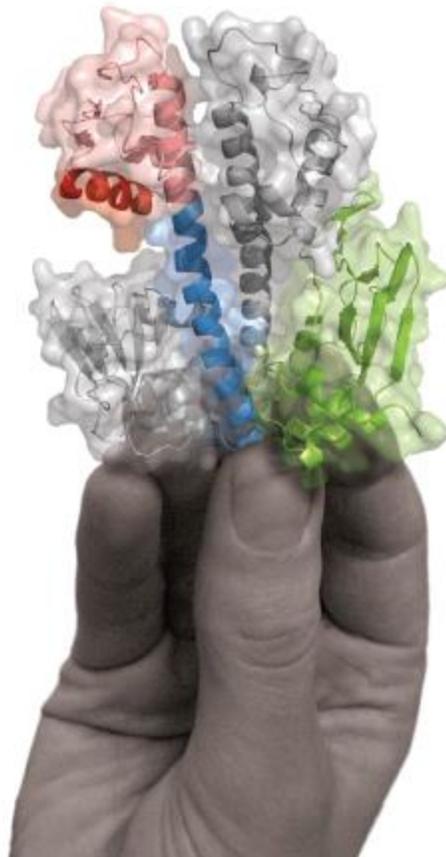




SAPIENZA  
UNIVERSITÀ DI ROMA



la Scienza a portata di mano



**Comunicazione  
delle  
Scienze Biomediche**

**Prof.ssa Cristina Cerboni**

*L'immunità innata:  
cellule dendritiche, cellule NK, granulociti  
(parte II)*

Anno Accademico 2023-2024

Il materiale presente in questo documento viene distribuito solamente per uso interno ed esclusivamente a scopo didattico.

# LE CELLULE DELL'IMMUNITA' INNATA

## I GRANULOCITI



Neutrophil	
Eosinophil	
Basophil	

## I MASTOCITI

Mast cell	
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## I MACROFAGI

Macrophage	
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## LE CELLULE DENDRITICHE



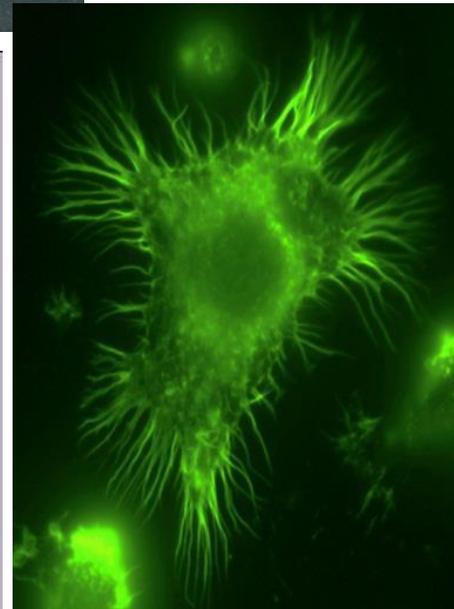
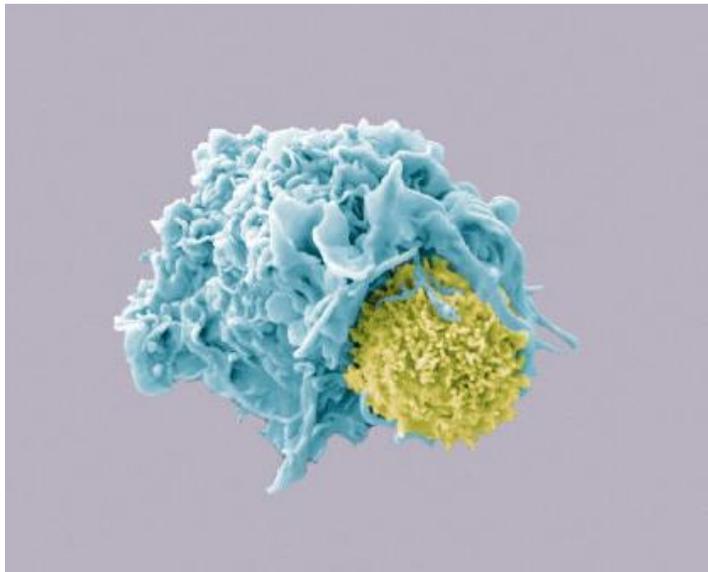
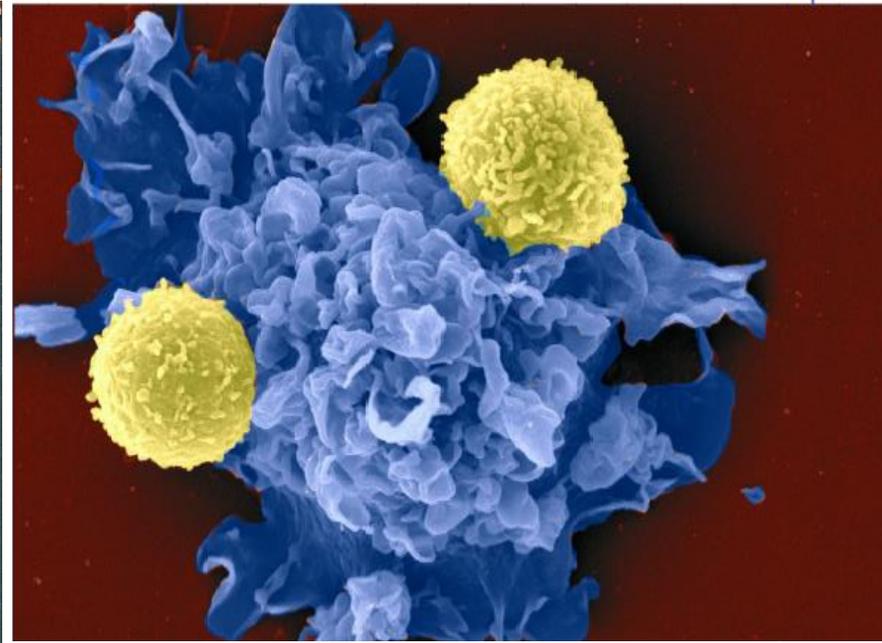
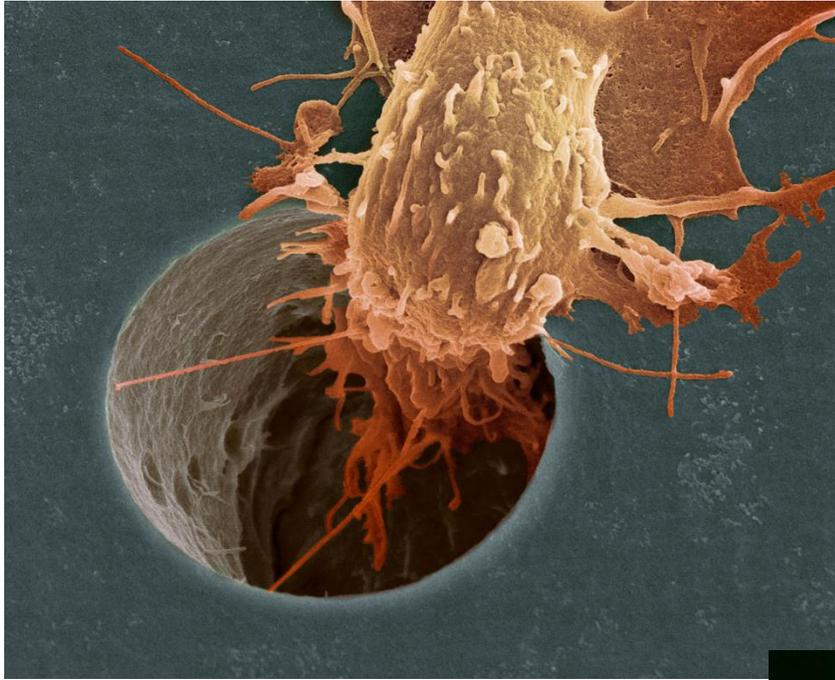
Dendritic cell	
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## LE CELLULE NATURAL KILLER (NK)

