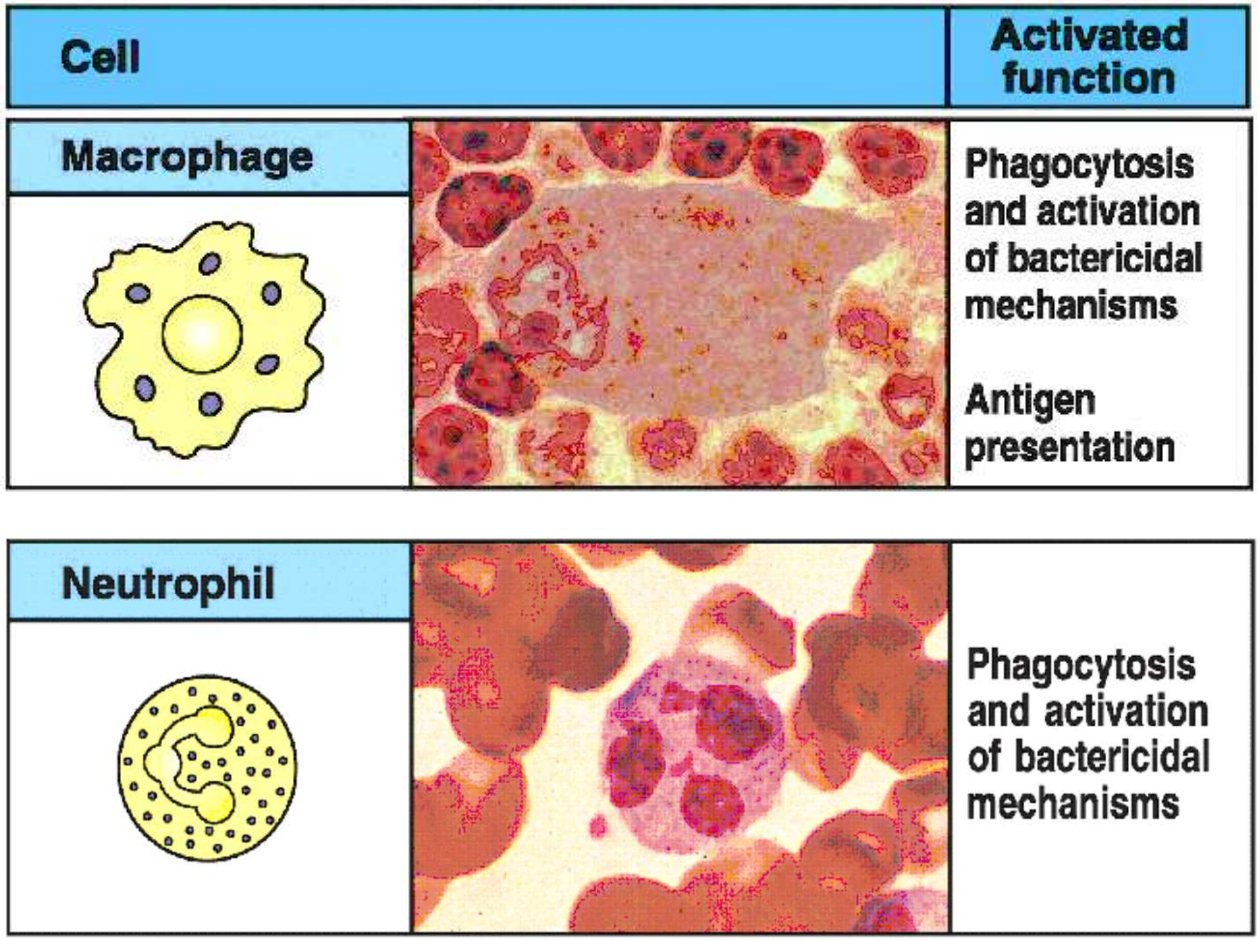
Phagocytosis is a type of endocytosis



Phagocytosis is a type of receptor mediated endocytosis

L'INGLOBAMENTO E LA DISTRUZIONE DEI MICROBI E' MEDIATA DA FAGOCITI

Macrofagi, neutrofili e cellule dendritiche effettuano una efficace fagocitosi e per questo sono chiamati *fagociti professionisti*

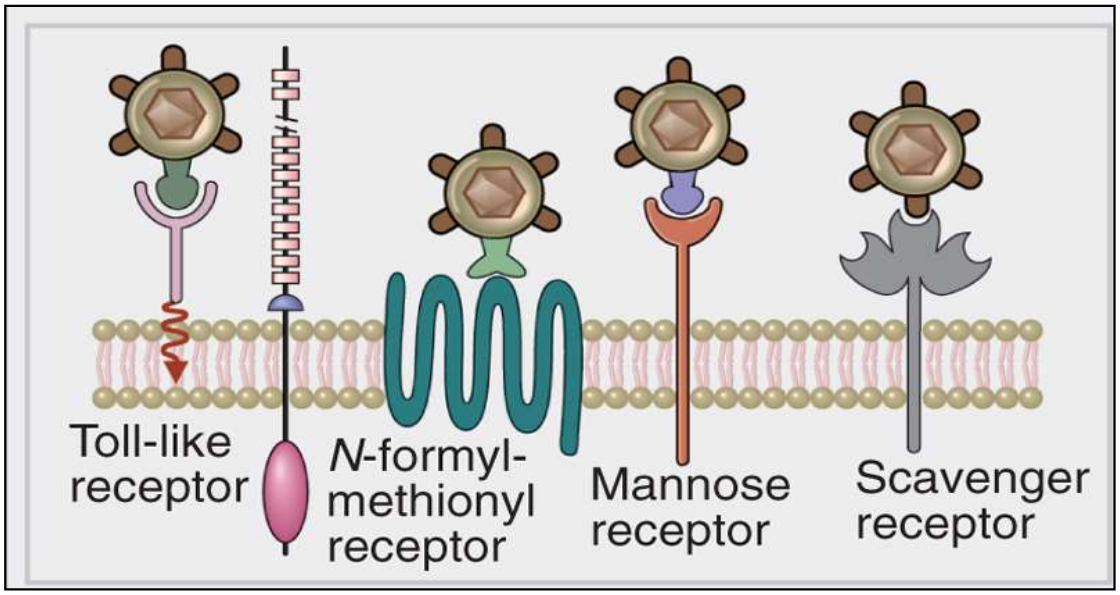


I macrofagi riconoscono il non-self (PAMPs) o il self modificato (DAMPs) ed eseguono una risposta effettrice (tramite fagocitosi, o secrezione di citochine per partecipare alla **funzione 1**). … ma nel frattempo interagiscono con linfociti T per promuovere la loro attivazione **(funzione 2)**.

I neutrofili svolgono prevalentemente funzioni microbicide (fagocitosi, rilascio di granuli contenenti enzimi litici e nettosi)

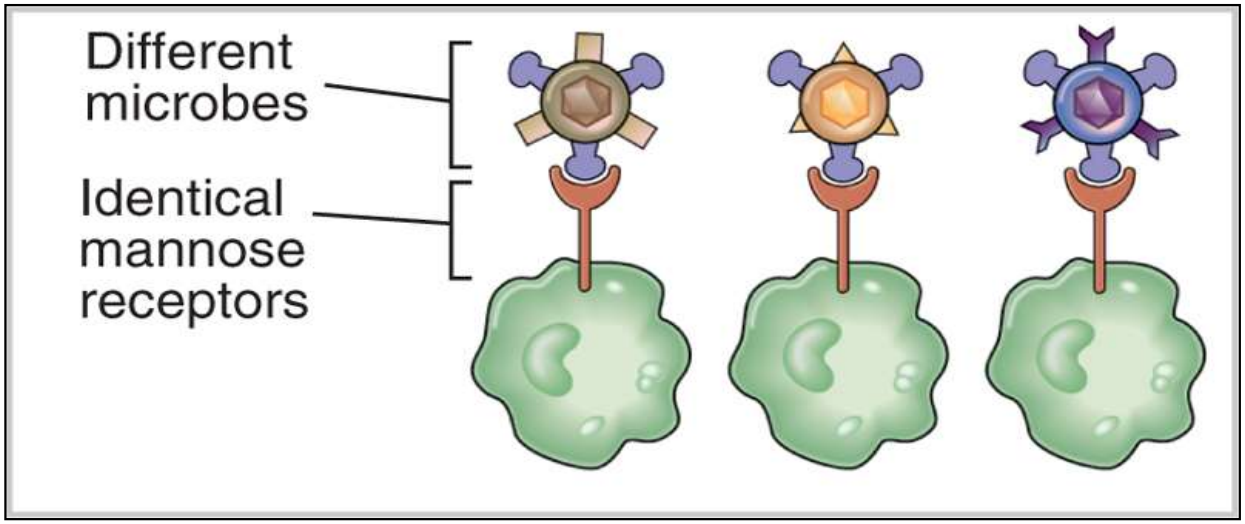
Che tipo di recettori possono mediare la fagocitosi?

Pathogen recognition receptors (PRR)



Recognition is mediated by specific invariant receptors for pathogen molecular structures which are not expressed by mammal cells ("molecular patterns").
Each cell type expresses several receptor types and is thus able to recognize different molecular patterns.

In addition different microorganisms share same molecular patterns and can thus be recognized by the same receptor type.



CRD: Carbohydrate recognition domain

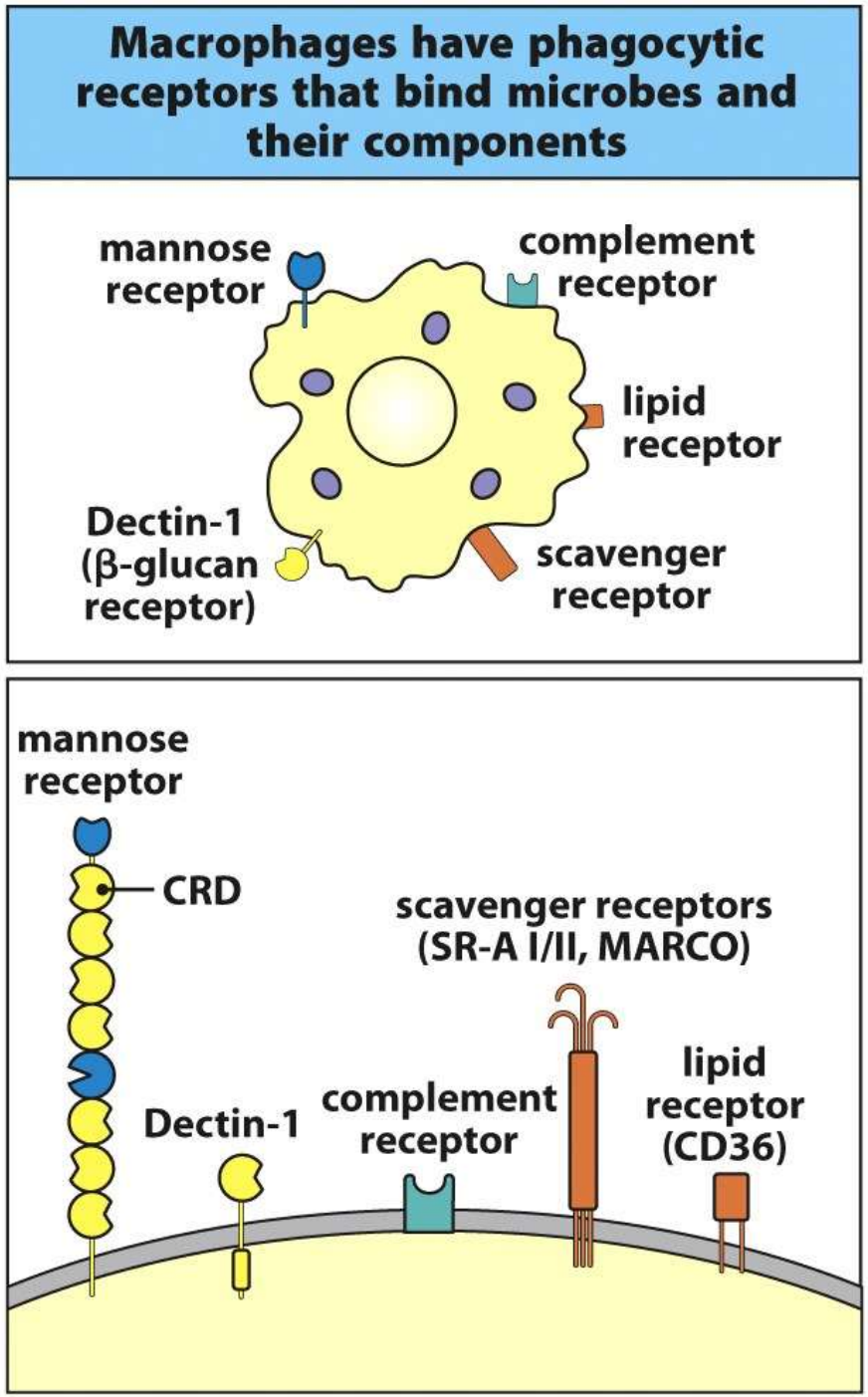


Figure 3.2 part 1 of 2 Janeway's Immunobiology, 8ed. (© Garland Science 2012)

Phagocytes take part to the innate immune response

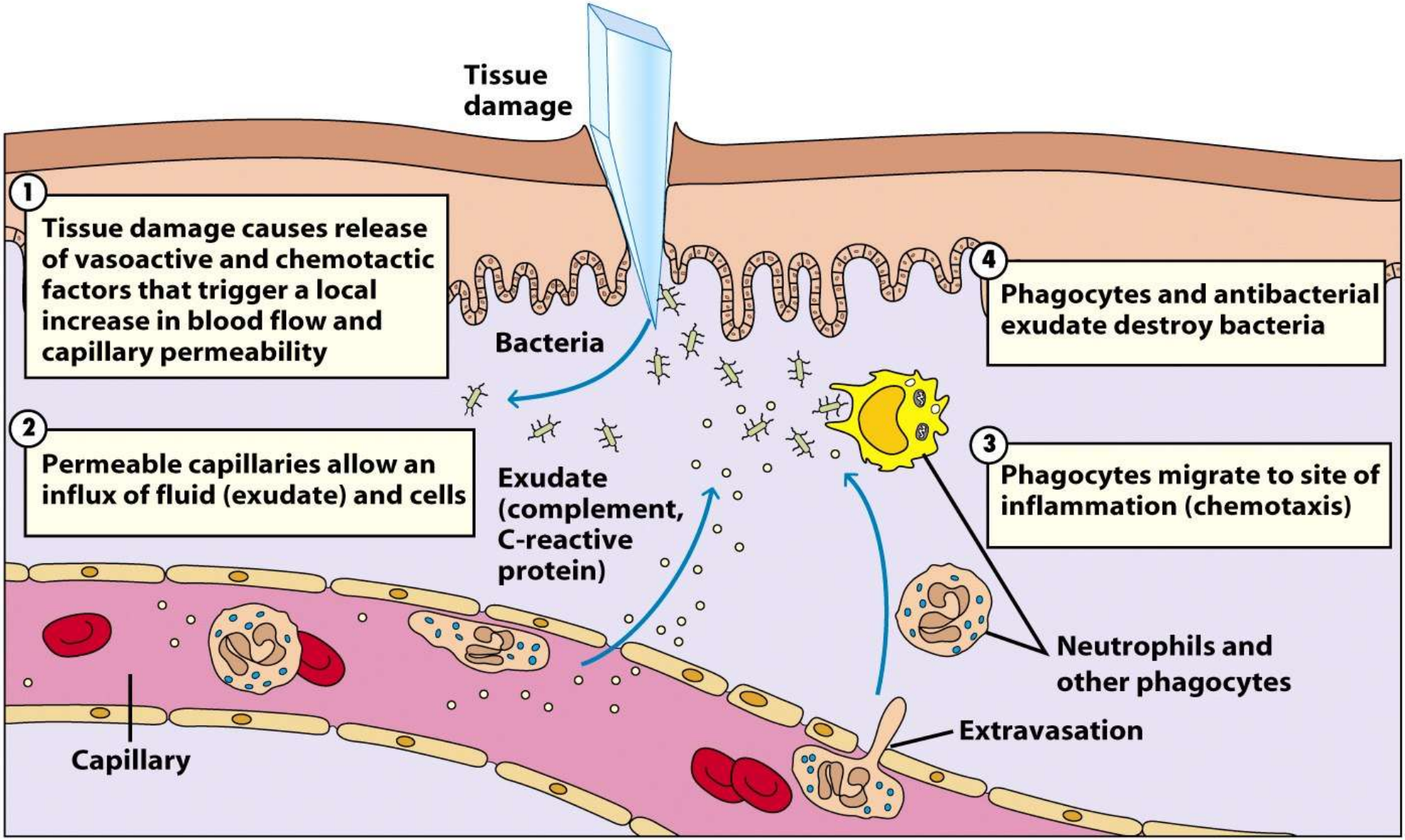


Figure 3-5
Kuby IMMUNOLOGY, Sixth Edition

Phagocytes take part to the innate immune response

- Neutrophils are the most abundant population among the circulating white blood cells.
- Neutrophils are short-lived cells (1-2 days) and they die immediately after completing a phagocytosis cycle.
- Dead neutrophils are the main components of pus formed in certain infections caused by extracellular bacteria called pyogenic bacteria (or pus generators).
- Macrophages, on the contrary, are tissue cells which have a long life and the capacity to continuously generate new lysosomes.

Phagocyte-mediated immune response is fundamental for protection

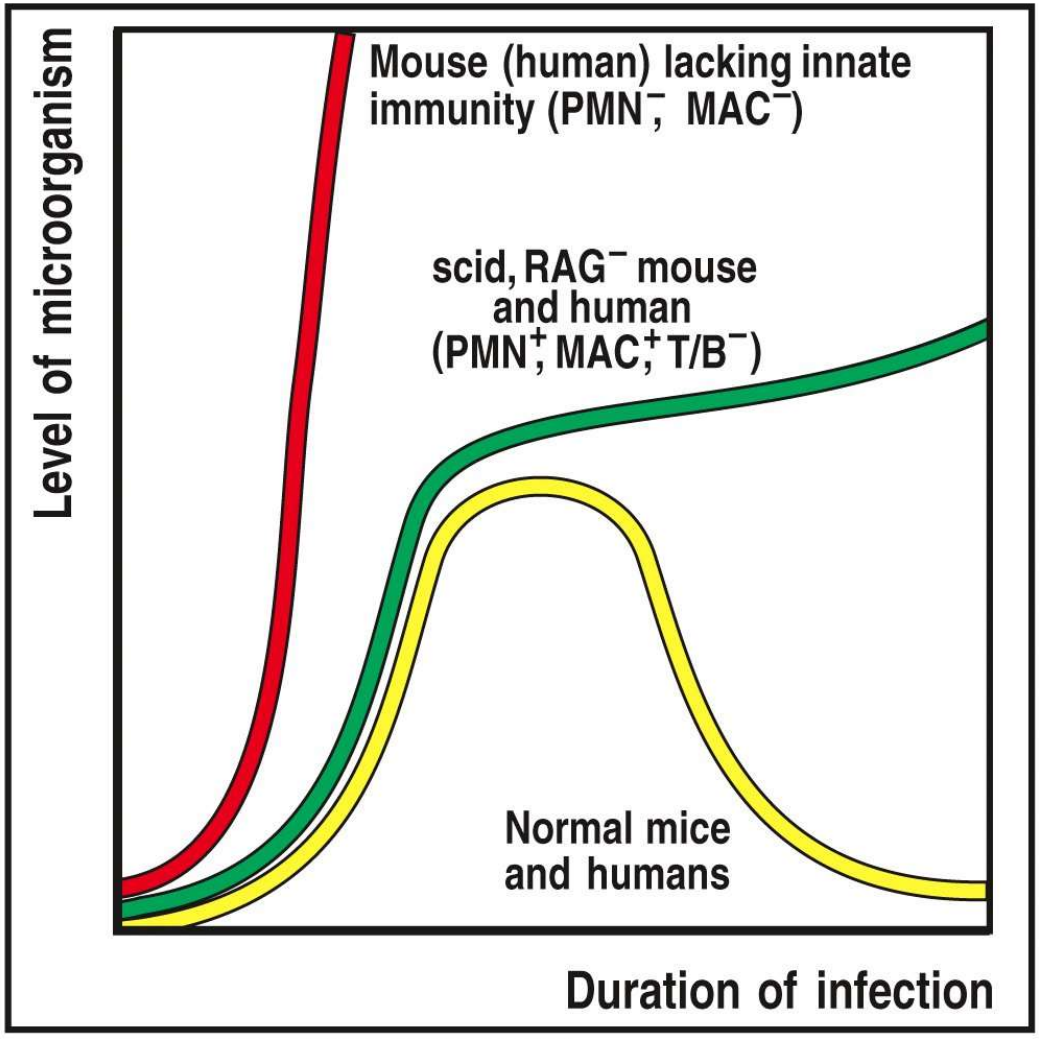


Figure 10-6 Immunobiology, 6/e. (© Garland Science 2005)