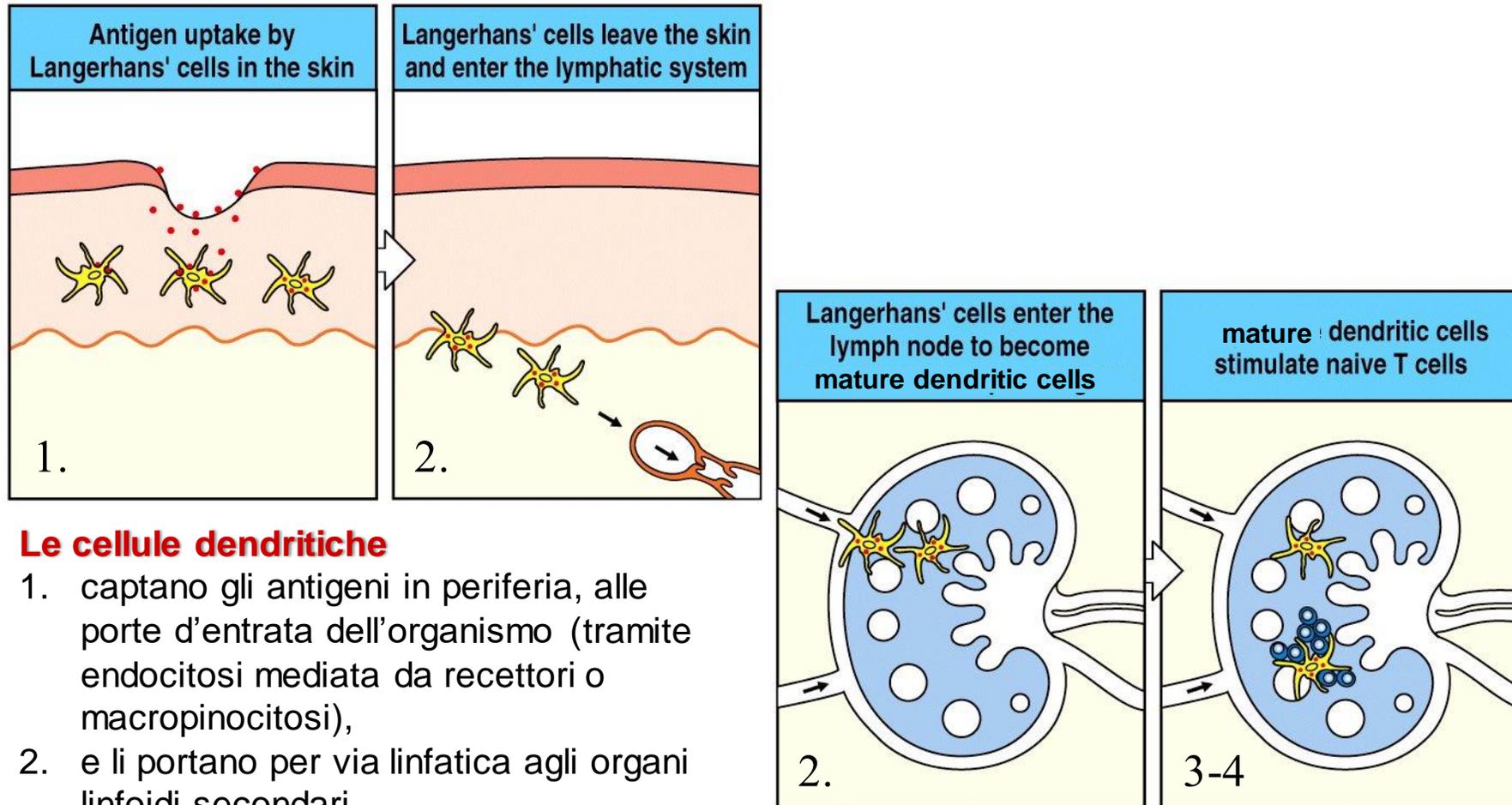
Natural killer (NK) cell	
--------------------------	--

# Le cellule dendritiche

dal greco “dendron”: albero

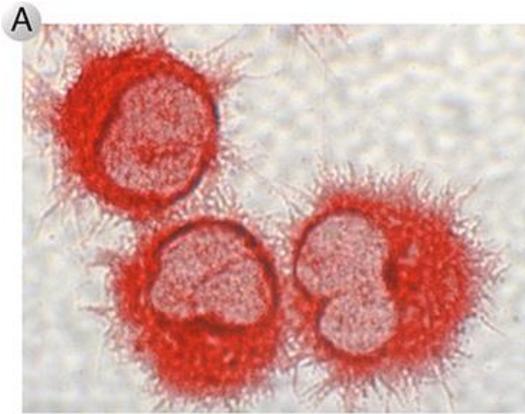


# Le cellule dendritiche

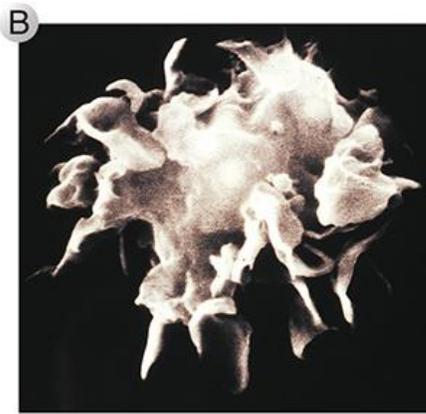


## Le cellule dendritiche

1. captano gli antigeni in periferia, alle porte d'entrata dell'organismo (tramite endocitosi mediata da recettori o macropinocitosi),
2. e li portano per via linfatica agli organi linfoidi secondari,
3. dove presentano l'antigene ai linfociti T,
4. e danno inizio alla risposta immunitaria acquisita.

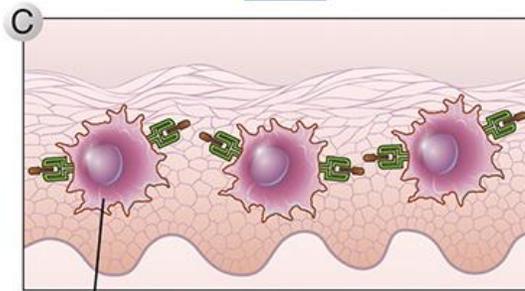


Cute

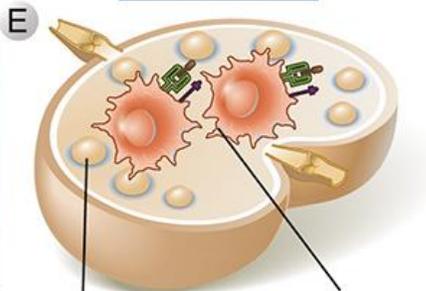
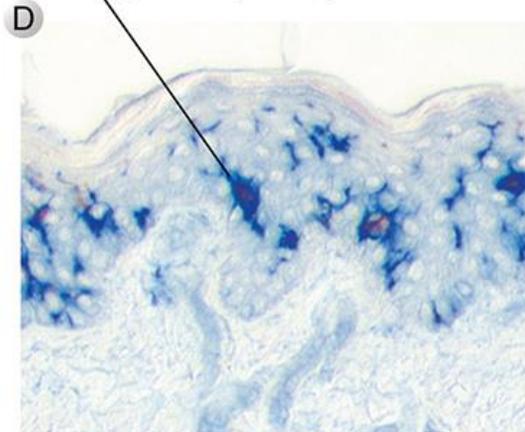


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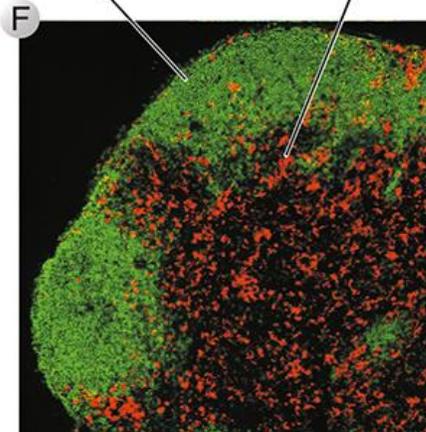
Linfonodo



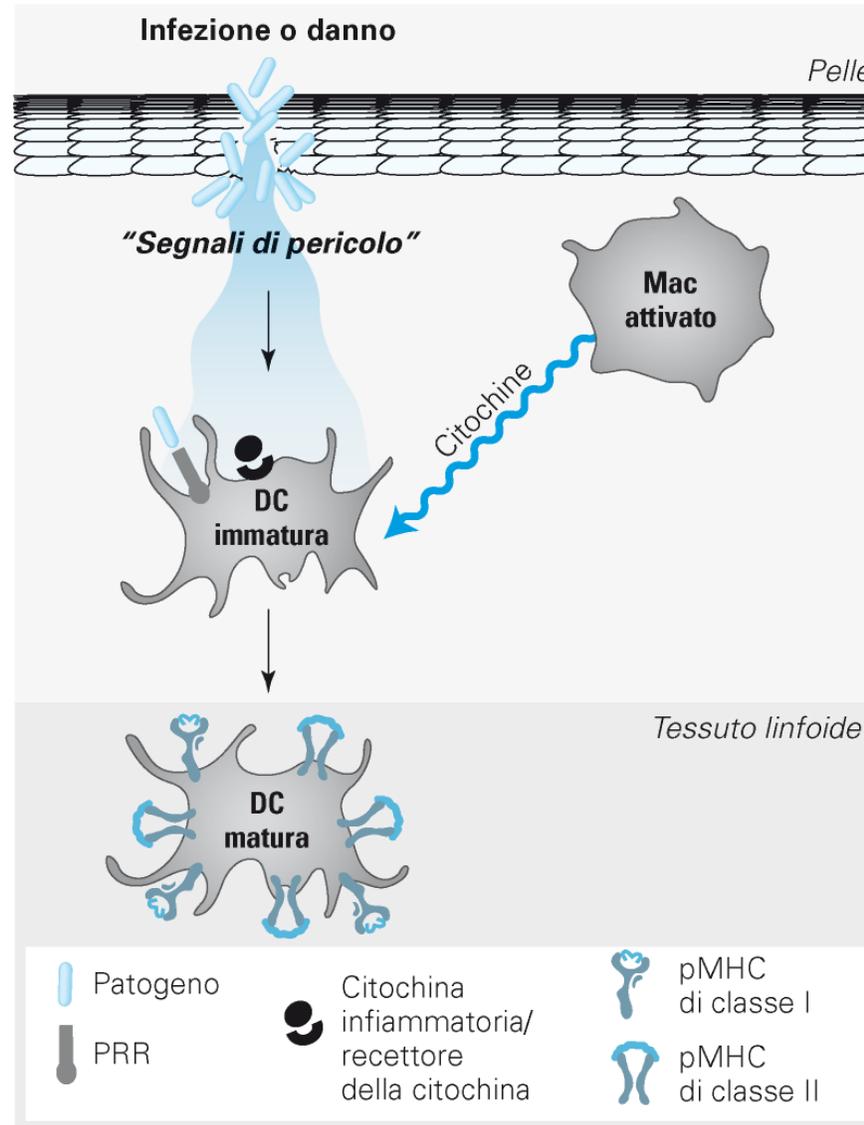
Cellula dendritica (cellula di Langerhans) nell'epidermide



Follicolo  
Cellula dendritica nel linfonodo



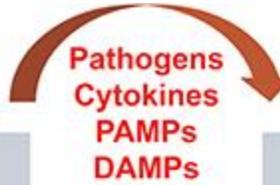
# Maturazione di una cellula dendritica



## Immature Dendritic Cells



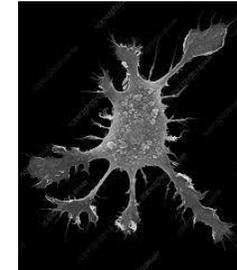
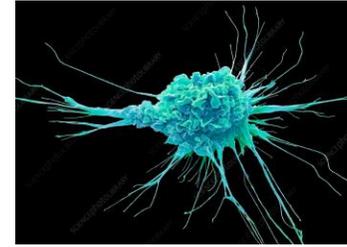
- ↓ Co-stimulatory molecules
- ↓ MHC II Expression
- ↓ Secretion of pro-inflammatory cytokines
- ↑ Endocytic/Uptake capacity
- ↓ CCR7 expression
- ↓ Glycolysis