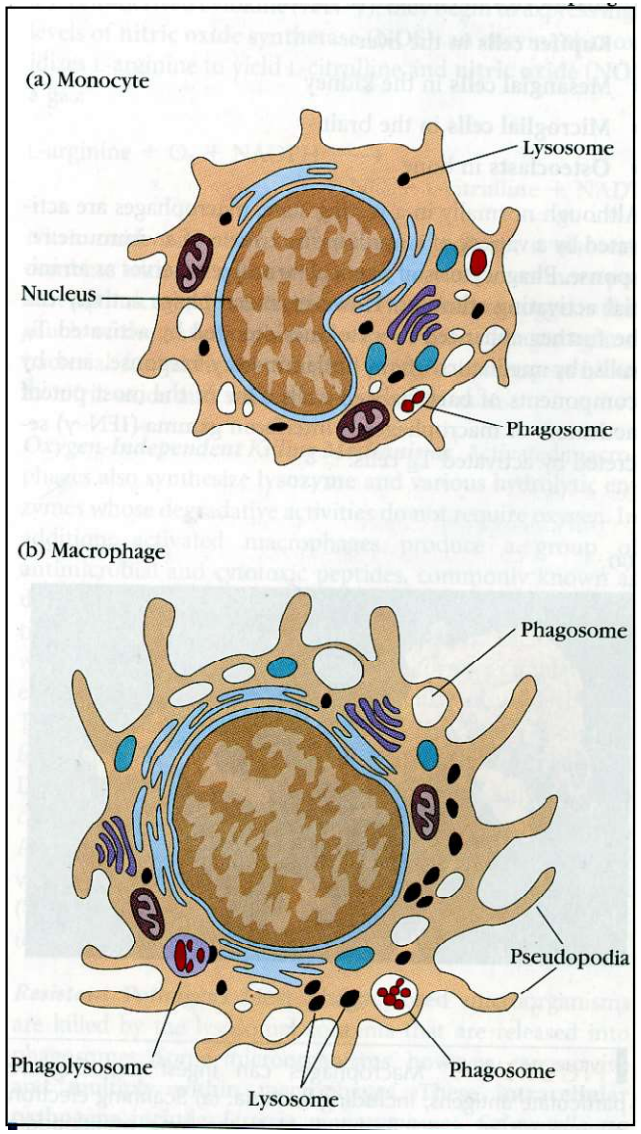
PMN: polymorphonuclear cells; MAC: Macrophages; T/B: T and B lymphocytes

ORIGINE DEI MACROFAGI e NEUTROFILI

Macrophages differentiate from circulating monocytes when these cells enter the tissues

Monocyte-macrophage differentiation:

- Increase in size of 5-10 times
- Increase in the number of organelles (lysosomes)
- Increased phagocytic capacity



ORIGINE DEI MACROFAGI
contributo dei monociti circolanti alla generazione di due tipi di macrofagi

MONOCITI INFIAMMATORI (MACROFAGI INFIAMMATORI O IMMUNOLOGICAMENTE STIMOLATI)

I monociti richiamati dal sangue durante la risposta infiammatoria differenziano in macrofagi dotati di proprietà fagocitiche citotossiche e capacità proinfiammatoria e di presentazione dell'antigene

MONOCITI RESIDENTI (MACROFAGI RESIDENTI)

Hanno una funzione di sentinella e clearence e così facendo contribuiscono a mantenere le funzioni tissutali

Iniziano la risposta infiammatoria acuta e il rimodellamento vascolare

Importante: Per i neutrofili abbiamo solo la controparte circolante che, come detto, ha vita breve e non diventa residente una volta entrata in tessuti

Origine dei macrofagi:
i precursori dei macrofagi residenti e infiammatori possono essere identificati nel sangue

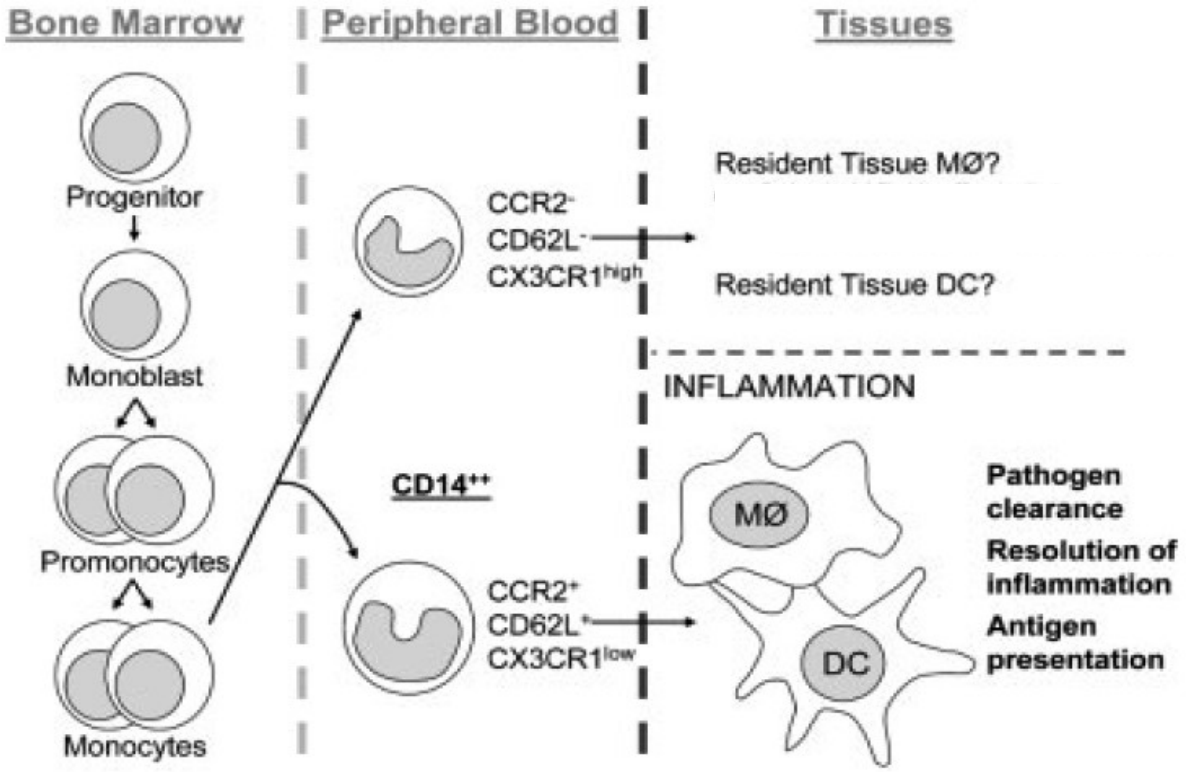
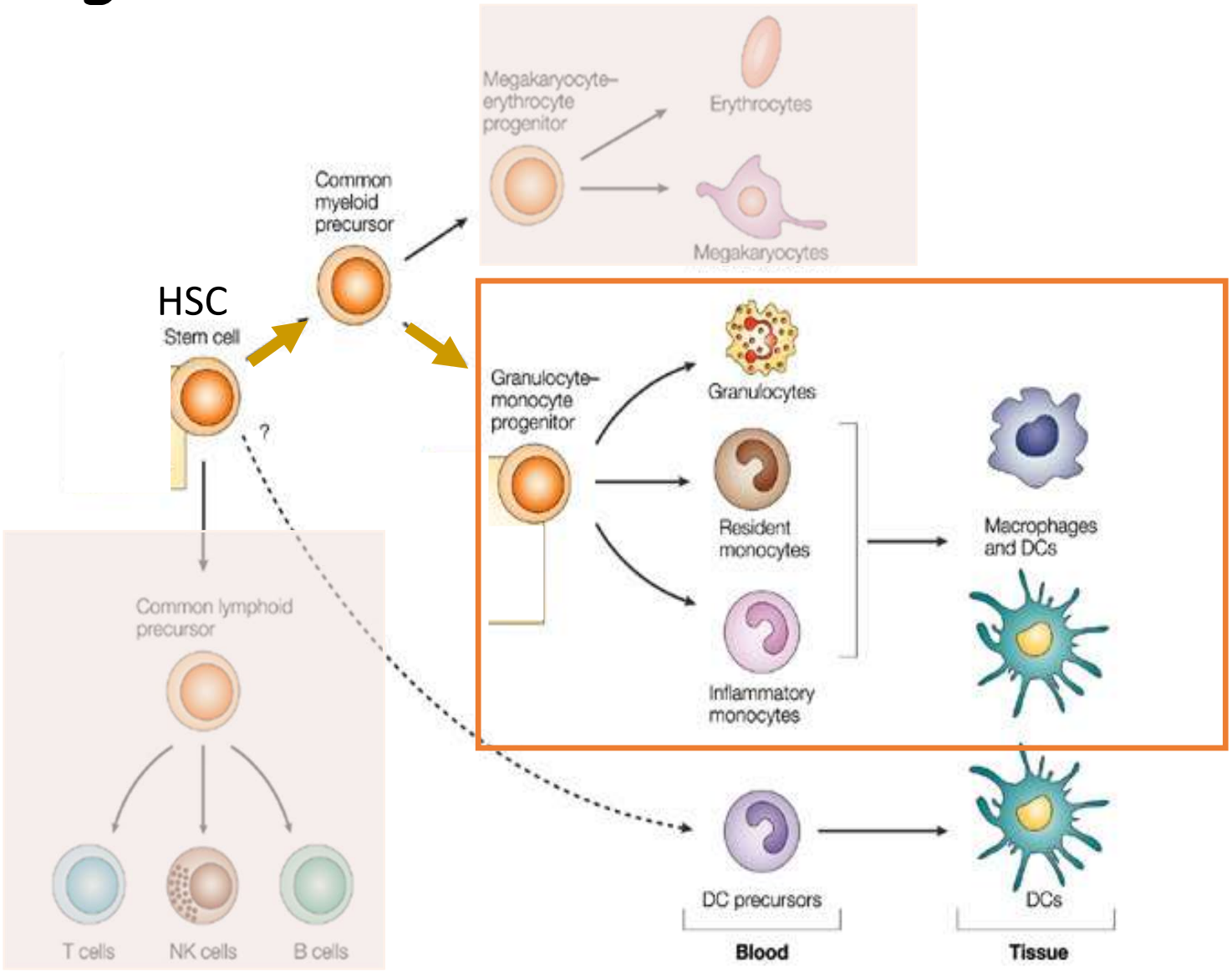


Figure 3. Differentiation and distribution of mononuclear phagocytes. Distinct subpopulations of circulating monocytes are thought to give rise to resident tissue macrophages, DC and osteoclasts compared with cells recruited by an inflammatory or immunologic stimulus. Further phenotypic heterogeneity

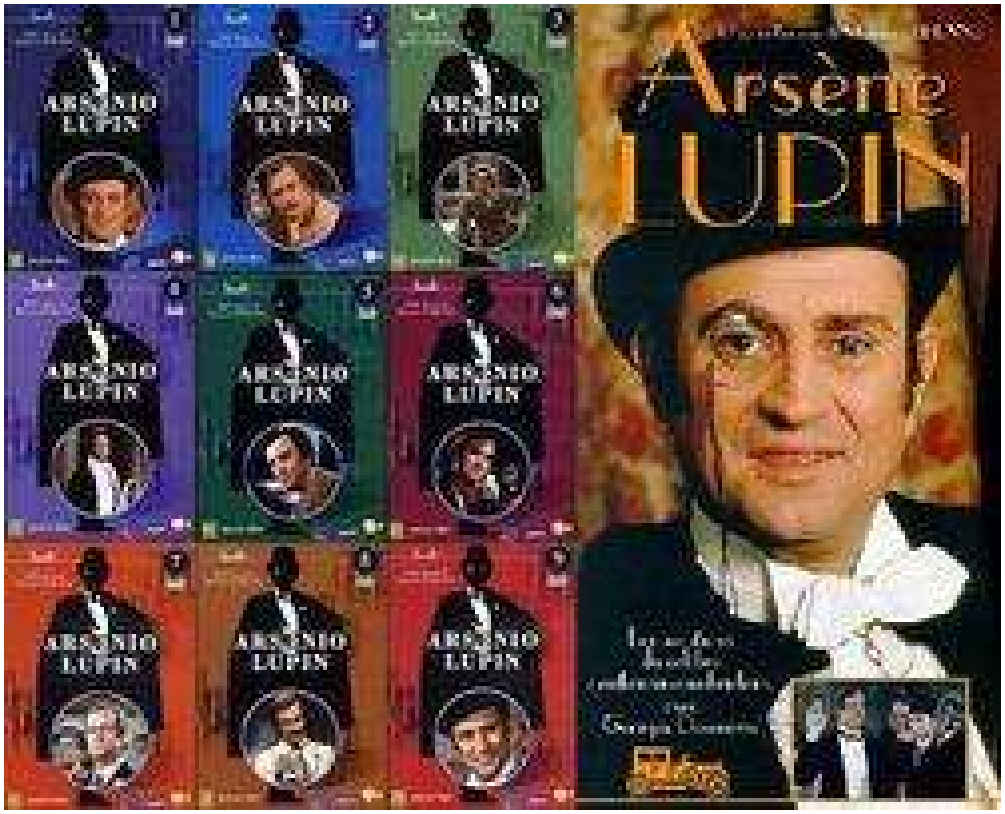
NELL'UOMO I MONOCITI INFIAMMATORI SONO CD14+CCR2+ E I MONOCITI RESIDENTI SONO CD16+CX3CR1+

After birth, myeloid phagocytes are generated in the bone marrow



Phagocytes are cells of the hematopoietic system

Grazie alla loro versatilità (plasticità), i macrofagi sono abili trasformisti …come Arsenio lupin



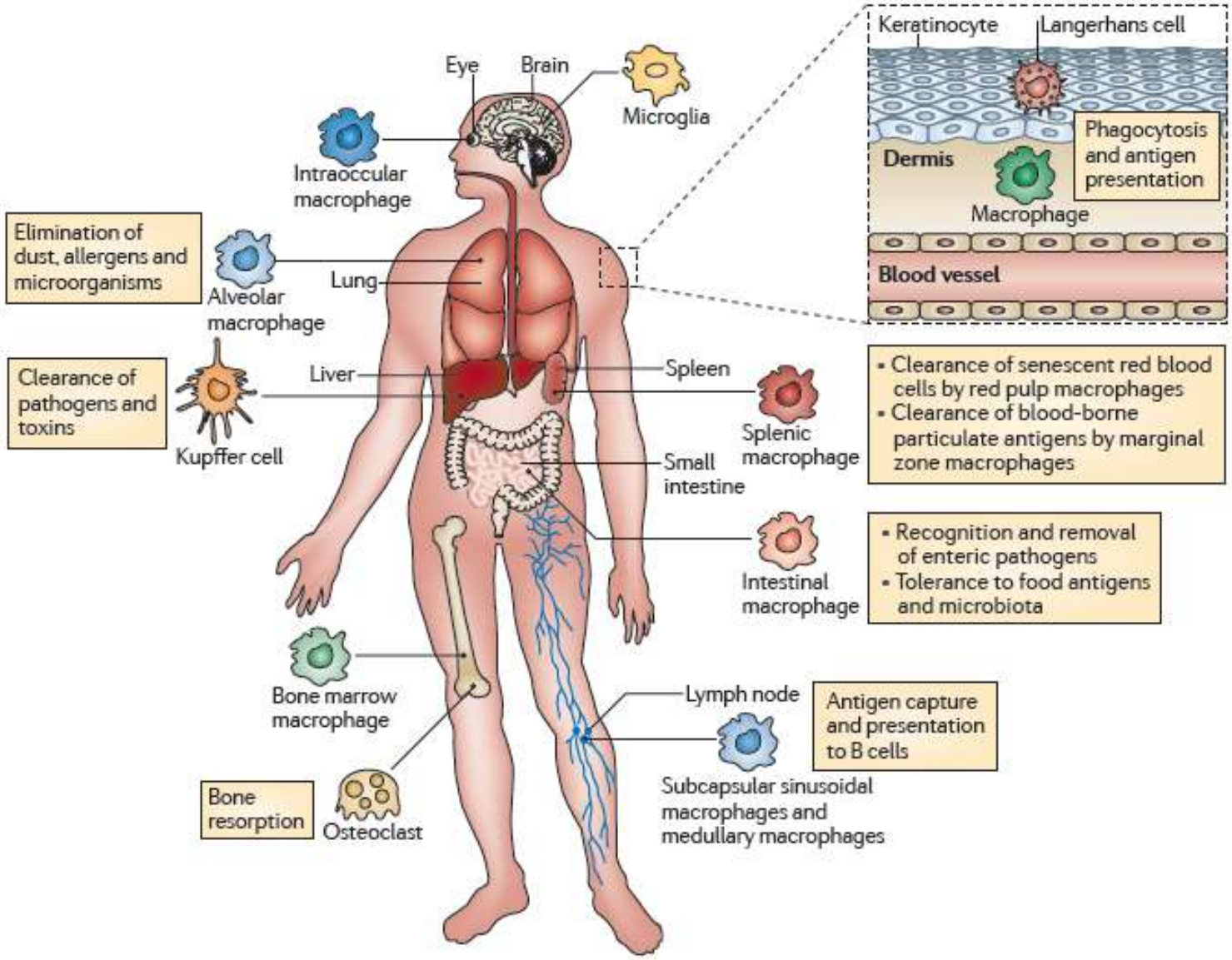
I macrofagi si trovano in quasi tutti i tessuti come cellule mobili per perlustrare l'ambiente ma anche immobili e svolgono una serie di funzioni degradative e difensive «housekeeping»,

I monociti differenziano in macrofagi con **caratteristiche funzionali differenti a seconda dell'ambiente in cui si trovano**.

Vedremo poi che i macrofagi stessi possono acquisire un comportamento pro- o anti-infiammatorio se opportunamente stimolati

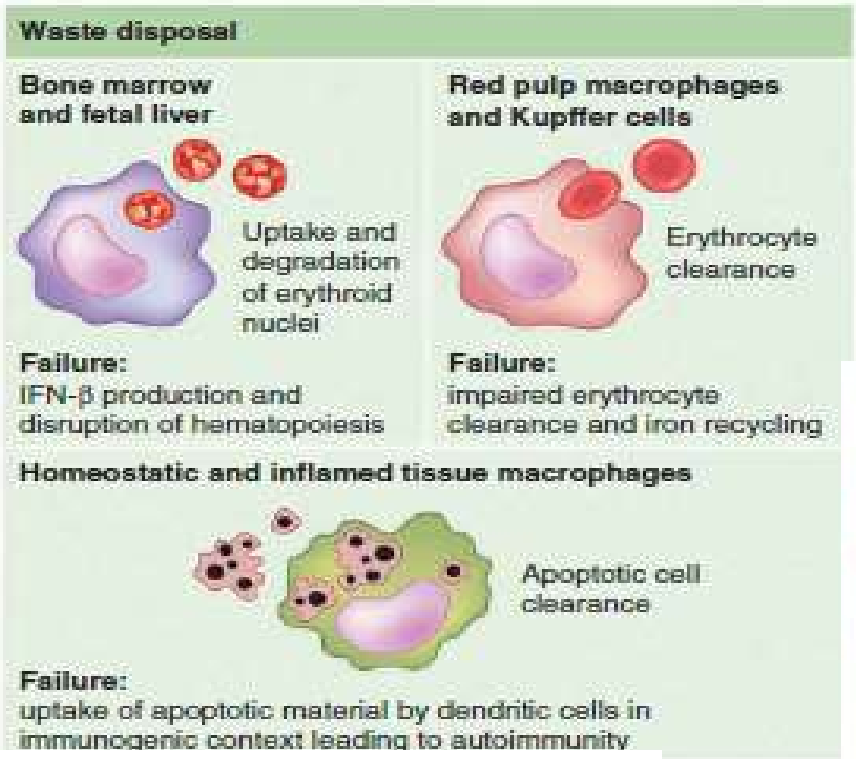
Funzioni fisiologiche dei macrofagi tissutali

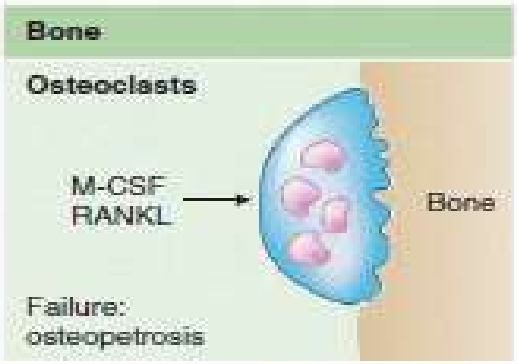
One of the major hallmarks of macrophages is their heterogeneity, which is reflected by their specialized function in a particular microenvironment.