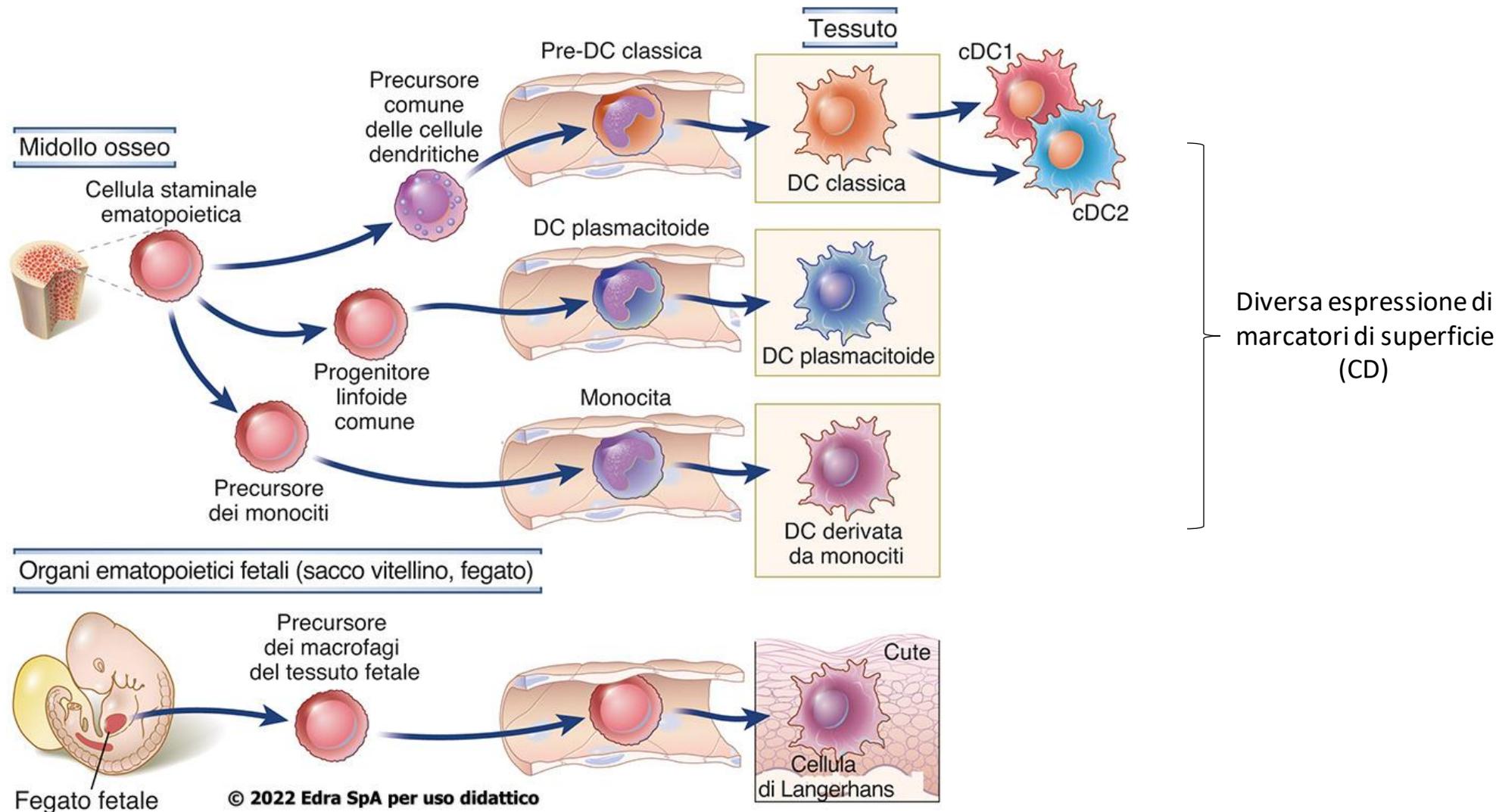
## Mature Dendritic Cells

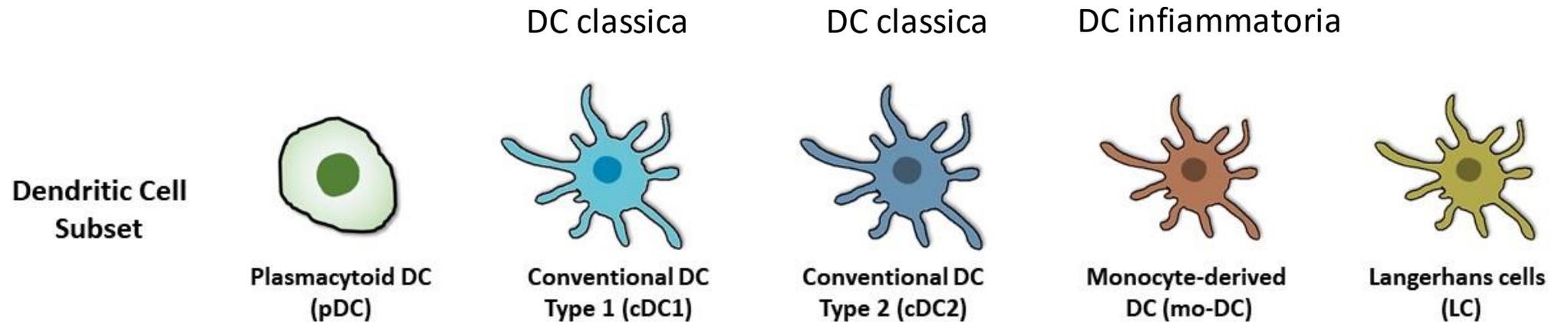


- ↑ Co-stimulatory molecules
- ↑ MHC II Expression
- ↑ Secretion of pro-inflammatory cytokines
- ↓ Endocytic/Uptake capacity
- ↑ CCR7 expression
- ↑ Glycolysis