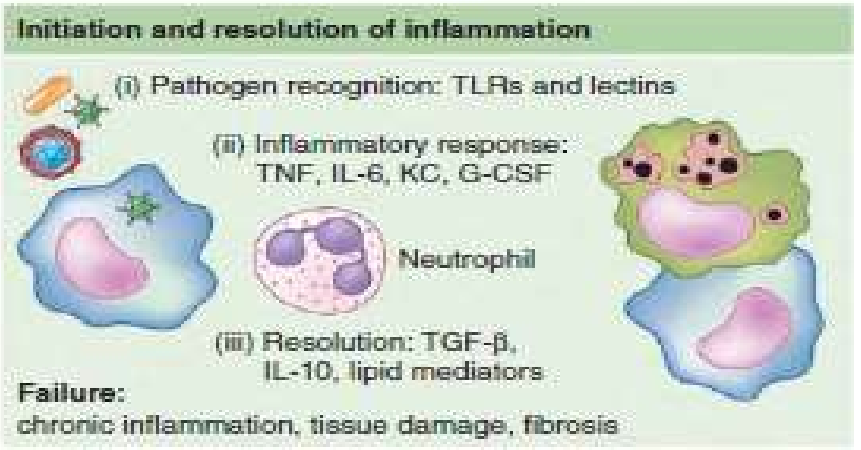


Transcriptional profiling of murine tissue macrophages isolated from the peritoneum, lung, spleen and the brain reveals that there is high diversity but some overlap in genes encoding molecules involved in phagocytosis of apopototic cells and PAMPS/DAMPS recognition

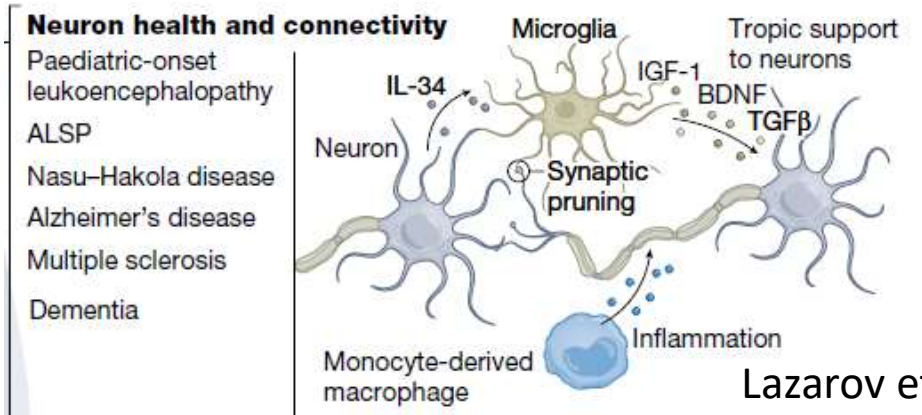
Funzioni fisiologiche e protettive dei macrofagi tissutali ed effetti di alterazioni funzionali







Davies et al. Nature Imm 2013

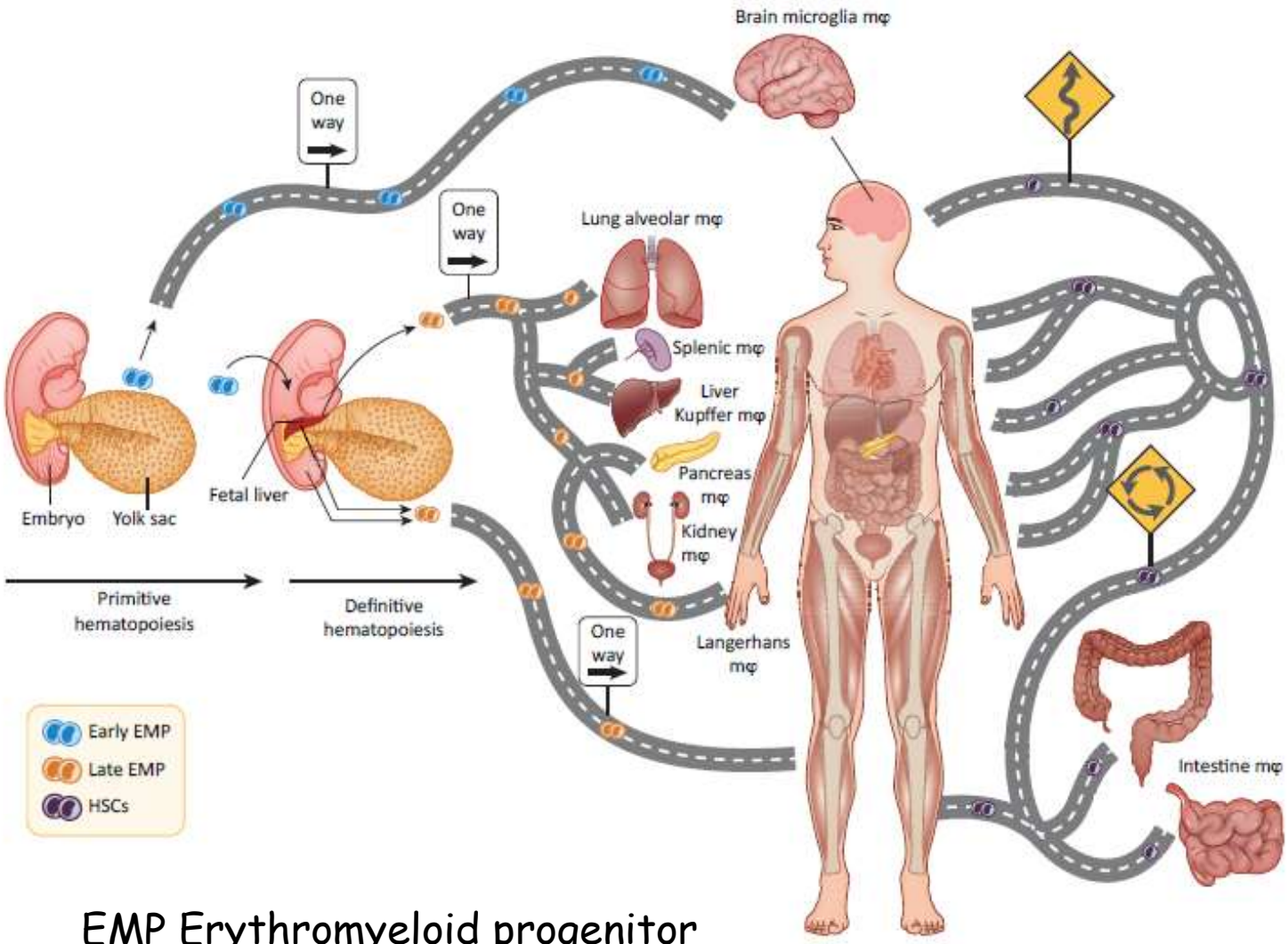


Lazarov et al. Nature Rev Imm 2023

- In contrast to the **classical model of macrophage development**, where macrophages differentiate from circulating monocytes…

….some type of tissue-resident macrophages **arise from yolk sac (sacco vitellino) or fetal liver-derived progenitors** and persist throughout life.

Road of Macrophage Ontogeny into tissues

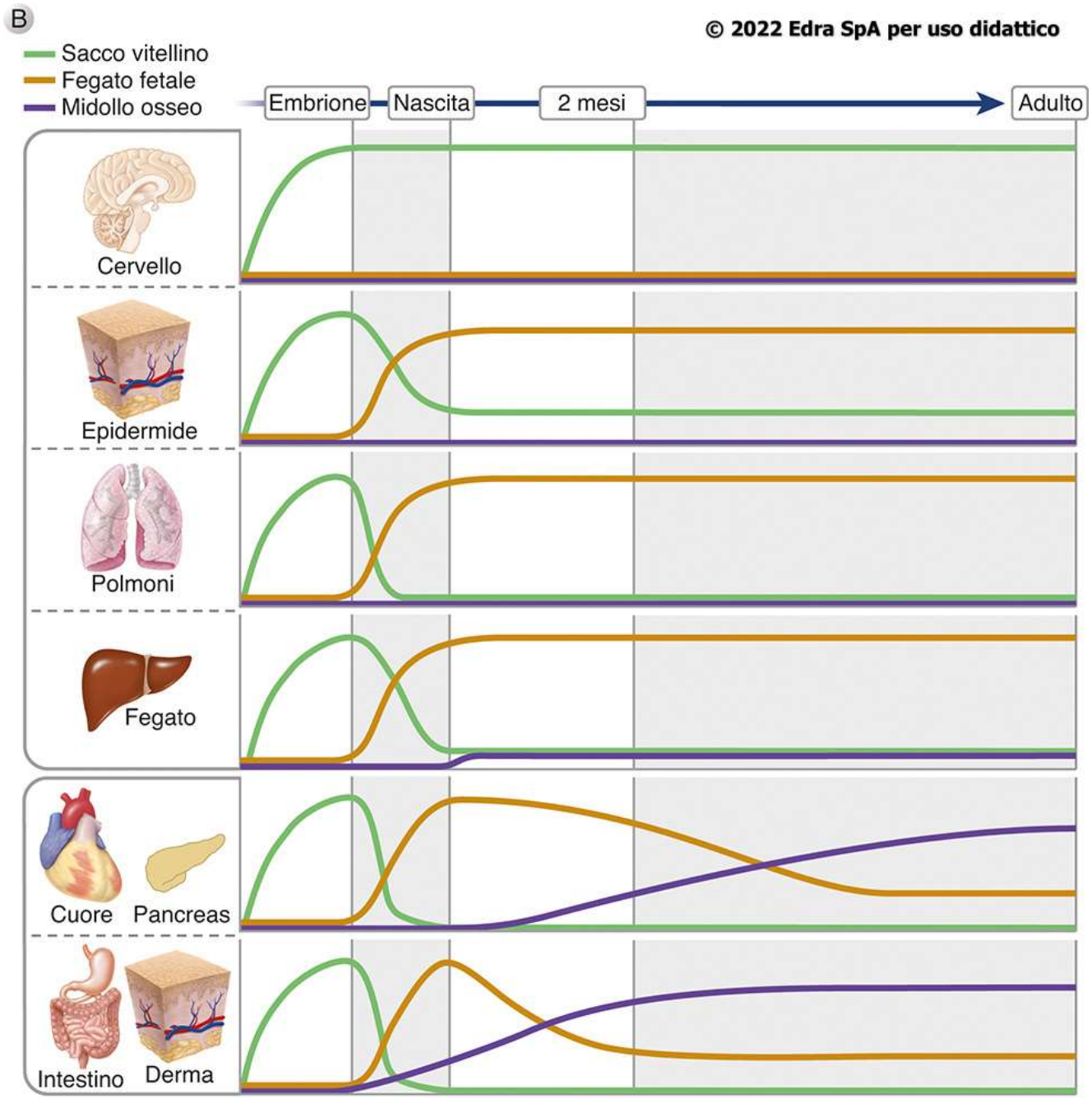


There are various types of macrophages in tissues, which arise from at least three distinct sources: the embryonic yolk sac (YS), the fetal liver (FL), and the **bone marrow (BM).**

EMP Erythromyeloid progenitor

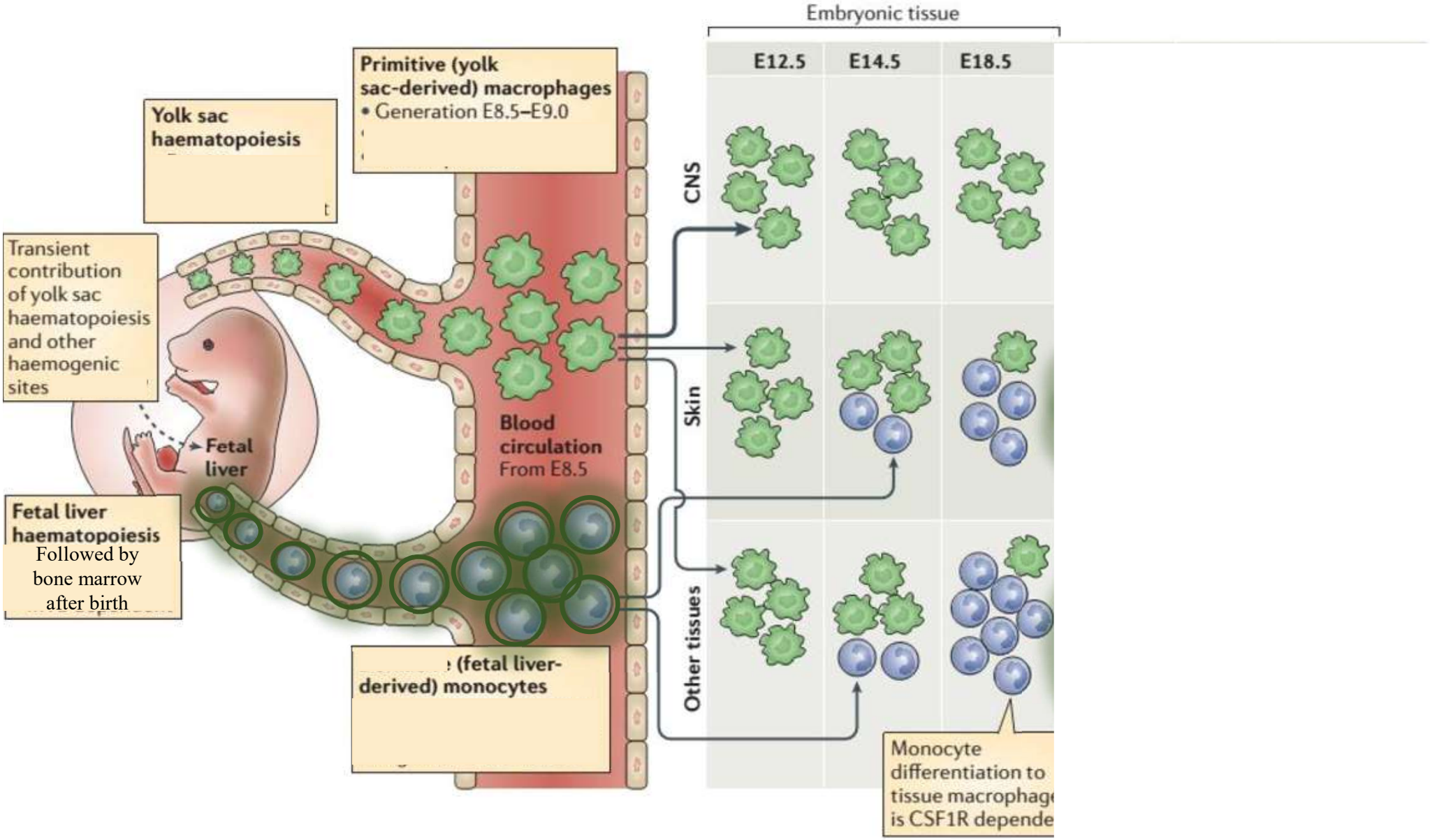
HSC Hematopoietic Stem Cell

Macrophage Ontogeny into tissues



Certain tissues continue to recruit adult monocytes from the bone marrow to generate resident macrophages and **replace with time** the embryonic-derived macrophages.

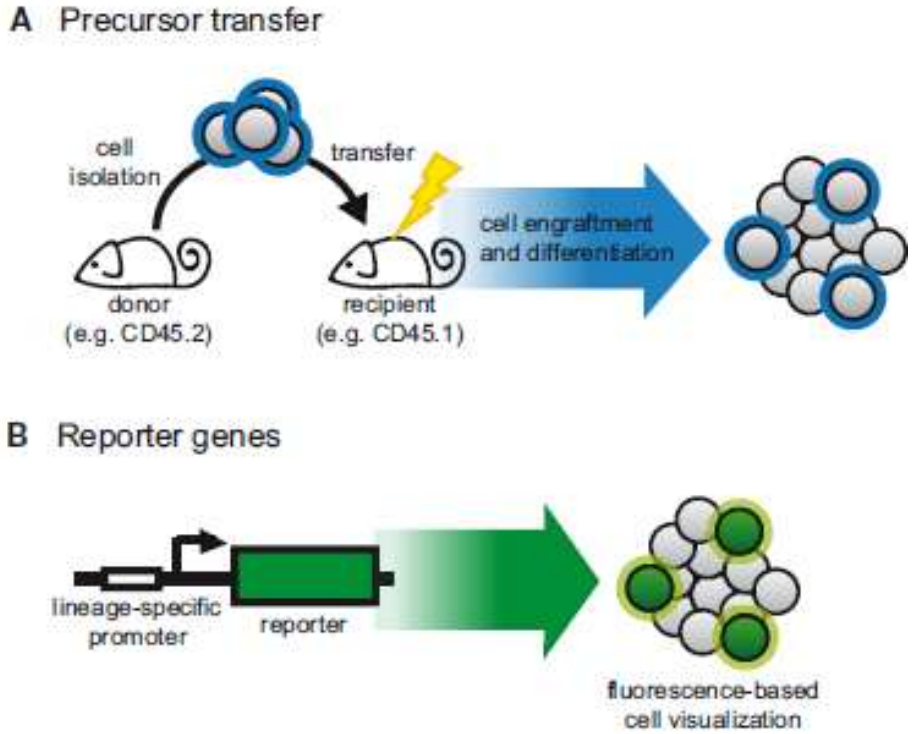
How to track the developmental origin of tissue immune cells (fate mapping).



Fluorescent monocyte precursors (after birth)

How to track the developmental origin of tissue immune cells (fate mapping)

The transfer of purified and pre-marked precursor cells into congenic recipients is the most accessible form of fate mapping as a variety of labeling options can be used to distinguish between donor and host cells (A).

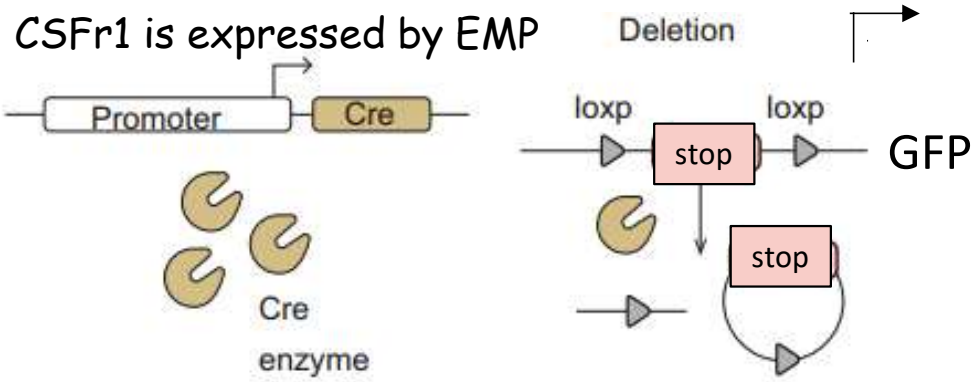


Lineage Restricted Reporters (lineage tracing) is used when the availability of isolatable progenitor cells is limiting and when populations are ontogenetically heterogeneous (B).

Fate Mapping studies

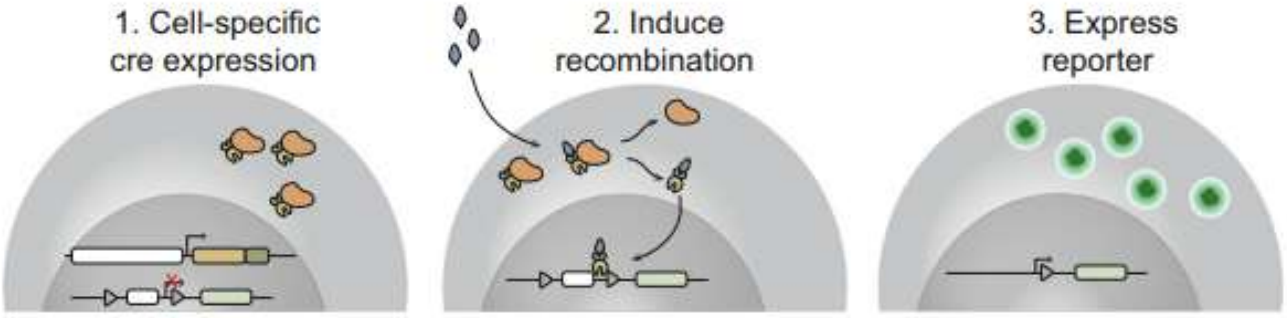
Fate-mapping mice are used to understand how cells of different developmental origins contributed to the assembly of organs and tissues.

In this system **Cre recombinase** drives a fluorescent protein that remains continually expressed UNDER THE CONTROL of a lineage-specific promoter



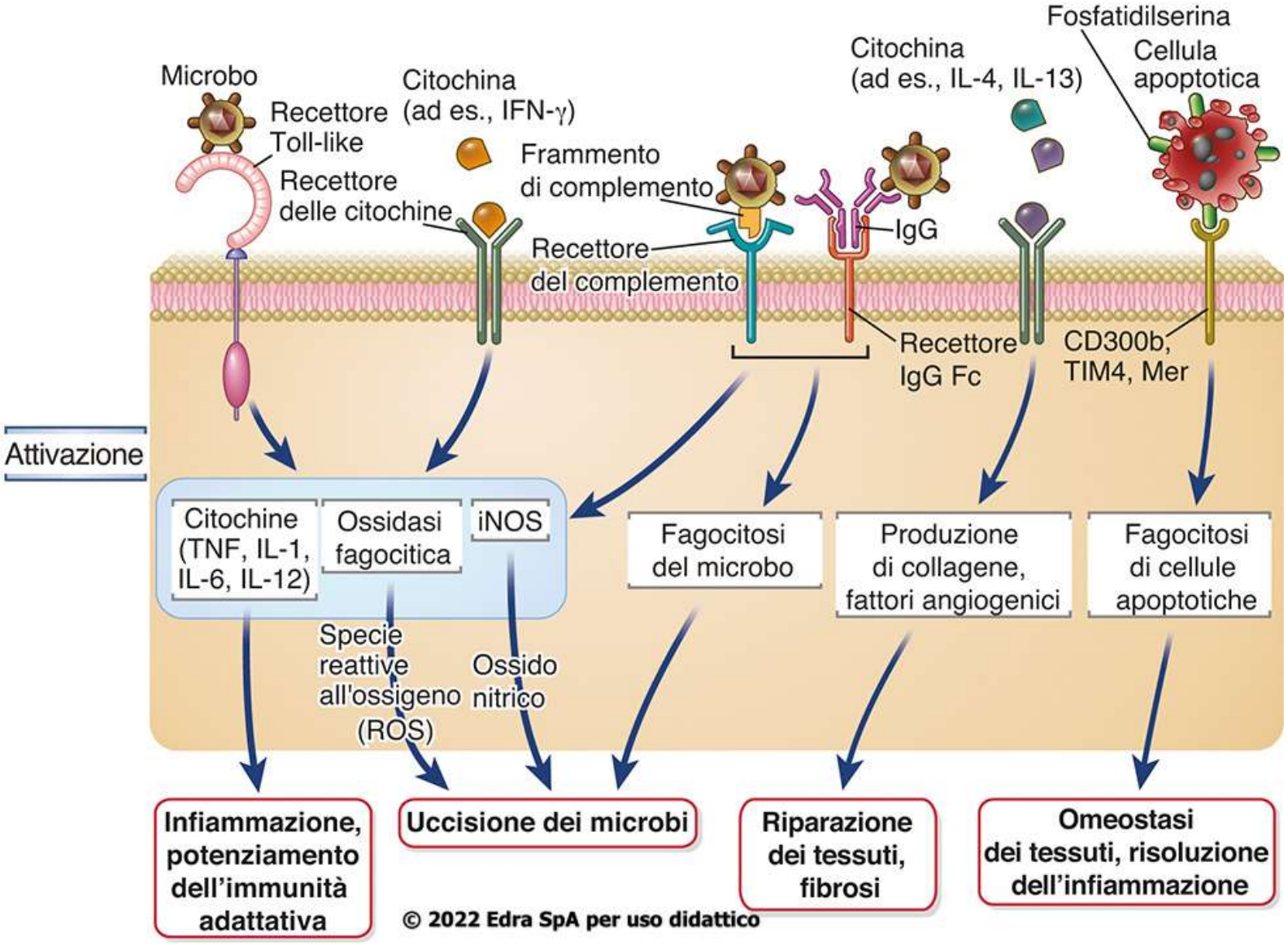
The selection of the promoter for lineage-defining genes is crucial to effective lineage tracing

When the reporter is expressed, it becomes a permanent, heritable mark of the targeted cell population.



Which MAIN functions do phagocytes accomplish?

Funzioni dei macrofagi



PHAGOCYTOSIS

RECEPTOR MEDIATED ENDOCYTOSIS

ENGULFEMENT OF PARTICULATE MATERIAL (CAN BE PATHOGENIC MICROORANISMS OR APOPTOTIC CELLS)

Tissue migration of leukocytes is mediated by chemotatic factors released during infection

Phagocytosis was at first described by **Ilya Metchnicoff** at the end of 19th century

The key experiments was to inject carmine (carminio, un pigmento rosso) into starfish larvae

In 1908, Paul Ehrlich shared the Nobel Prize in Physiology or Medicine with Ilya Metchnikoff "in recognition of their work on immunity."

How do phagocytes recognize pathogens?

Functional classes of microbial recognition receptors capable of binding conserved portions of these molecules

1. **Endocytic pattern-recognition receptors**
   engulfment of pathogens

2. **Chemotactic pattern-recognition receptors**
   Migration to infection site

3. **Signaling pattern-recognition receptors**
   Production of effector molecules that participate to the immune response and affect the nature of adaptive response

Recettori per la fagocitosi

Table 1 | **Selected receptors involved in phagocytosis**

Type of phagocytosis	Receptors	Ligands
Opsonic phagocytosis	Fc receptor family (FcγRI, FcγRIIA and FcγRIIIA)	Antibody-opsonized targets
	Complement receptors (CR1, CR3 and CR4)	Complement-opsonized targets
Non-opsonic phagocytosis	Dectin 1	β-glucan
	Macrophage receptor MARCO	Bacteria (undefined specific ligand)
	Scavenger receptor A	Bacteria (diverse charged molecules)
	αVβ5 integrin	Apoptotic cells
Triggered (nonspecific) phagocytosis	Toll-like receptors	Various, including lipopolysaccharides and lipopeptides

FcγR, Fc receptor for IgG.

Opsonins facilitate recognition, binding, ingestion, and killing of microorganisms by phagocytes

Scavenger receptors in NON opsonic phagocytosis

Class	Scavenger receptor	Ligands	Expression profile
A	SR-A	AcLDL, OxLDL, β-amyloid, molecular chaperones, ECM, AGE, apoptotic cells, activated B-cell, bacteria	Macrophages, mast, dendritic, endothelial and smooth muscle cells
A	MARCO	AcLDL, OxLDL, apoptotic cells, B cells, bacteria	Macrophages, dendritic cells
B	SR-B	HDL, LDL, OxLDL, apoptotic cells	Monocytes/macrophages, hepatocytes and adipocytes
B	CD36	AcLDL, OxLDL, HDL, LDL, VLDL, β-amyloid, AGE, apoptotic cells	Macrophages, platelets, adipocytes, epithelial and endothelial cells
E	LOX-1	OxLDL, molecular chaperones, ECM, AGE, apoptotic cells, activated platelets, bacteria	Endothelial and smooth muscle cells, macrophages, and platelets
F	SRECI/II	AcLDL, OxLDL, molecular chaperones, apoptotic cells	Endothelial cells and macrophages
G	SR-PSOX	OxLDL and bacteria	Macrophages, smooth muscle, dendritic, endothelial cells, and B- and T cells.
H	FEEL-I/II	AcLDL, molecular chaperones, ECM, AGE, bacteria	Monocytes/macrophages, endothelial cell

Destruction of antibody-coated pathogens via Fc receptors

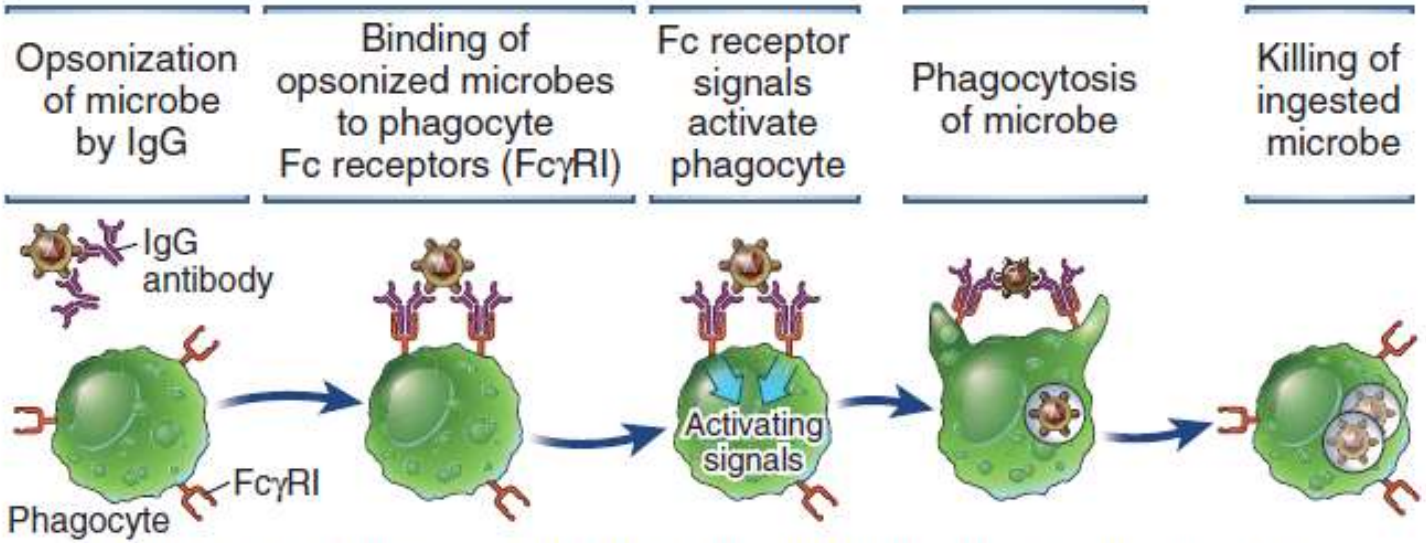
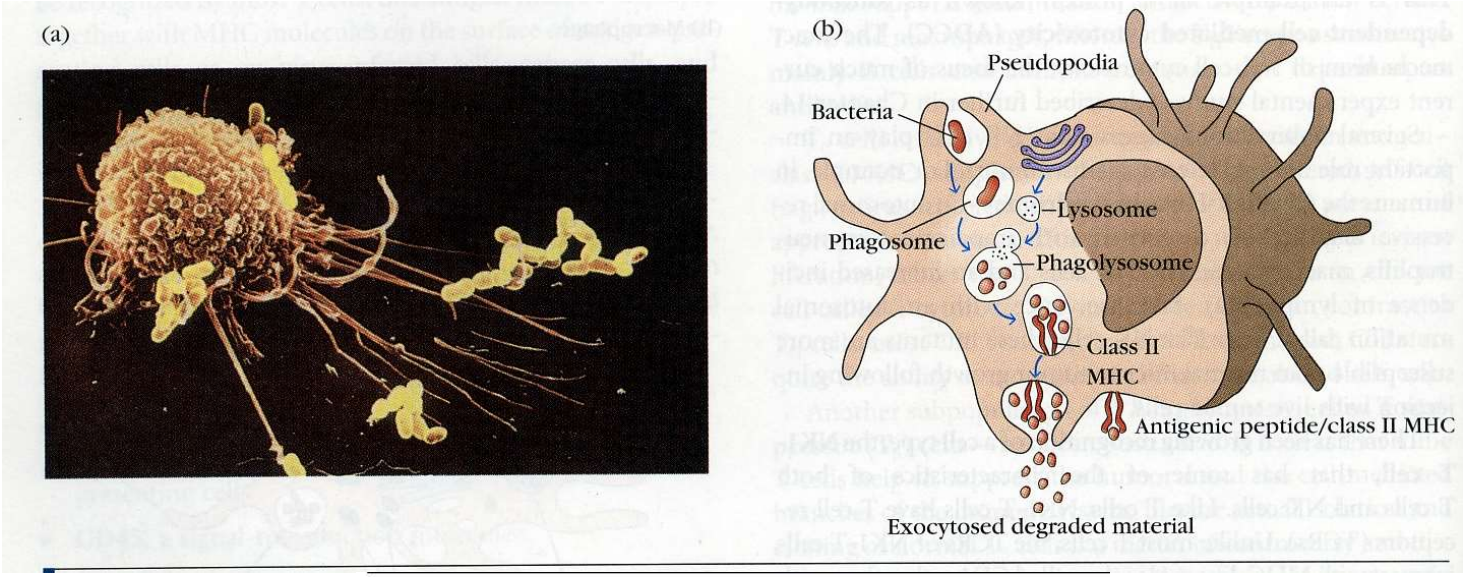


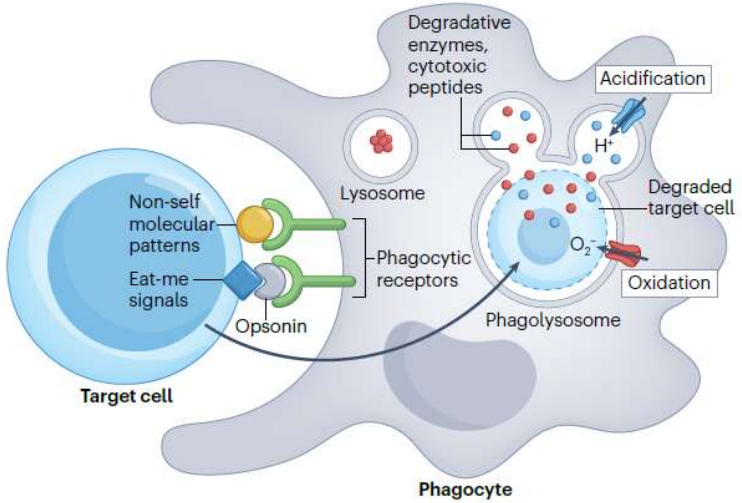
FIGURE 13.4 Antibody-mediated opsonization and phagocytosis of microbes. Antibodies of certain IgG subclasses bind to microbes and are then recognized by Fc receptors on phagocytes. Signals from the Fc receptors promote the phagocytosis of the opsonized microbes and activate the phagocytes to destroy these microbes. The microbicidal mechanisms of phagocytes are described in Chapters 4 (see Fig. 4.13) and 10 (see Fig. 10.7).

The process of coating particles to promote their phagocytosis is called opsonization

dal greco antico *ópson*, cibo

Phagocytosis





Zipper-like mechanism of injestion

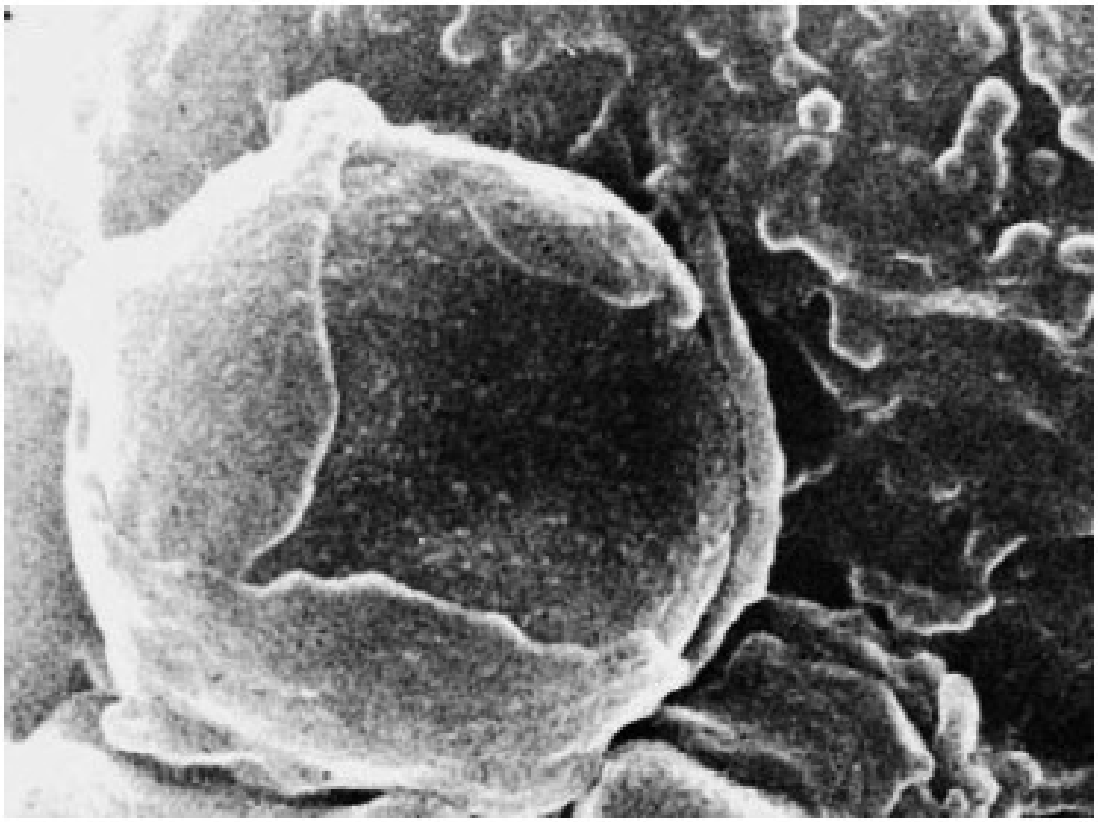


Figure 2. Electron micrograph to illustrate extension of macrophage pseudopodia during engulfment of an IgG-opsonised sheep erythrocyte, by a zipper-type mechanism.

Phagocytosis is a multi-phase event that requires:

Chemotaxis

Adhesion

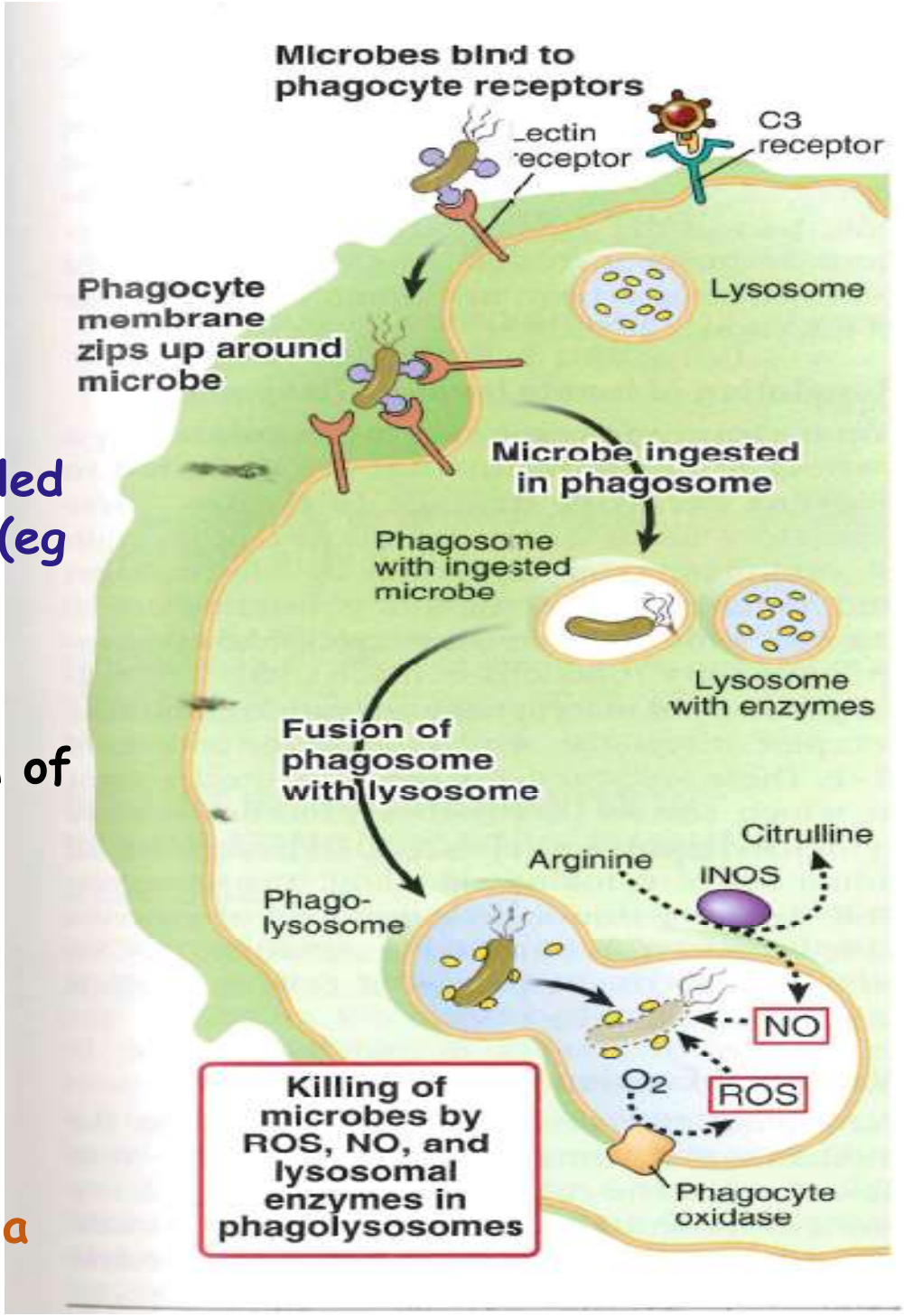
Recognition

Induction of membrane protrusions called pseudopodia that envelop the microbe (eg the zipper-like mechanism of the Fc receptor)

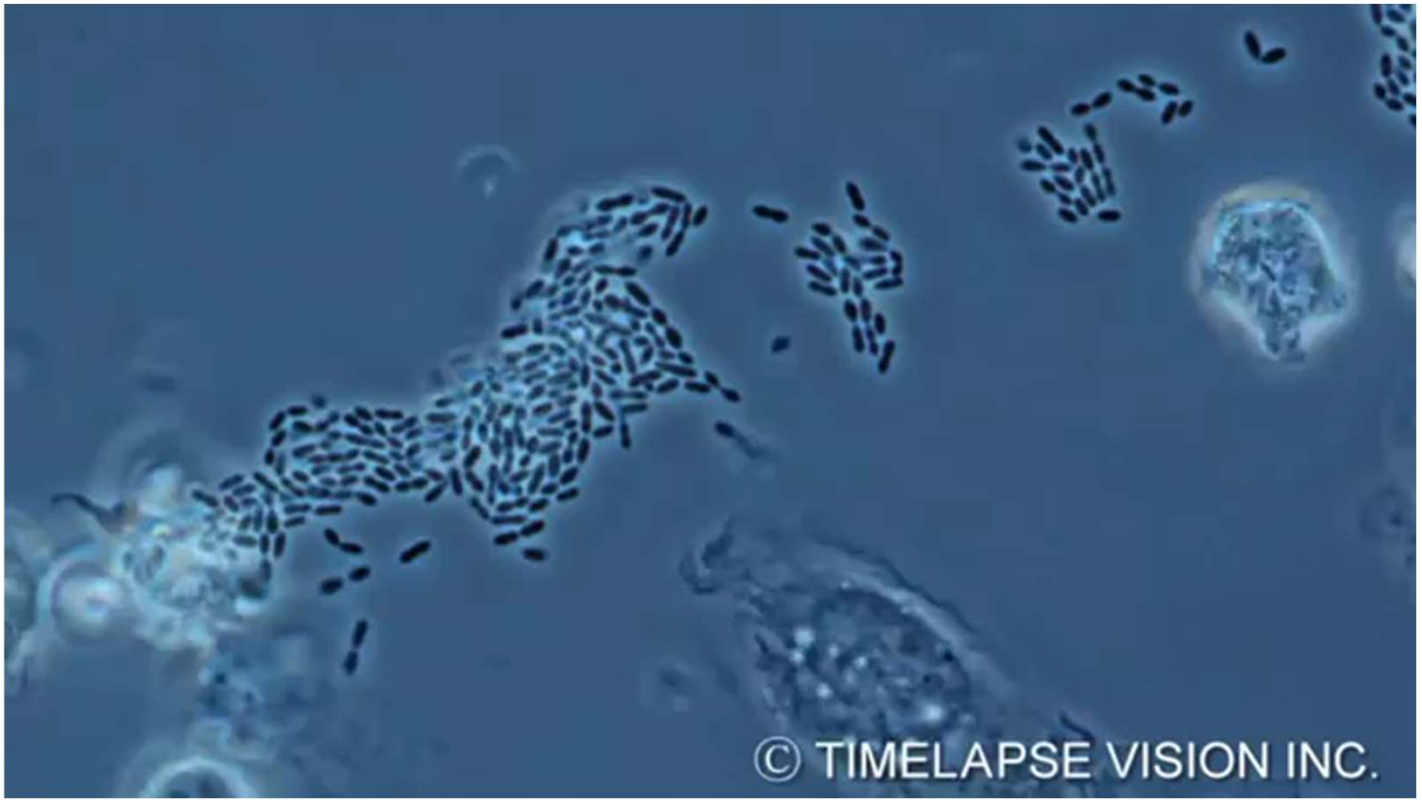
Pseudopodia fusion allows incorporation of the microbe in a structure called a phagosome