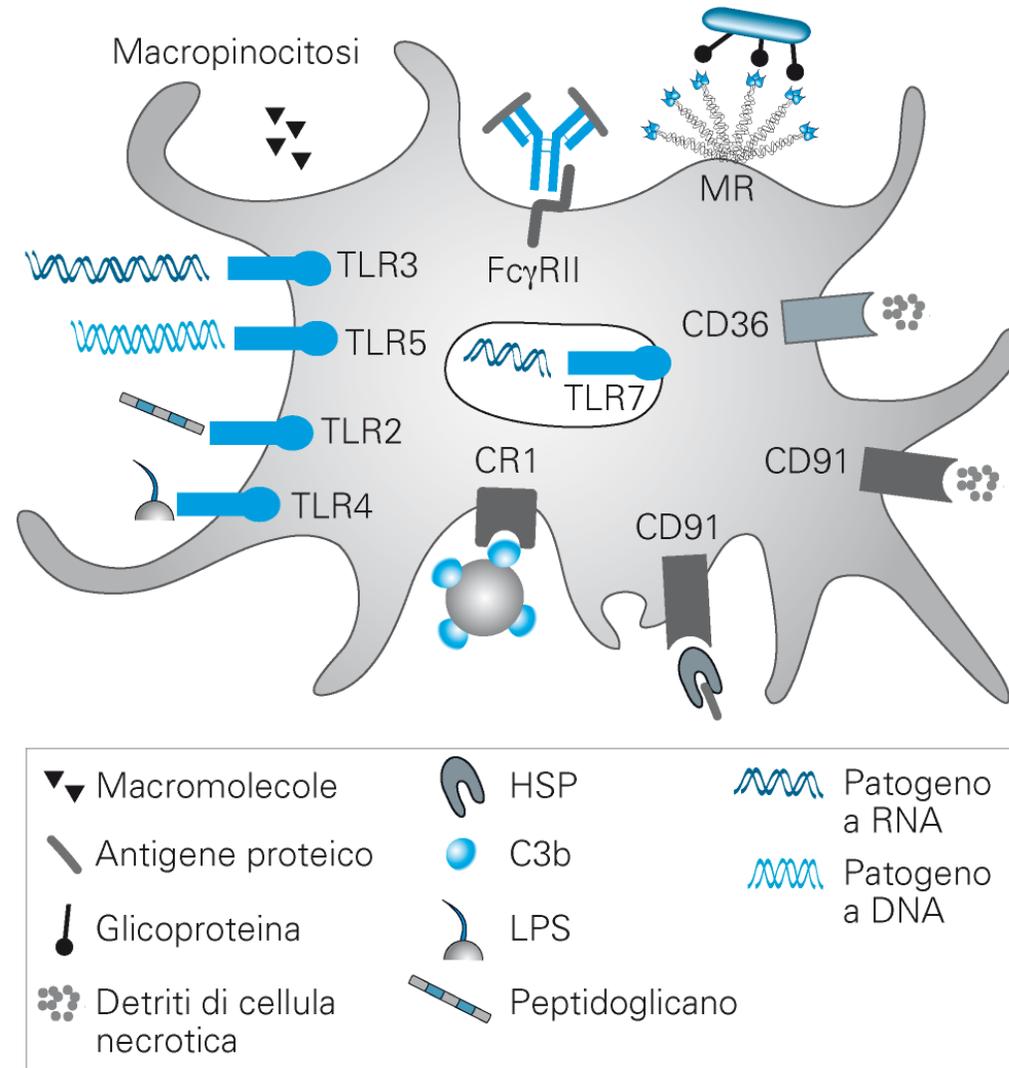
# Diversi tipi di cellule dendritiche



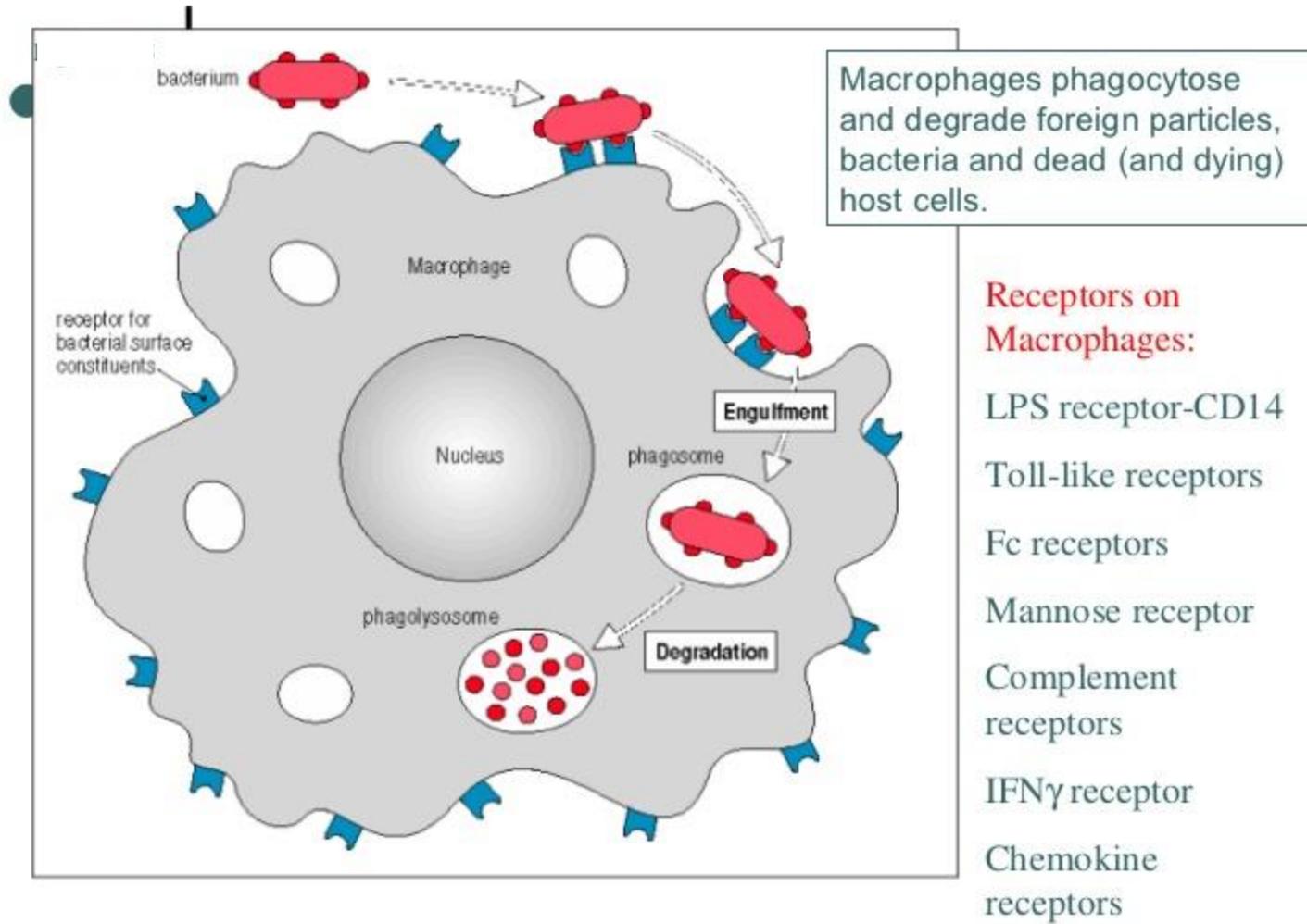


Description	Plasma cell-like morphology. Rapid production of type I and type III interferons for viral infections.	Notable for cross-presentation, effective in priming CD8+ T cells against extracellular antigens.	Dominant DC subset in blood, tissues, and lymphoid organs. Robust repertoire of pattern recognition receptors.	Differentiates from monocytes during inflammation. Has dendritic cell morphology.	Macrophages in origin, but with dendritic cell-like phenotype. Resides in squamous epithelium.
Cell Markers	CCR7, CD45RA, CD209, CLEC4C, LILRB4, NRP1, B220, SiglecH, IFN- $\alpha$	BTLA, CADM1, CD8A, CLEC9A, ITGAE, ITGAX, LY75, THBD, XCR1	CD14, CD163, CLEC10A, NOTCH2, ITGAM, SIRPA, CX3CR1, CD1C, CD2	CD14, CD1A, CD1C, CD209, FCER1, ITGAM, MRC1, SIRPA	CD1A, CD207, ID2
Transcription Factors	IRF4, IRF7, IRF8	BATF3, ID2, IRF8, ZBTB46	ID2, IRF4, KLF4, ZBTB46	IRF4, KLF4, ZBTB46	

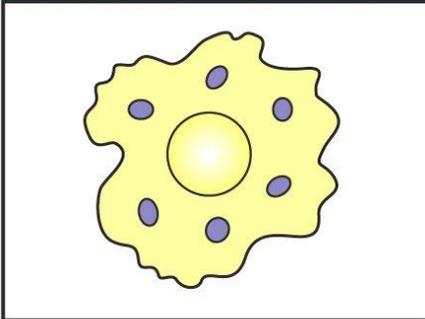
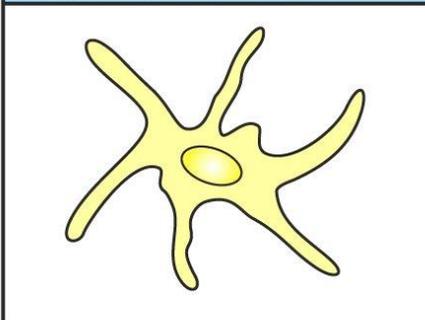
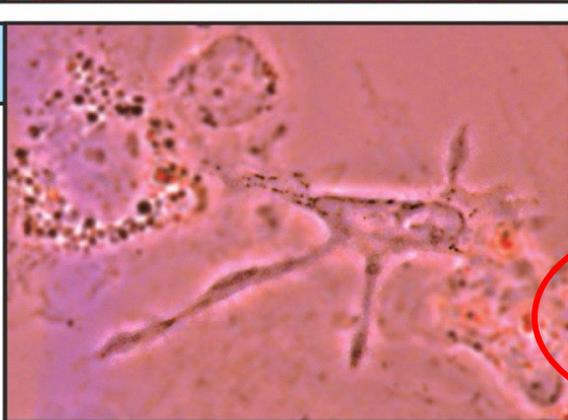
# Esempi di meccanismi per catturare l'antigene in una cellula dendritica...



# ... e in un macrofago!



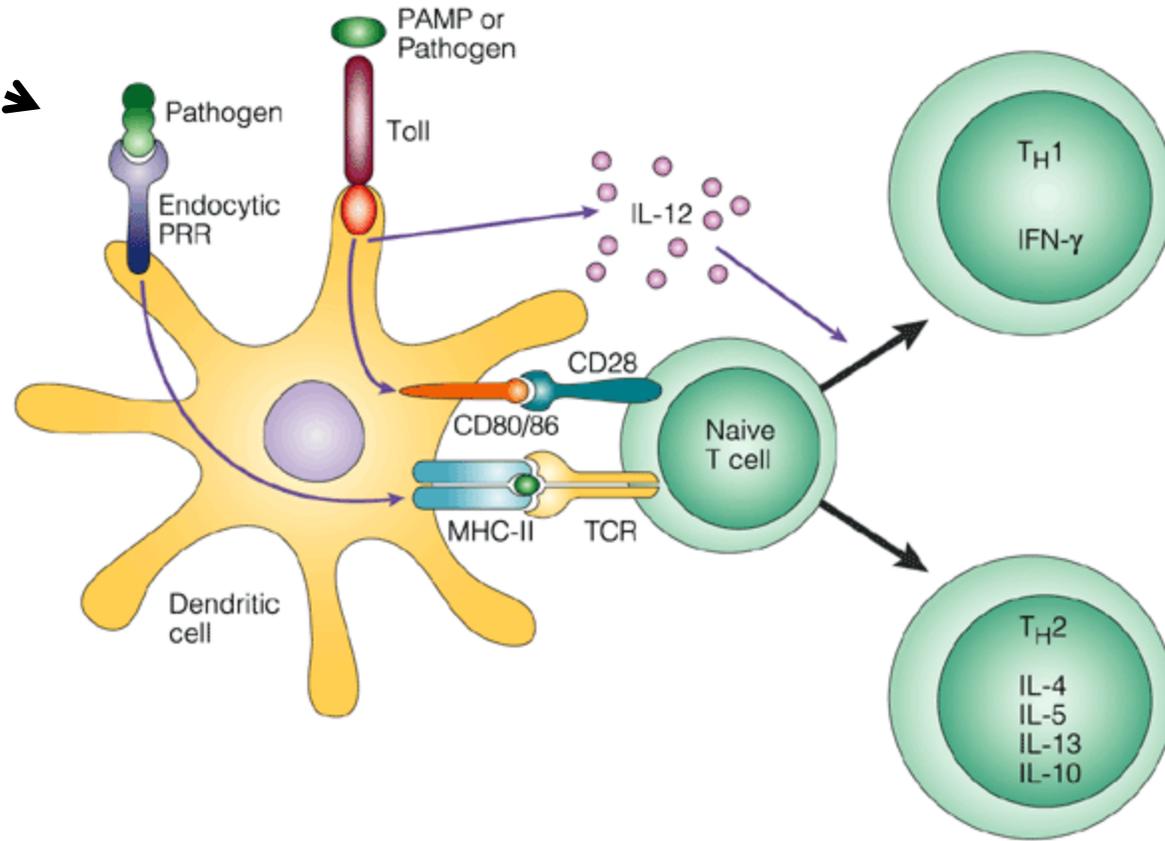
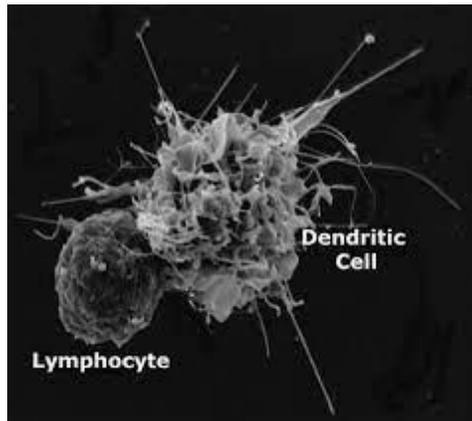
## Oltre ai macrofagi anche le cellule dendritiche funzionano da cellule accessorie presentanti l'antigene...