Fusion of the phagosome with the lysosome and formation of a phagolysosome

The digested content is eliminated by a process of exocytosis



Phagocytosis is a multi-phase event



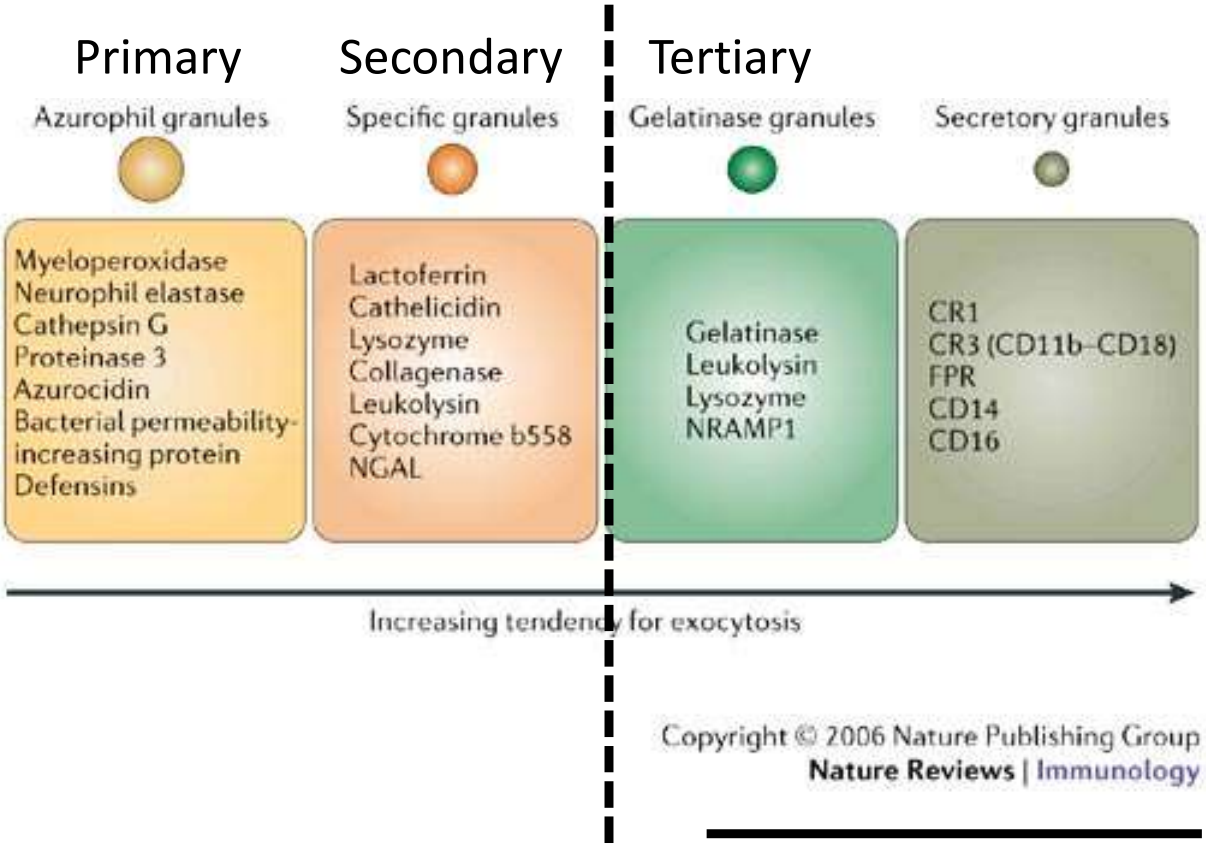
Activated neutrophils and macrophages kill phagocytosed microbes by the action of microbicidal molecules in phagolysosomes

Factors that help destroy microorganisms within a phagolysosome

Class of mechanism	Specific products
Acidification	pH=~3.5–4.0, bacteriostatic or bactericidal
Toxic oxygen-derived products	Superoxide O_2^-, hydrogen peroxide H_2O_2, singlet oxygen $^1O_2^*$, hydroxyl radical OH^*, hypohalite OCl^-
Toxic nitrogen oxides	Nitric oxide NO
Antimicrobial peptides	Defensins and cathelicidin
Enzymes	Lysozyme—dissolves cell walls of some Gram-positive bacteria. Acid hydrolases—further digest bacteria
Competitors	Lactoferrin (binds Fe) and vitamin B_{12}-binding protein

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

Neutrophil granules (in addition to lysosomes)



I granuli vengono esocitati precocemente durante il processo di extravasazione

The activation of NADPH oxidase, which occurs when neutrophils or macrophages 'sense' a pathogen via pattern-recognition receptors, entails the phosphorylation and translocation of preformed cytosolic components to the membrane-bound catalytic core of the enzyme

Il processo di esplosione respiratoria è un meccanismo per produrre metaboliti estremamente tossici

Phagocyte oxidase reduces molecular oxygen into reactive oxygen species (ROS) such as superoxide radicals, with the reduced form of nicotinamide adenine dinucleotide phosphate (NADPH) acting as a cofactor

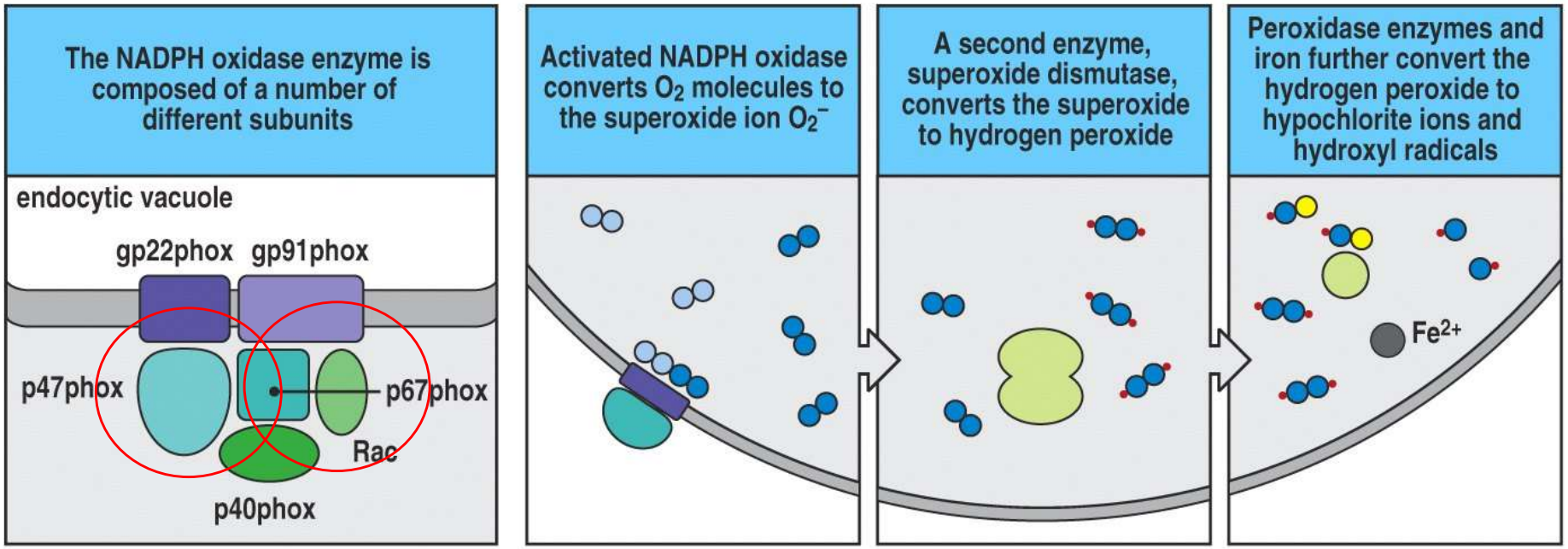
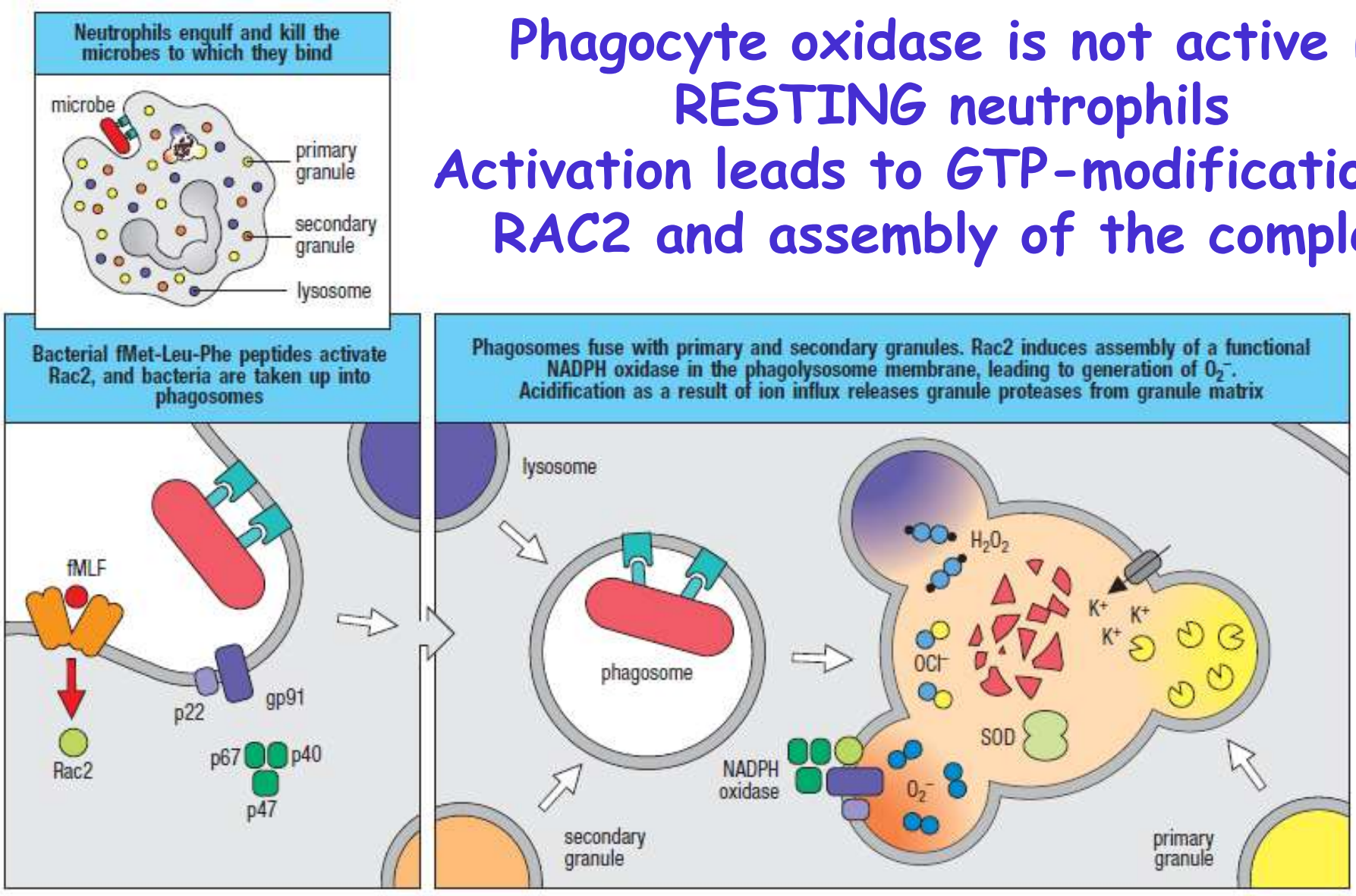


Figure 2-7 Immunobiology, 6/e. (© Garland Science 2005)

Superoxide anion is enzymatically dismutated into hydrogen peroxide, which is used by the enzyme (myelo)peroxidase to produce microbicidal oxidized halogens and reactive oxygen species (ROS) that are toxic for bacteria.

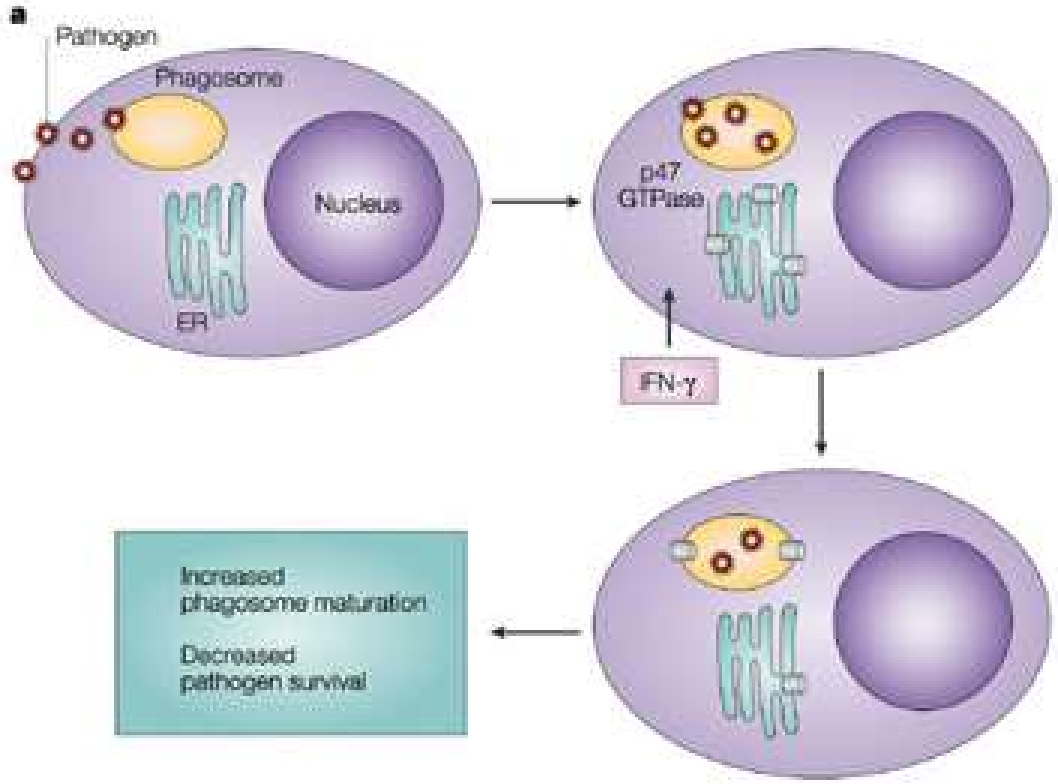
The process by which ROS are produced is called the **respiratory burst**, because it requires oxygen consumption (cellular respiration).

Phagocyte oxidase is not active in RESTING neutrophils
Activation leads to GTP-modification of RAC2 and assembly of the complex



Furthermore, phagosome acidification dissociates granule enzymes (such as cathepsin G and elastase (yellow) from their proteoglycan matrix, leading to their cleavage and activation by lysosomal proteases

Phagocyte oxidase is not active in RESTING macrophages as it cannot be assembled



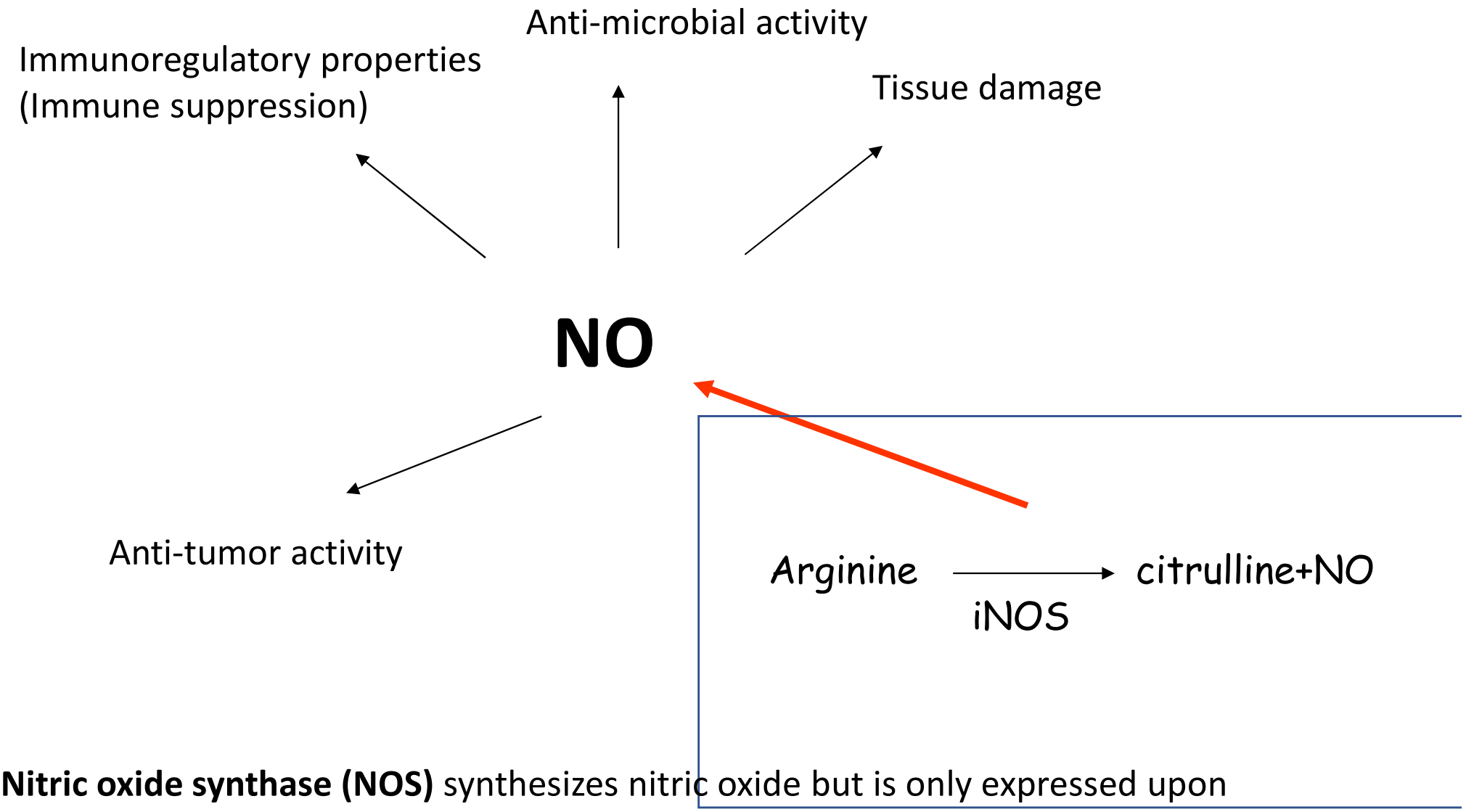
IFN gamma activated macrophages induce production of the oxidase subunit GTPase p47

Factors that help destroy microorganisms within a phagolysosome

Class of mechanism	Specific products
Acidification	pH=~3.5–4.0, bacteriostatic or bactericidal
Toxic oxygen-derived products	Superoxide O_2^-, hydrogen peroxide H_2O_2, singlet oxygen $^1O_2^*$, hydroxyl radical OH^*, hypohalite OCl^-
Toxic nitrogen oxides	Nitric oxide NO
Antimicrobial peptides	Defensins and cationic proteins
Enzymes	Lysozyme—dissolves cell walls of some Gram-positive bacteria. Acid hydrolases—further digest bacteria
Competitors	Lactoferrin (binds Fe) and vitamin B_{12}-binding protein

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

Nitric oxide is ha powerful anti-microbial compound but have also other activities



Nitric oxide synthase (NOS) synthesizes nitric oxide but is only expressed upon activation (**iNOS, also called NOS2**, stands for inducible NOS)

MECCANISMI COINVOLTI NEL KILLING DEI BATTERI

Class of mechanism	Specific products
Acidification	pH=~3.5–4.0, bacteriostatic or bactericidal
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Figure 2-6 Immunobiology, 6/e. (© Garland Science 2005)

Killing mechanisms

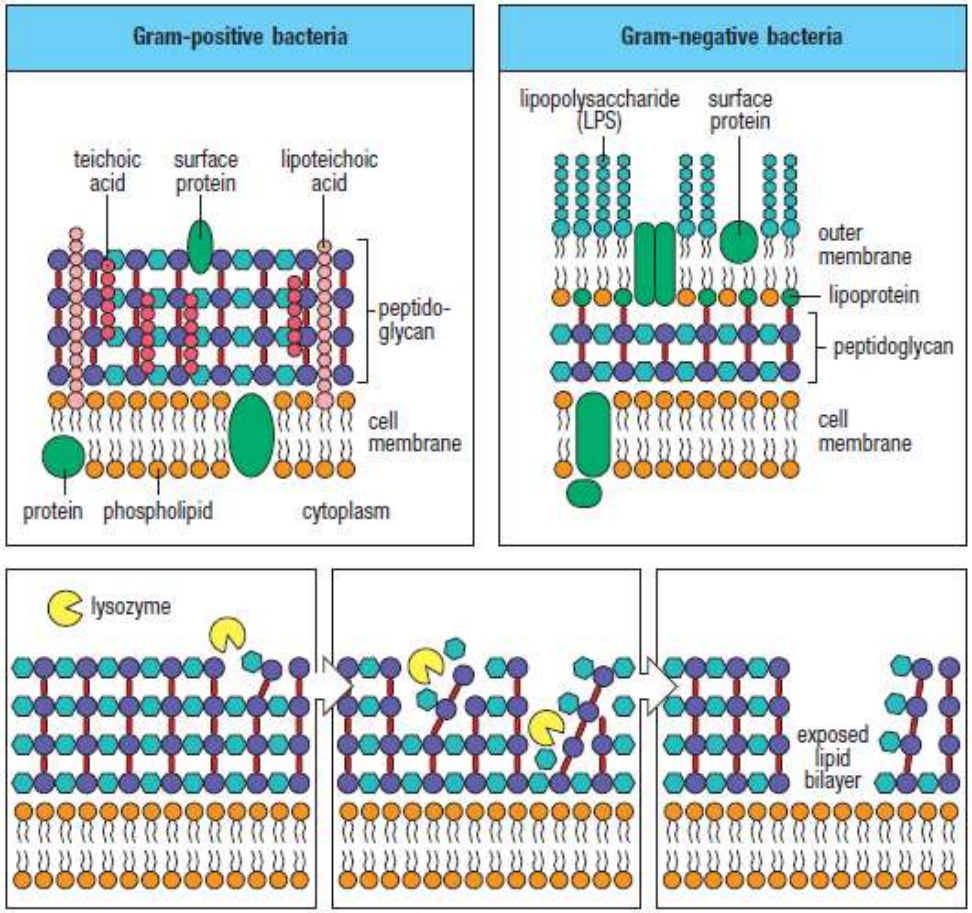
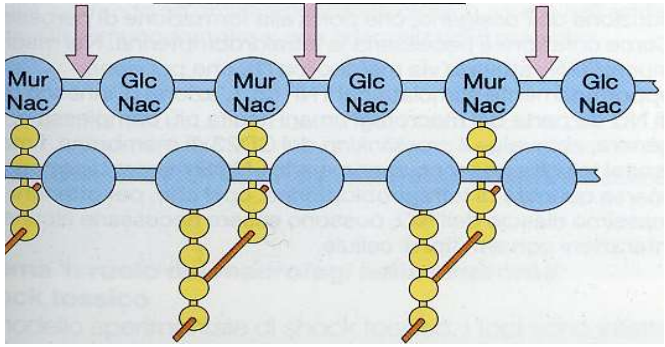


Fig. 2.9 Lysozyme digests the cell walls of Gram-positive and Gram-negative bacteria. Upper panels: the peptidoglycan of bacterial cell walls is a polymer of alternating residues of β-(1,4)-linked *N*-acetylglucosamine (GlcNAc) (large turquoise hexagons) and *N*-acetylmuramic acid (MurNAc) (purple circles) that are cross-linked by peptide bridges (red bars) into a dense three-dimensional network. In Gram-positive bacteria (upper left panel), peptidoglycan forms the outer layer in which other molecules are embedded such as teichoic acid and the lipoteichoic acids that link the peptidoglycan layer to the bacterial cell membrane itself. In Gram-negative bacteria (upper right panel), a thin inner wall of peptidoglycan is covered by an outer lipid membrane that contains proteins and lipopolysaccharide (LPS). Lipopolysaccharide is composed of a lipid, lipid A (turquoise circles), to which is attached a polysaccharide core (small turquoise hexagons). Lysozyme (lower panels) cleaves β-(1,4) linkages between GlcNAc and MurNAc, creating a defect in the peptidoglycan layer and exposing the underlying cell membrane to other antimicrobial agents. Lysozyme is more effective against Gram-positive bacteria because of the relatively greater accessibility of the peptidoglycan.

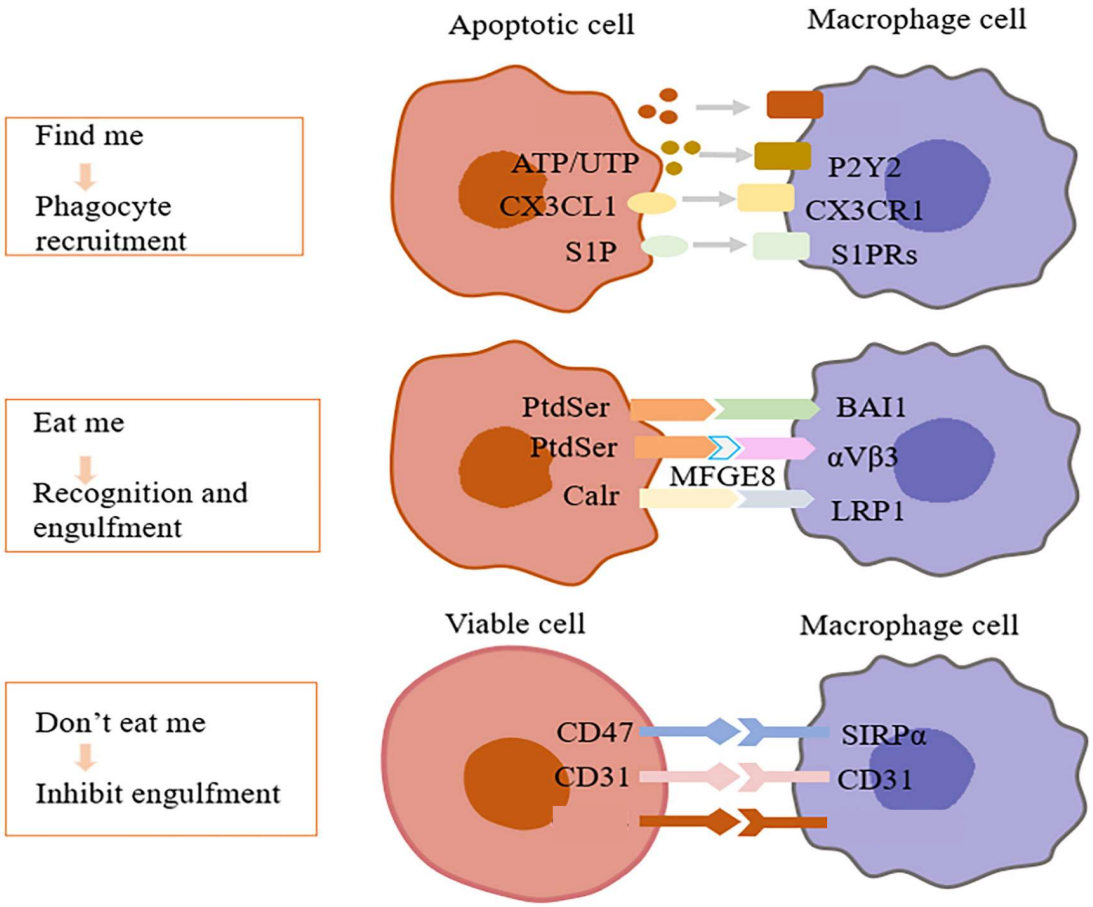
Lysozyme action



La fagocitosi ha anche un ruolo in processi fisiologici promuovendo l'eliminazione di cellule apoptotiche

Fagocitosi di cellule apoptotiche

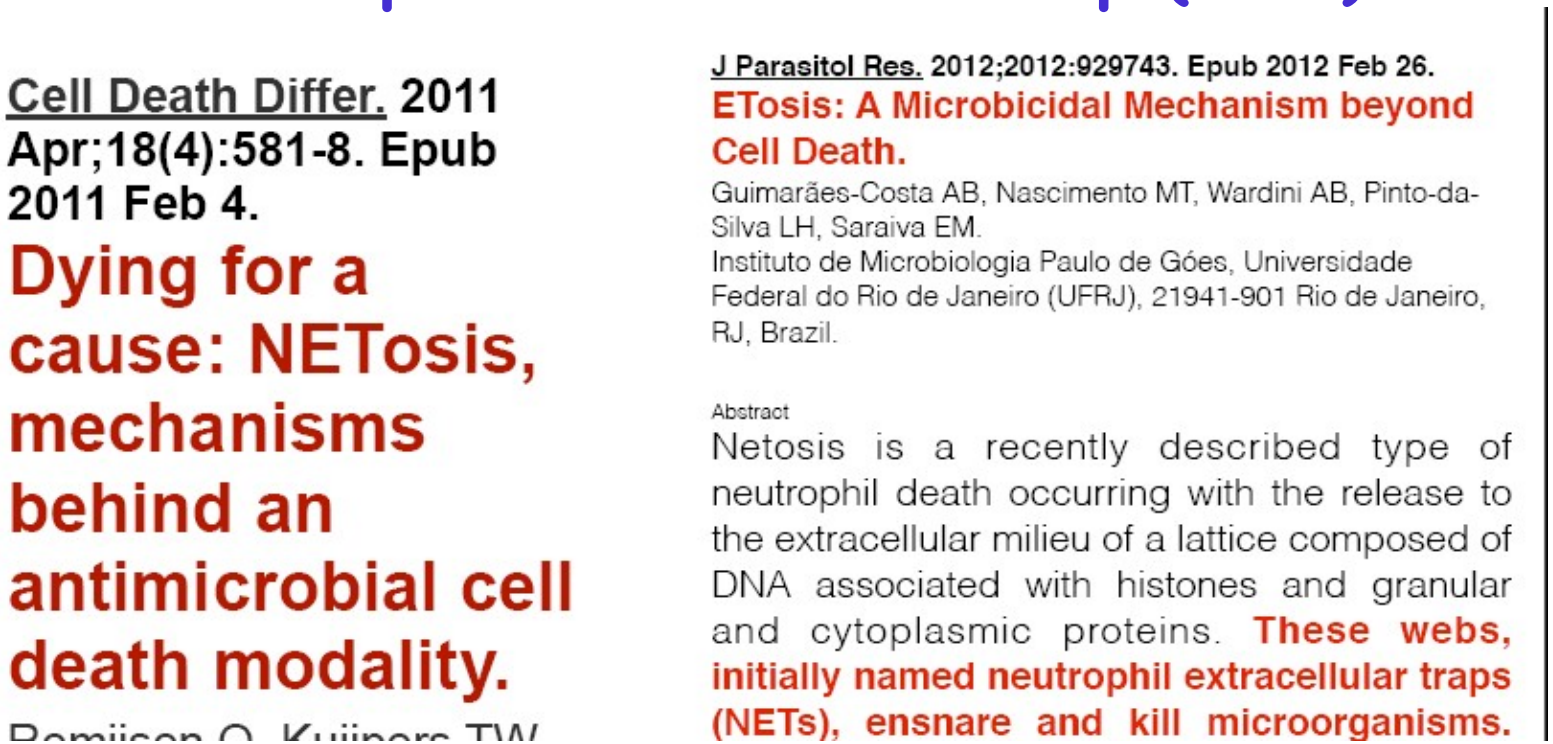
The human body turns over 100 billion cells every day to maintain normal development, tissue homeostasis, or physiological function by clearing unwanted (damaged, dysfunctional, aged, or harmful) cells.



Efferocytosis is a highly conservative physiological process, involving **phagocytes** and **apoptotic cells (ACs)**. Effective clearance of ACs should be the ultimate destination of apoptosis, and it is also a key link to prevent inflammation and maintain tissue homeostasis under physiological conditions

An alternaive killing mechanism in neutrophils: Neutrophil extracellular trap (NET)

Neutrophil extracellular trap (NET)



Cell Death Differ. 2011 Apr;18(4):581-8. Epub 2011 Feb 4.

Dying for a cause: NETosis, mechanisms behind an antimicrobial cell death modality.

Remijsen Q, Kuijpers TW

J Parasitol Res. 2012;2012:929743. Epub 2012 Feb 26.

ETosis: A Microbicidal Mechanism beyond Cell Death.

Guimarães-Costa AB, Nascimento MT, Wardini AB, Pinto-da-Silva LH, Saraiva EM.

Instituto de Microbiologia Paulo de Góes, Universidade Federal do Rio de Janeiro (UFRJ), 21941-901 Rio de Janeiro, RJ, Brazil.

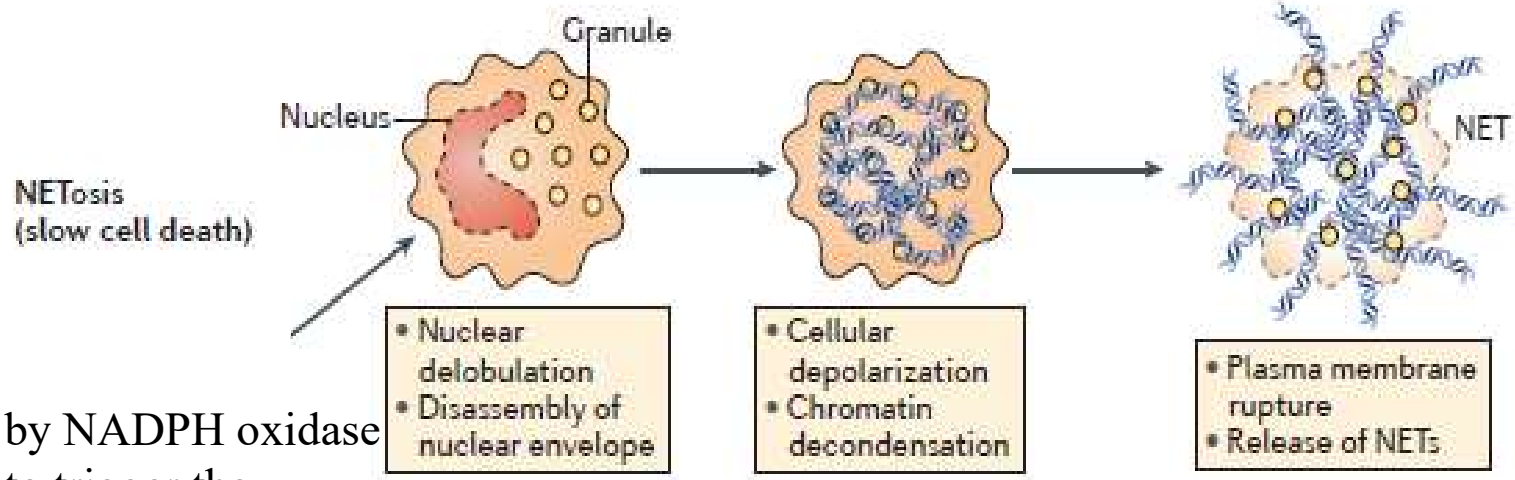
Abstract

Netosis is a recently described type of neutrophil death occurring with the release to the extracellular milieu of a lattice composed of DNA associated with histones and granular and cytoplasmic proteins. **These webs, initially named neutrophil extracellular traps (NETs), ensnare and kill microorganisms.**

DNA and granule content extrusion, which form extracellular networks on which bacteria and fungi are trapped and killed.

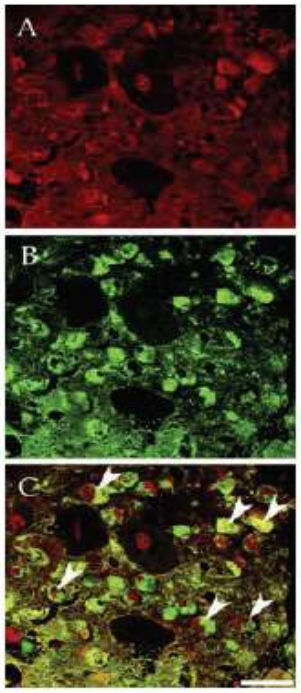
NETs are large, extracellular, web-like structures composed of cytosolic and granule proteins that are assembled on a scaffold of decondensed chromatin

Formazione dei NET mediante NETosi



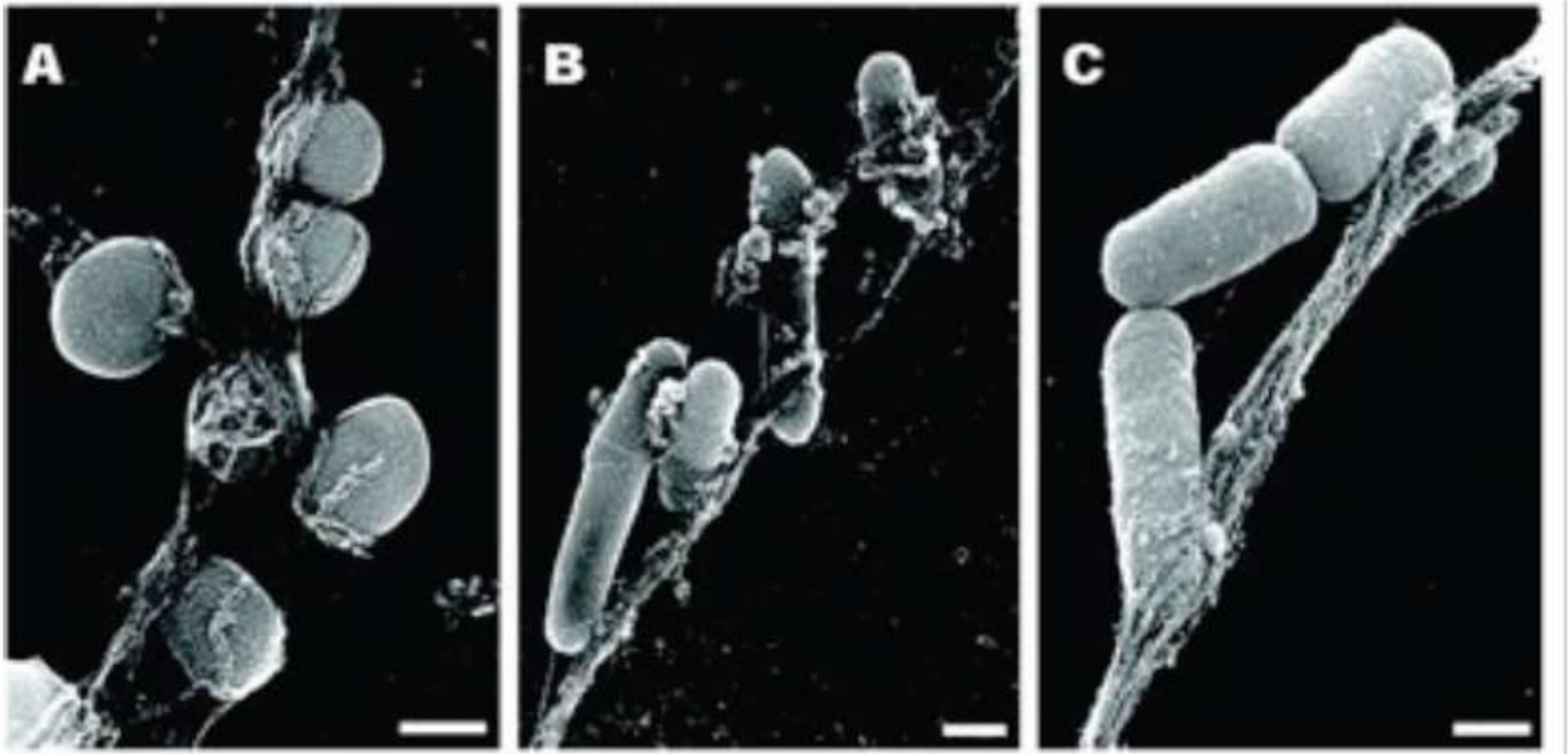
ROS generated by NADPH oxidase stimulate MPO to trigger the activation and translocation of neutrophil elastase (NE) from azurophilic granules to the nucleus, where NE proteolytically processes histones to disrupt chromatin packaging

Pus consists of numerous neutrophils in various stages of NETosis surrounded by NETs. Semithin cryosection of pus from a Molluscum contagiosum lesion stained for NE (green) and chromatin (red). Bar, 20 µm.



Brinkmann V , Zychlinsky A J Cell Biol 2012;198:773-783

NEUTROPHIL EXTRACELLULAR TRAPS ARE COMPOSED OF CHROMATIN AND GRANULE ENZYMES



What else? Macrophages produce cytokines

How do the cytokines of innate immunity work?