Cell	Activated function
<p data-bbox="614 344 1039 439"><b>Macrophage</b></p> 	 <p data-bbox="1607 344 1931 601">Phagocytosis and activation of bactericidal mechanisms</p> <p data-bbox="1607 601 1931 758">Antigen presentation</p>
<p data-bbox="614 758 1039 858"><b>Dendritic cell</b></p> 	 <p data-bbox="1607 758 1931 958">Antigen uptake in peripheral sites</p> <p data-bbox="1607 958 1931 1178">Antigen presentation in lymph nodes</p>

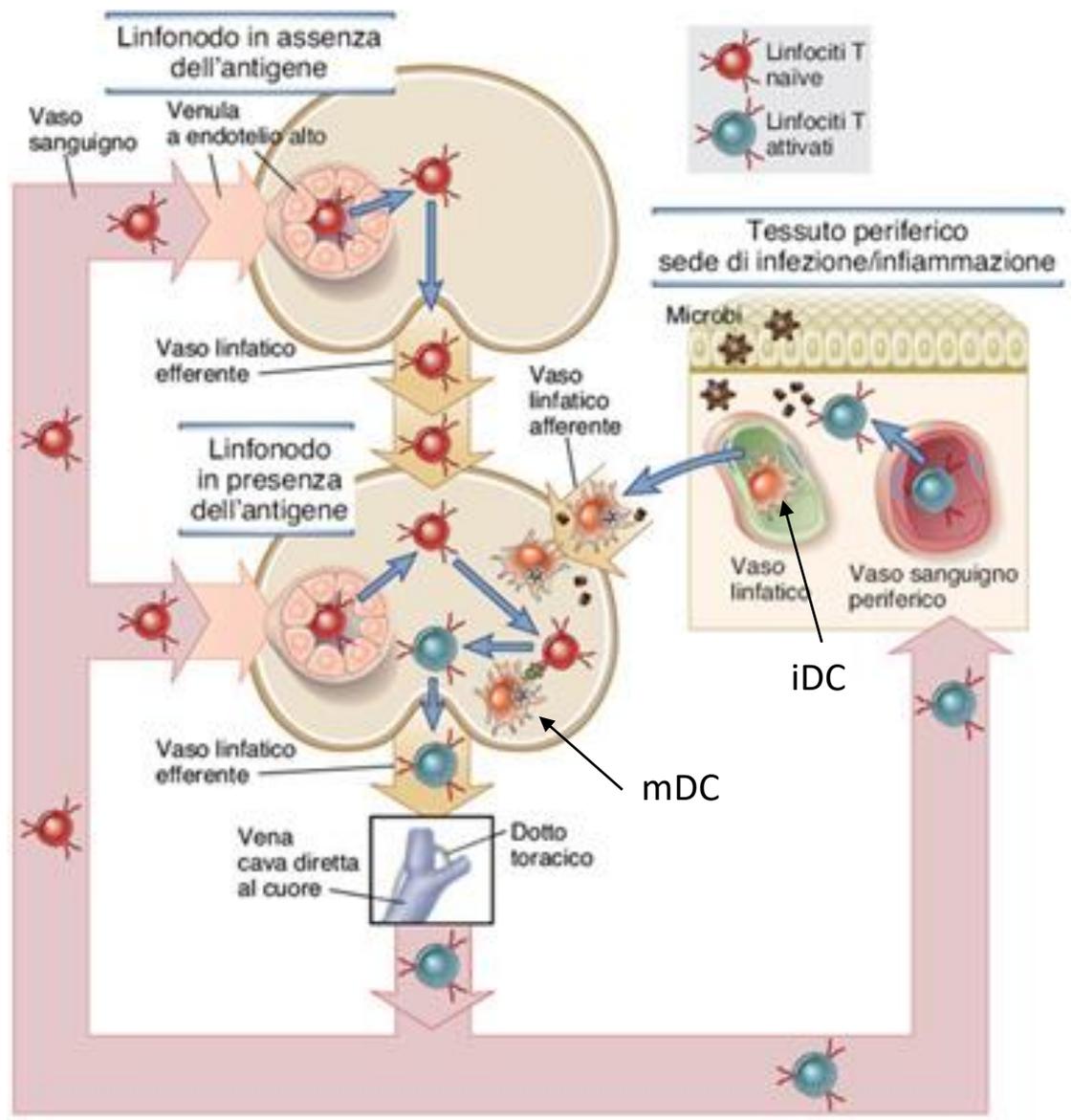
... e **ATTIVANO** e **ISTRUISCONO** i linfociti dell'immunità adattativa!



# ... e ATTIVANO e ISTRUISCONO i linfociti dell'immunità adattativa!

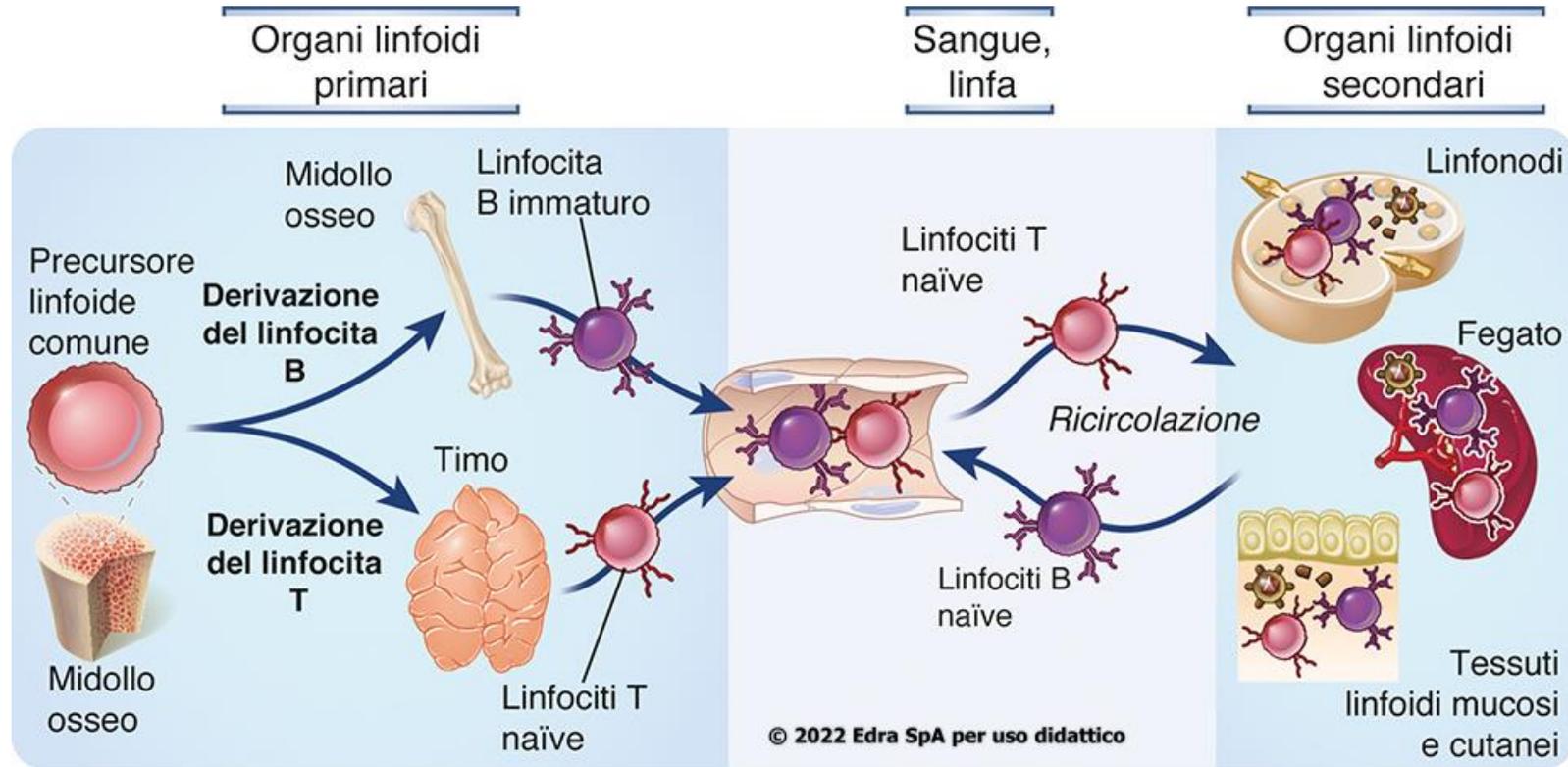


# e ATTIVANO e ISTRUISCONO i linfociti dell'immunità adattativa!

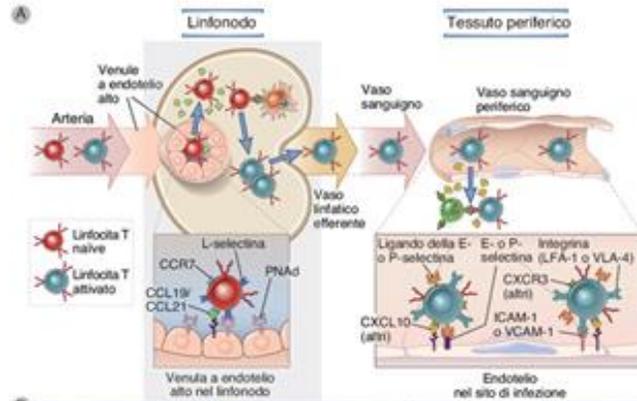


Le riprenderemo nell'immunità adattativa!