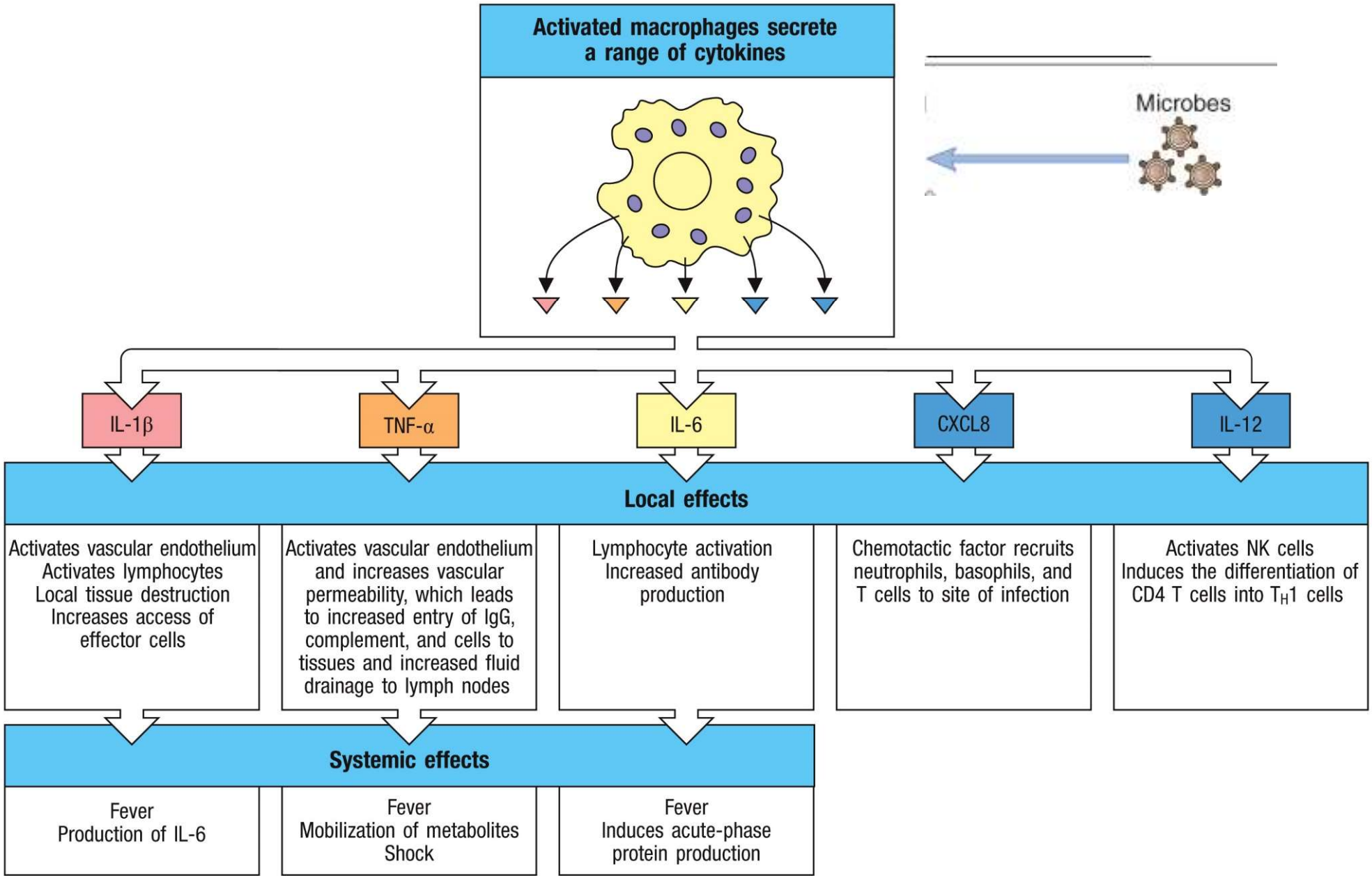
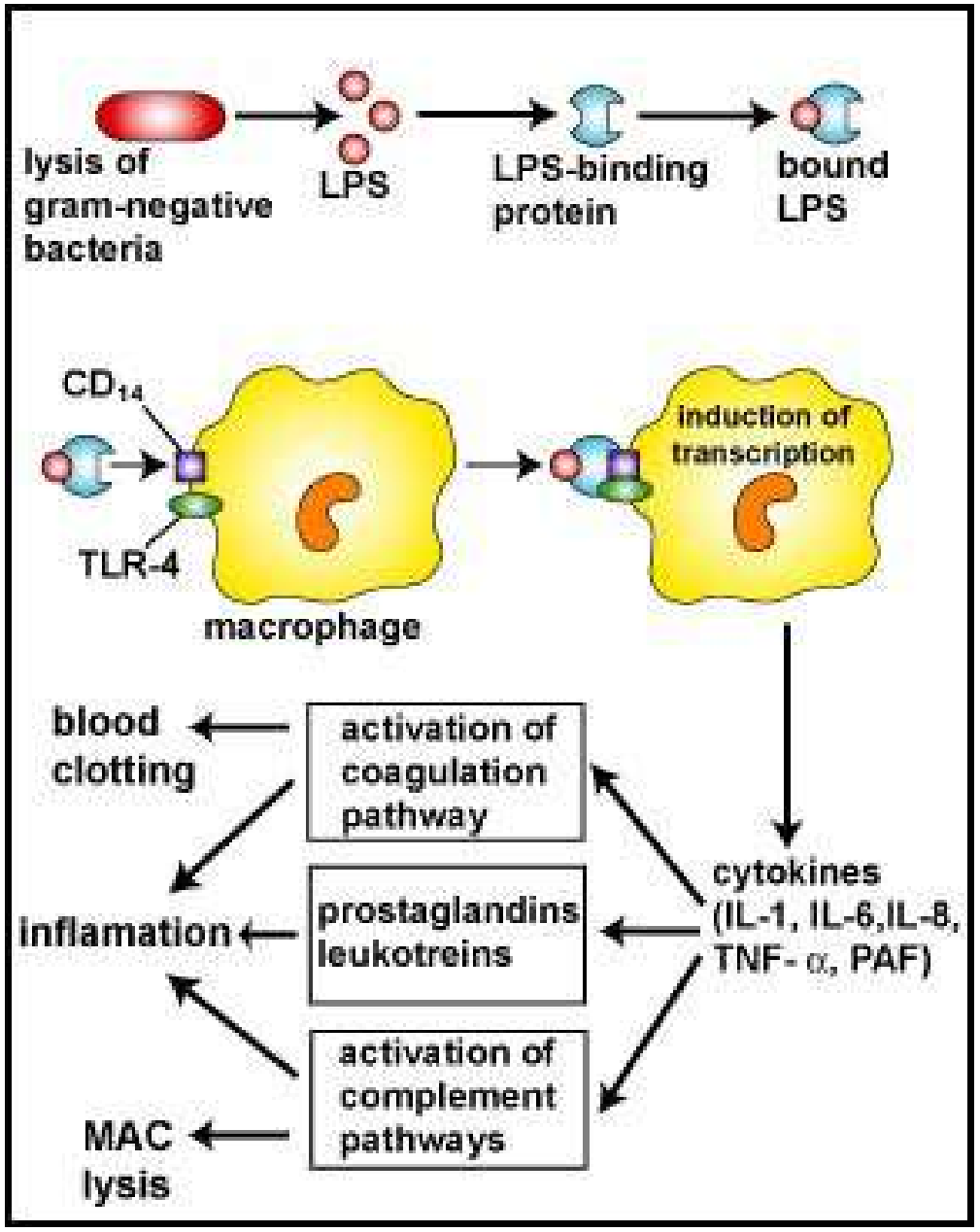


Figure 3.27 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)

Lypopolysaccharid (LPS) recognition and cell activation



The activation of the resulting macrophage leads to the release of chemicals involved in the defensive response called cytokines, including IL-1, IL-6, IL-8, TNF-alpha and PAF.

The cytokines then bind to cytokine receptors on target cells and begin the inflammatory process

L'inflammasoma

Gli inflammasomi sono complessi multiproteici che si formano in seguito a presenza di DAMPS e PAMPS nel citosol.
Hanno la funzione di generare la forma matura di citochine pro-infiammatorie molto importanti chiamate IL-1beta e IL-18

In aggiunta possono indurre la morte delle cellule che producono queste citochine (piroptosi) per promuoverne il rilascio

TABLE 2.2 Distinguishing Properties of Neutrophils and Macrophages

	Neutrophils	Macrophages
Origin	HSCs in bone marrow	HSCs in bone marrow (in inflammatory reactions) Many tissue-resident macrophages: stem cells in yolk sac or fetal liver (early in development)
Life span in tissues	1-2 days	Inflammatory macrophages: days or weeks Tissue-resident macrophages: years
Responses to activating stimuli	Rapid, short lived, enzymatic activity	More prolonged, slower, often dependent on new gene transcription
Phagocytosis	Rapid ingestion of microbes	Prolonged ability to ingest microbes, apoptotic cells, tissue debris, foreign material
Reactive oxygen species	Rapidly induced by assembly of phagocyte oxidase (respiratory burst)	Less prominent
Nitric oxide	Low levels or none	Induced following transcriptional activation of iNOS
Degranulation	Major response; induced by cytoskeletal rearrangement	Not prominent
Cytokine production	Low levels per cell	Major functional activity, large amounts per cell, requires transcriptional activation of cytokine genes
NET formation	Rapidly induced, by extrusion of nuclear contents	No
Secretion of lysosomal enzymes	Prominent	Less

La capacità di degradare componenti antigeniche ha anche implicazioni importanti per la successiva capacità dei macrofagi di presentare l'antigene ai linfociti T helper

Three types of antigen-presenting cells are effective at different stages and in different types of immune responses

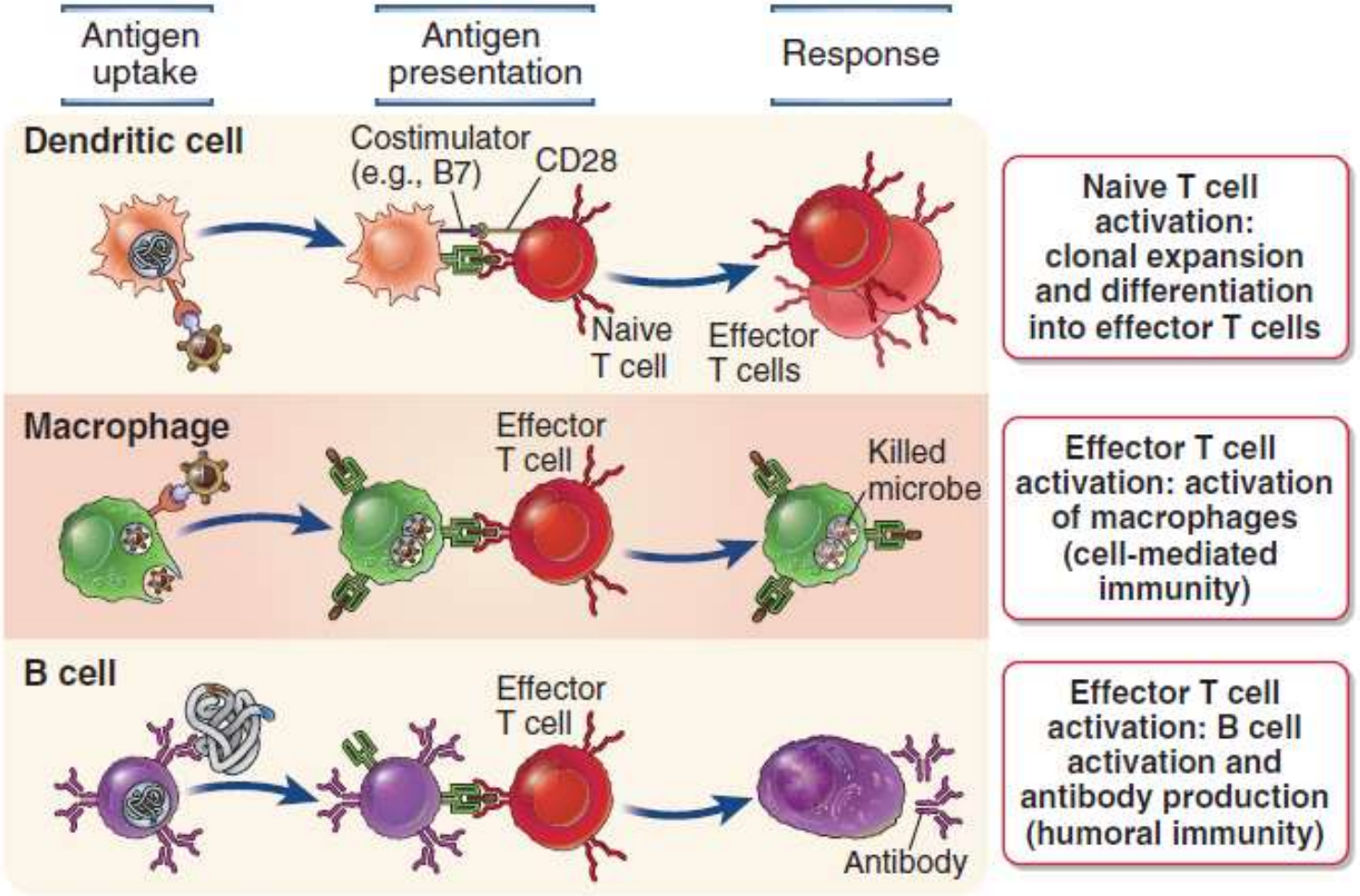
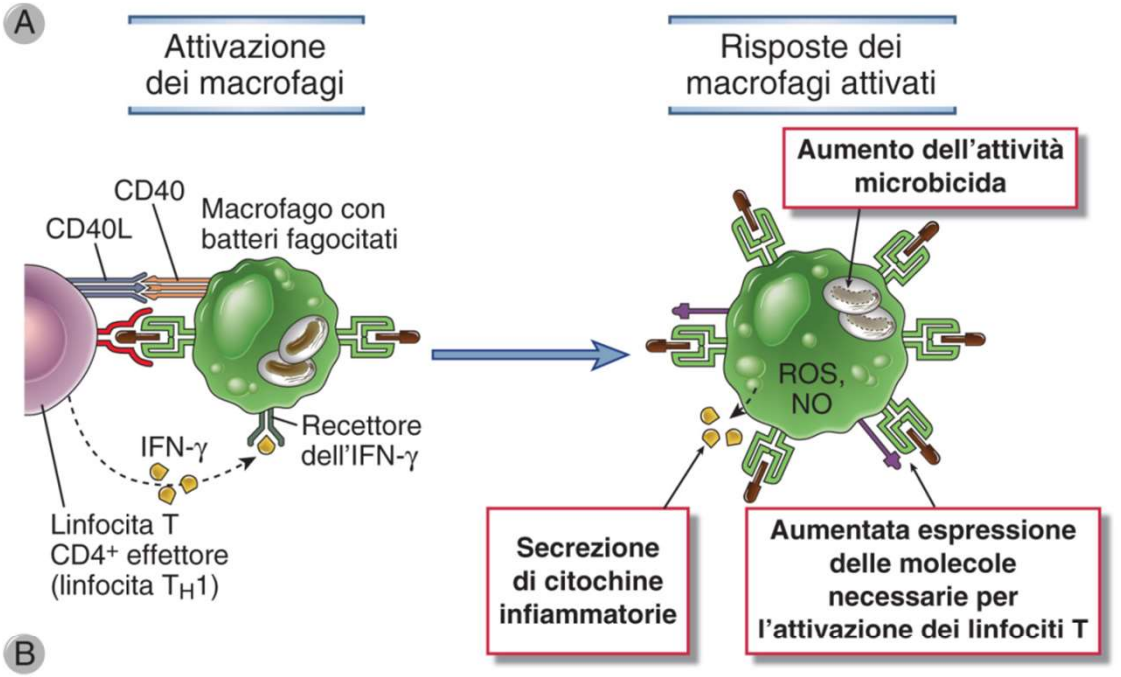


FIGURE 6.2 Functions of different antigen-presenting cells. The three major types of APCs for $CD4^+$ T cells function to display antigens at different stages and in different types of immune responses. Note that effector T cells activate macrophages and B lymphocytes by production of cytokines and by expressing surface molecules; these will be described in later chapters.

ATTIVAZIONE MACROFAGI DA PARTE DEI TH

A



B

Risposta dei macrofagi	Ruolo nell'immunità cellulo-mediata
Produzione di specie reattive , dell'ossigeno, dell'ossido nitrico, aumento dell'espressione degli enzimi lisosomiali	Uccisione dei microbi nei fagolisosomi (funzione effettrice dei macrofagi)
Secrezione delle citochine (TNF, IL-1, IL-12) e delle chemochine	TNF, IL-1, chemochine: reclutamento dei leucociti (infiammazione) IL-12: differenziazione T_H1, produzione dell'IFN-γ
Aumentata espressione delle molecole costimolatorie B7, delle molecole MHC	Aumentata attivazione dei linfociti T (amplificazione della risposta T)

g7.jpg

Immunologia cellulare e molecolare 7 ed

Attivazione dei macrofagi da parte dei linfociti T_H1. A. Una volta attivati dall'IFN-ã gamma; prodotto dai linfociti T_H1 e dalle interazioni CD40L/CD40, i macrofagi svolgono numerose funzioni che mirano all'eliminazione dei microrganismi, stimolano inoltre l'infiammazione e potenziano la capacità di presentare l'antigene. B. Nella tabella sono elencate le principali molecole che mediano le funzioni dei macrofagi. I macrofagi vengono inoltre attivati a esercitare le stesse funzioni durante le risposte innate (vedi Cap. 4).

Ma che succede se i macrofagi ricevono stimoli diversi dall'interferon gamma o dai PAMPs pro-infiammatori?

The type of stimulus promote generation of different type of macrophages

Classical activation → **Macrophages M1**
Powerful effectors of the inflammatory response that kill microorganisms and cancer cells and produce proinflammatory cytokines

Alternative activation → **Macrophage M2**
They control the inflammatory response
Response to parasites (Chitimase and Th2 polarization)
They eliminate of the debris
Promote angiogenesis and tissue repair

Classically and alternatively activated macrophages

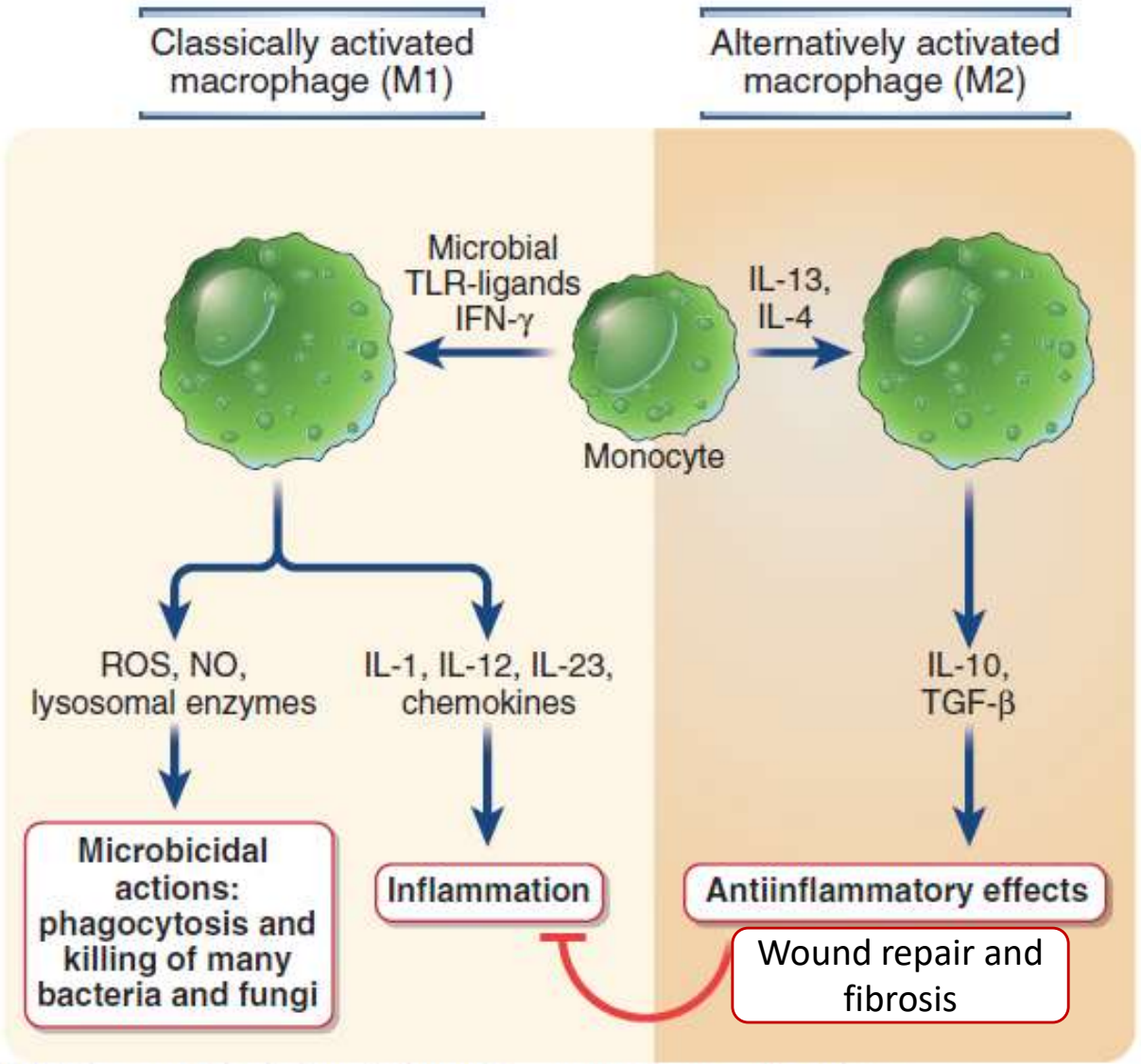


FIGURE 10.10 Classical and alternative macrophage activation. Different stimuli activate tissue macrophages to develop into functionally distinct populations. Classically activated macrophages are induced by microbial products and cytokines, particularly IFN-γ, and are microbicidal and involved in potentially harmful inflammation. Alternatively activated macrophages are induced by IL-4 and IL-13 produced by Th2 cells and other leukocytes and function to control inflammation and to promote tissue repair and fibrosis. Some investigators divide the M2 macrophage population into subpopulations, some of which are mainly antiinflammatory and others are responsible for tissue repair.