# I linfociti circolano!



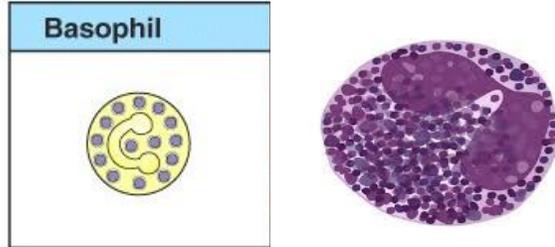
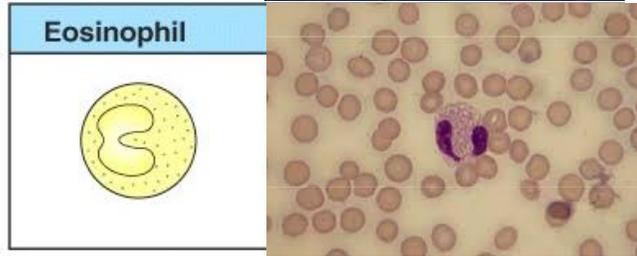
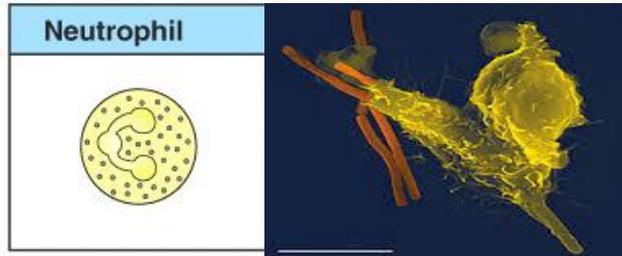
# Il reclutamento di linfociti e cellule dendritiche è finemente regolato!



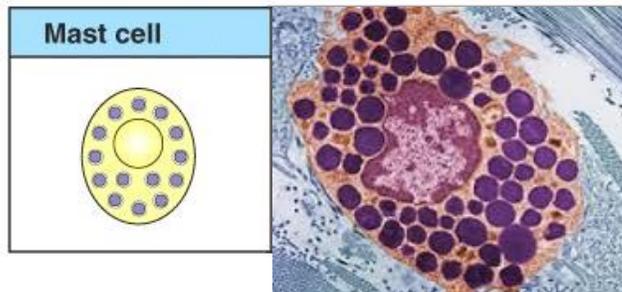
Recettore di homing dei linfociti T	Ligando sulla cellula endoteliale	Funzione dell'interazione recettore: ligando
<b>Linfociti T naïve</b>		
L-selectina	PNAd	Adesione iniziale debole dei linfociti T naïve alle venule a endotelio alto nel linfonodo
CCR7	CCL19 o CCL21	Attivazione delle integrine e chemotassi
LFA-1 ( $\beta 2$ -integrina)	ICAM-1	Arresto sulle cellule a endotelio alto nel linfonodo
<b>Linfociti T attivati (effettori e della memoria)</b>		
Ligando della E- e P-selectina	E- e P-selectina	Adesione iniziale debole dei linfociti T effettori e della memoria all'endotelio attivato dalle citochine nel sito periferico di infezione
CXCR3	CXCL10 (atrr)	Attivazione di integrine o chemotassi
CCR5	CCL4 (atrr)	Attivazione di integrine e chemotassi
LFA-1 ( $\beta 2$ -integrina) o VLA-4 ( $\beta 1$ -integrina)	ICAM-1 or VCAM-1	Arresto saldo su endotelio attivato da citochine nei siti di infezione