Different arginine usage by the two type of Macrophages

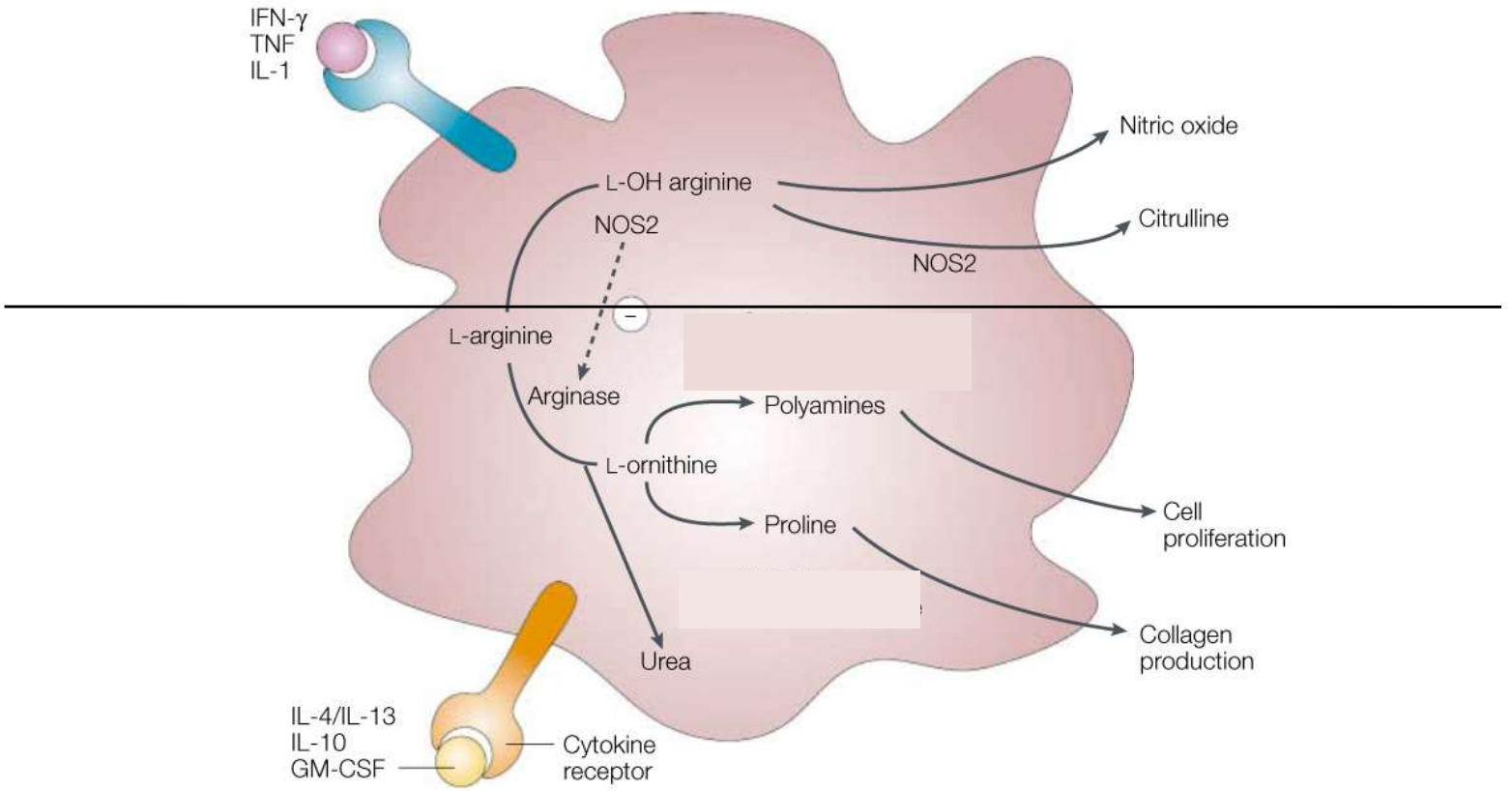
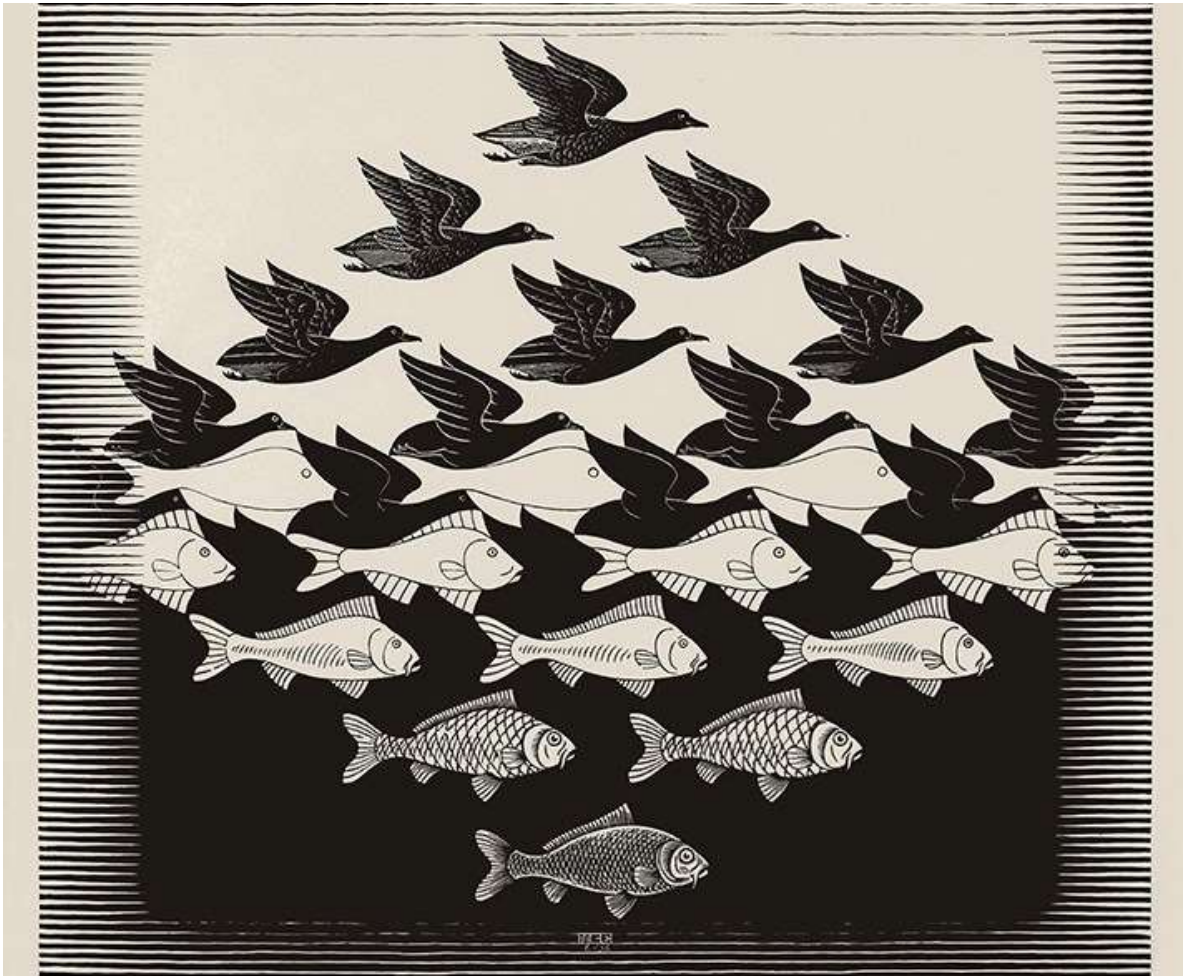


Figure 2 | **Differential utilization of L-arginine by activated macrophages.** Interleukin-4 (IL-4) and IL-13 promote arginase-dependent formation of L-ornithine and, ultimately, fibroblast proliferation and collagen production. Interferon-γ (IFN-γ) enhances the activity of nitric oxide synthase 2 (NOS2) to generate nitric oxide, and inhibits arginase. GM-CSF, granulocyte–macrophage colony-stimulating factor; TNF, tumour-necrosis factor. Adapted, with permission, from REF. 80 © (2001) The American Association of Immunologists, Inc.

Nowadays the M1/M2 classification is considered not to fully cover the total spectrum of in vivo macrophage phenotypes

M2



M1

«**Cielo e acqua**»
Escher

Macrophages are considered a doubled-edged sword in cancer biology

Originally, it was thought the body to recruit macrophages to tumors to defend itself from cancer, but there is increasing evidence that the tumor recruits them for its own support in a manner analogous to a wound attracting macrophages to assist in healing.

Classically activated macrophages are potent effector cells that kill microorganisms and tumor cells, produce copious amounts of proinflammatory cytokines, and activate cytotoxic T lymphocytes. These macrophages are the so-called **M1 macrophages**.

Conversely, macrophages can be activated in a manner that promotes tissue remodeling and repair and preferentially attract T-cell subsets devoid of cytotoxic functions such as regulatory T cells (Tregs) and T-helper type 2 (Th2) cells.
The **alternatively activated macrophages** are the ones referred to as **'M2'-like macrophages**.
When assessing the function of **Tumor-Associated Macrophages TAMs**, they show M2-like features with protumor functions (tumor survival, proliferation, angiogenesis, dissemination, and chemoresistance).

End of the lesson

Questions and answers

What are phagocytes?

What is the origin of phagocytes?

Phagocyte heterogeneity. Diversification of macrophages into subpopulations.

Phagocytosis, what else? Functions of phagocytes.

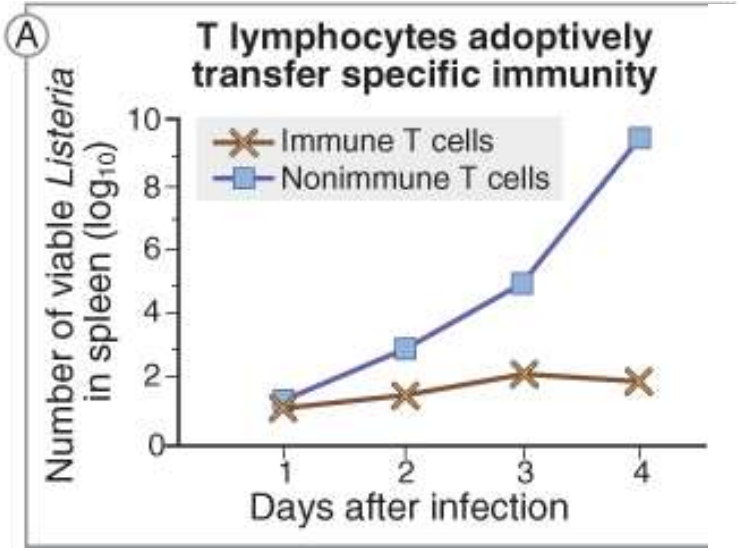
Table 1 | **Markers of circulating myeloid subsets in mice**

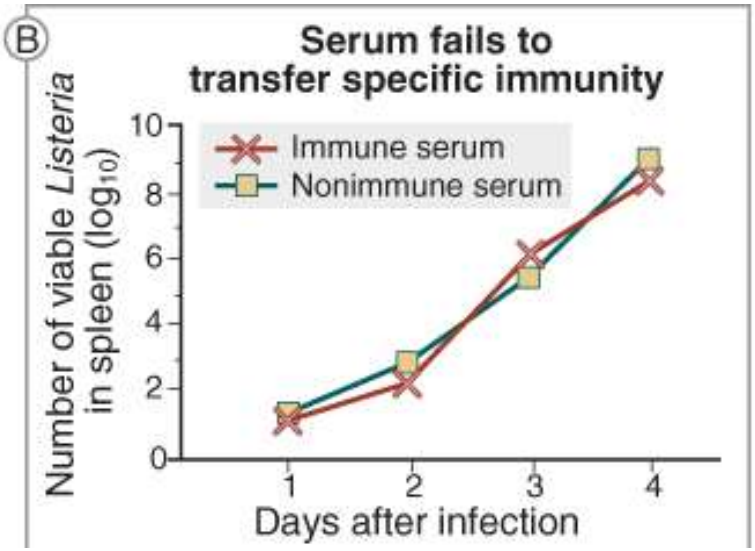
Markers	Granulocytes	Monocytes in inflamed tissues	Resident monocytes in non-inflamed tissues	References
Adhesion molecules				
CD62L	+++	+++	–	11,150
LFA1/$\alpha_L\beta_2$	+++	+++	++	8,126
CD11b/α_M	+++	+++	+++	8,126
CD11c/α_X	–	–	–	8,126
CD49d/α_4	Regulated	+++	N.D.	126,151
Pecam1	+++	+++	+	152,153
Chemokine receptors				
CX_3CR1	–	+	+++	8,11,154
CCR2	–	+	–	11
Other molecules				
Gr1	+++	++	–	8,155
F4/80	–	+	+	8,156
7/4	+++	+++	N.D.	126

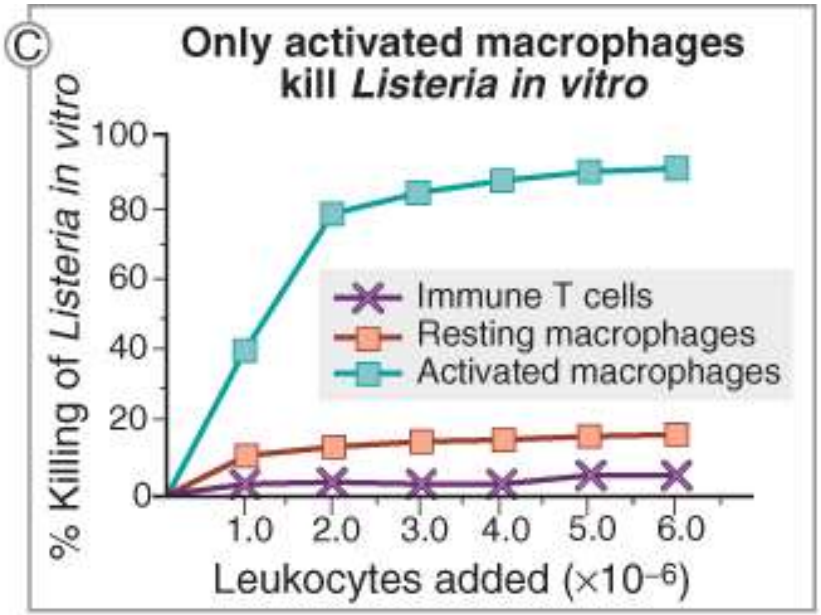
α_4, α_4-integrin chain; $\alpha_L\beta_2$, $\alpha_L\beta_2$-integrin; α_M, α_M-integrin chain; α_X, α_X-integrin chain; CCR2, CC-chemokine receptor 2; CD62L, CD62 ligand; CX_3CR1, CX_3C-chemokine receptor 1; LFA1, lymphocyte function-associated antigen 1; N.D., not determined; Pecam1, platelet/endothelial cell-adhesion molecule 1.

NELL'UOMO I MONOCITI INFIAMMATORI SONO CD14+CCR2+ E I MONOCITI RESIDENTI SONO CD16+CX3CR1+

I linfociti T sono importanti per la risposta a patogeni mediata da macrofagi

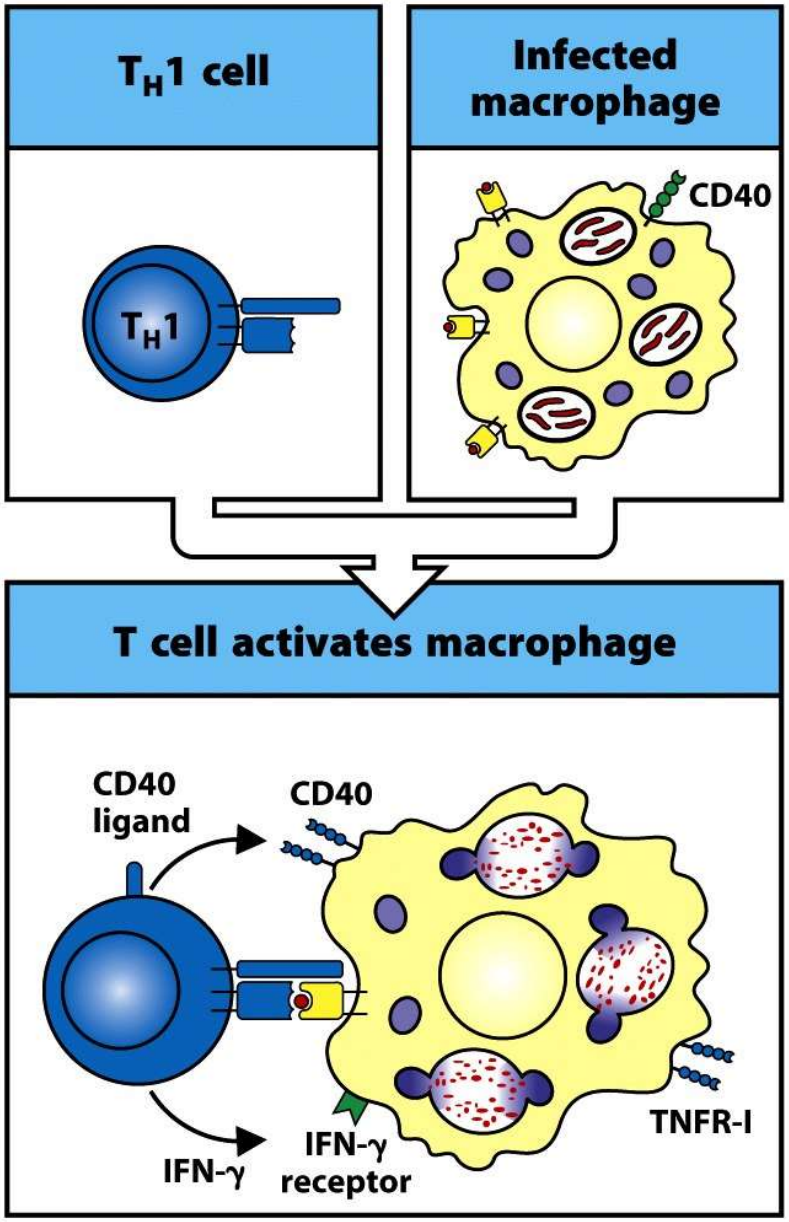






. Abbas & Lichtman: Cellular and Molecular Immunology 5e www.stu

Perché i linfociti T sono importanti per l'attivazione dei macrofagi?



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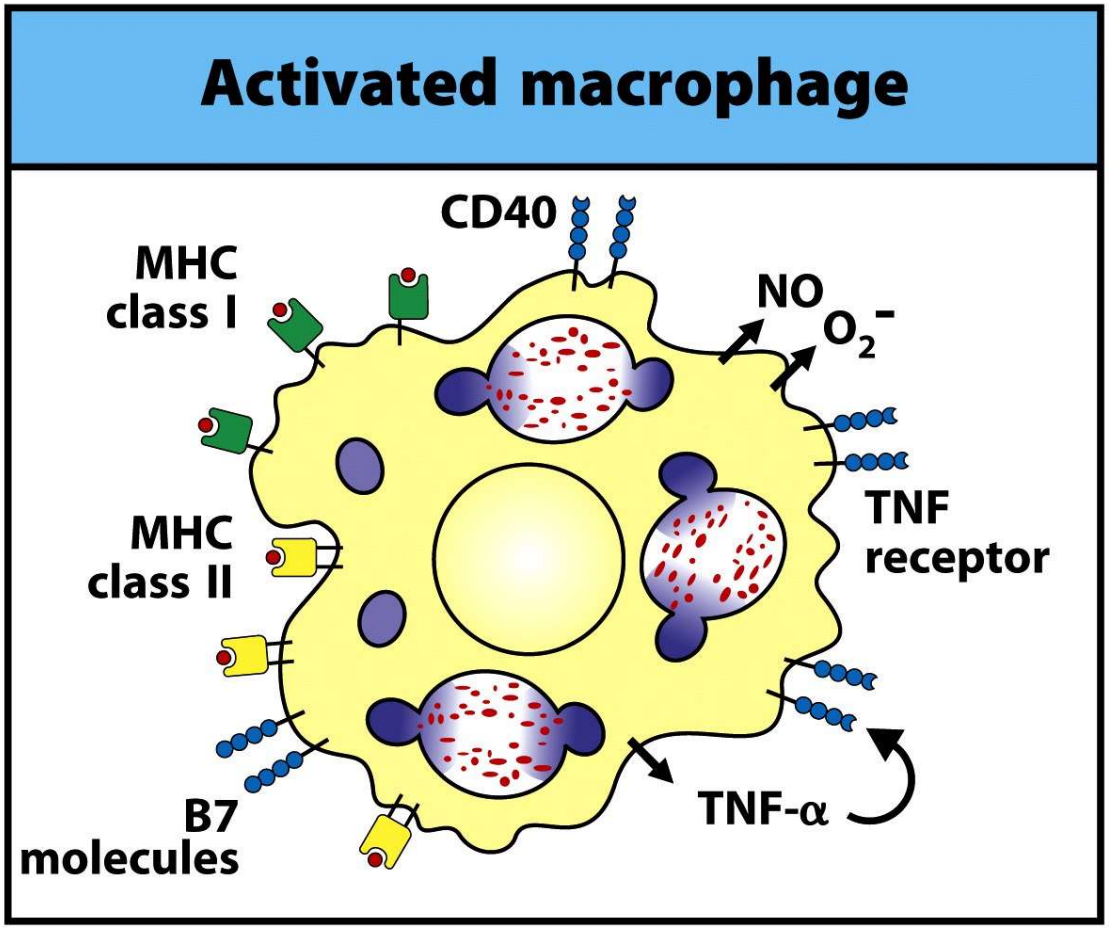

Figure 8-42 Immunobiology, 7ed. (© Garland Science 2008)