# LE CELLULE DELL'IMMUNITA' INNATA

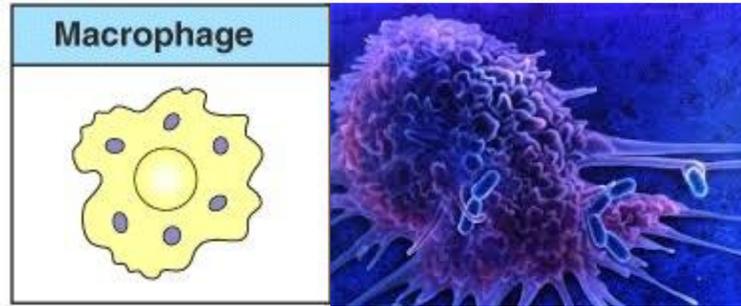
## I GRANULOCITI



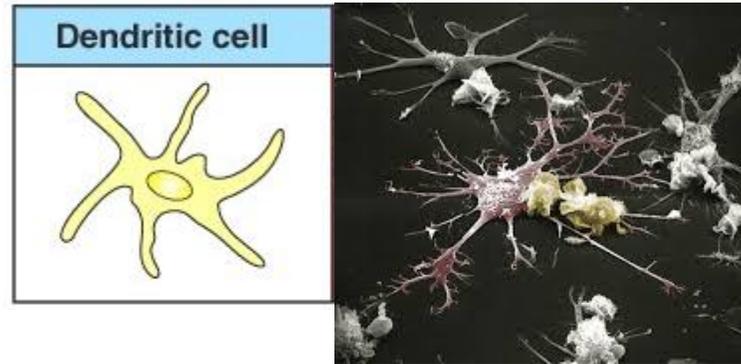
## I MASTOCITI



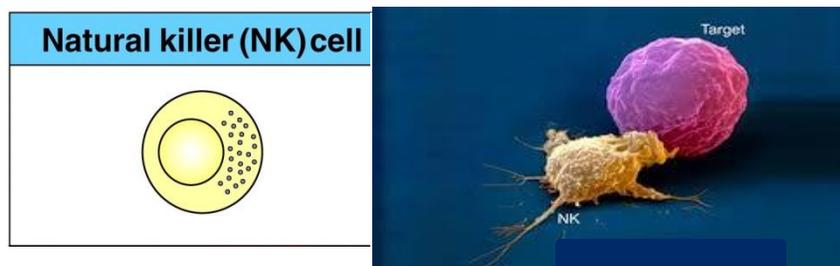
## I MACROFAGI



## LE CELLULE DENDRITICHE

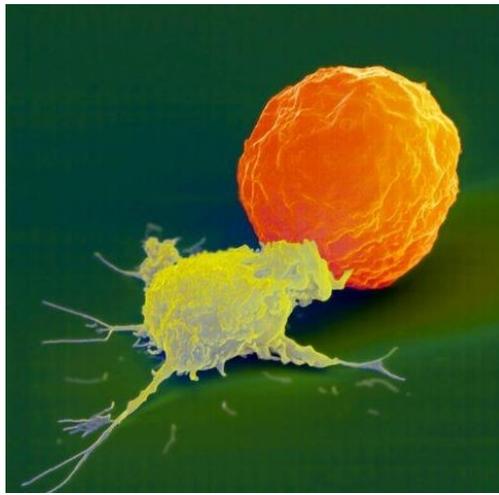
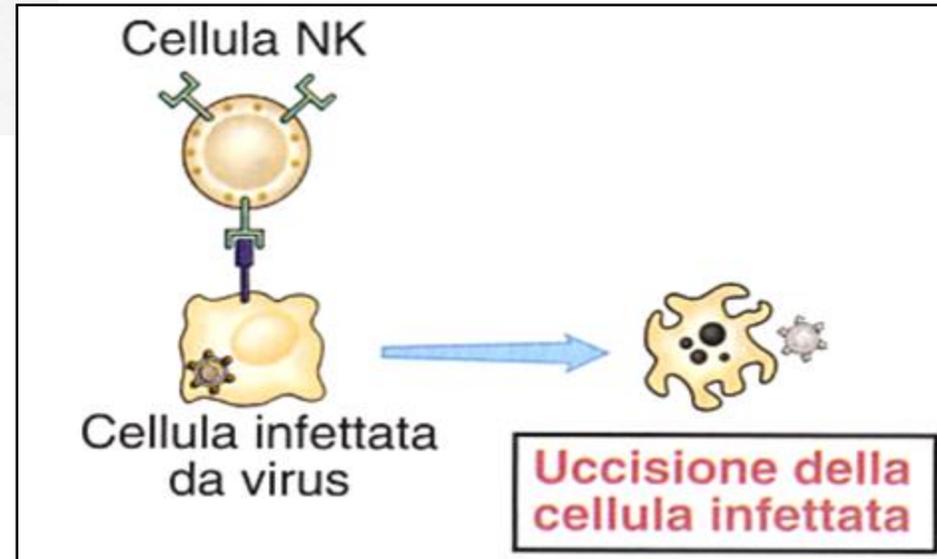
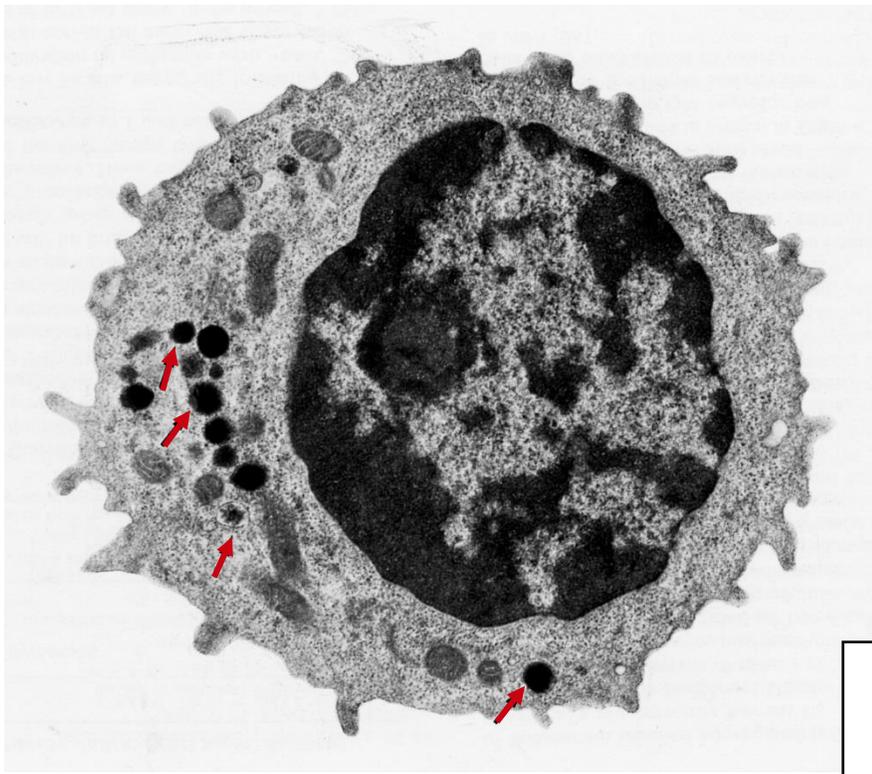


## LE CELLULE NATURAL KILLER (NK)



# CELLULE EFFETTRICI DELL'IMMUNITA' INNATA:

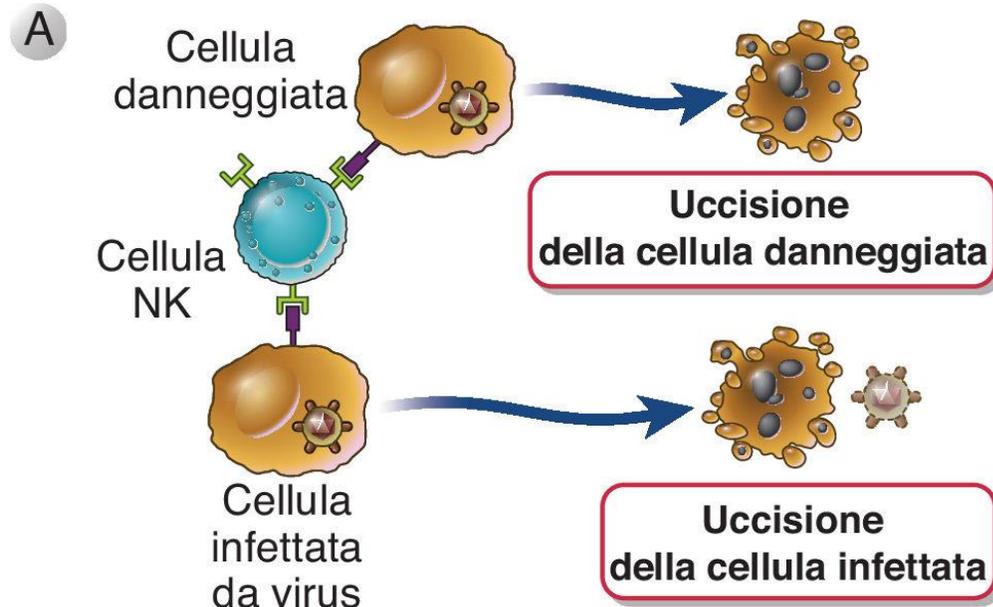
## LE CELLULE NATURAL KILLER (NK)



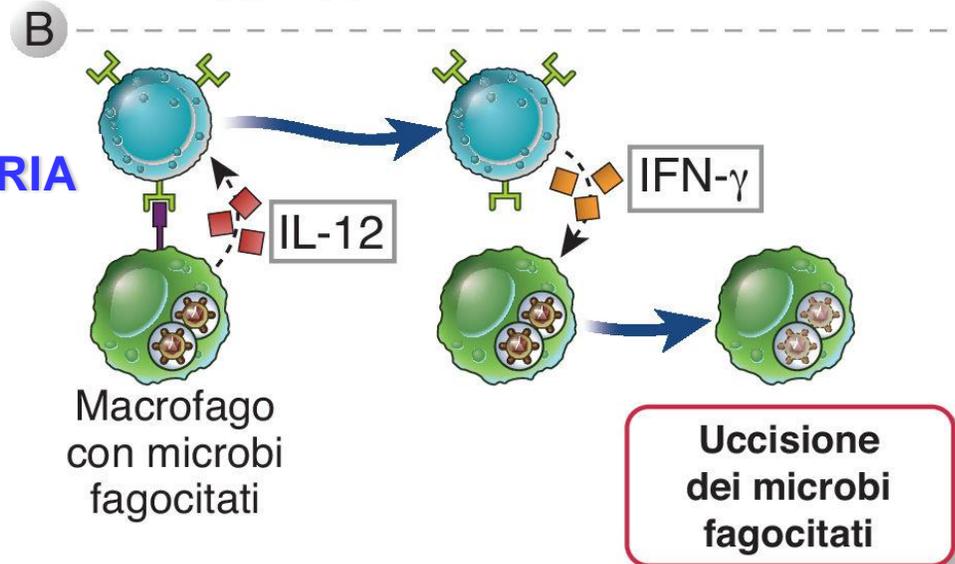
*Una cellula Natural Killer (in giallo)  
attacca una cellula infettata da virus  
(in arancione)*

# LE FUNZIONI DELLE CELLULE NK

CITOTOSSICA



IMMUNOREGOLATORIA



# LE FUNZIONI DELLE CELLULE NK

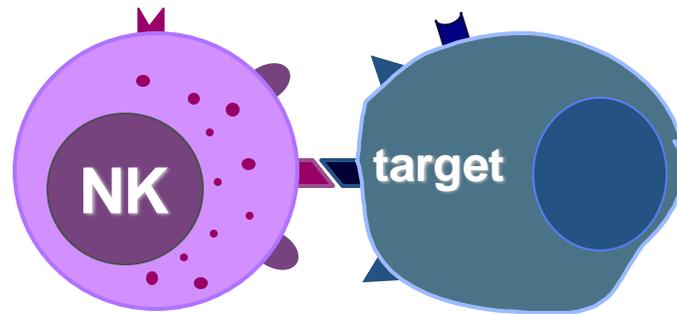
- **Citotossicità naturale**

- costitutiva
- non MHC-ristretta
- priva di memoria immunologica

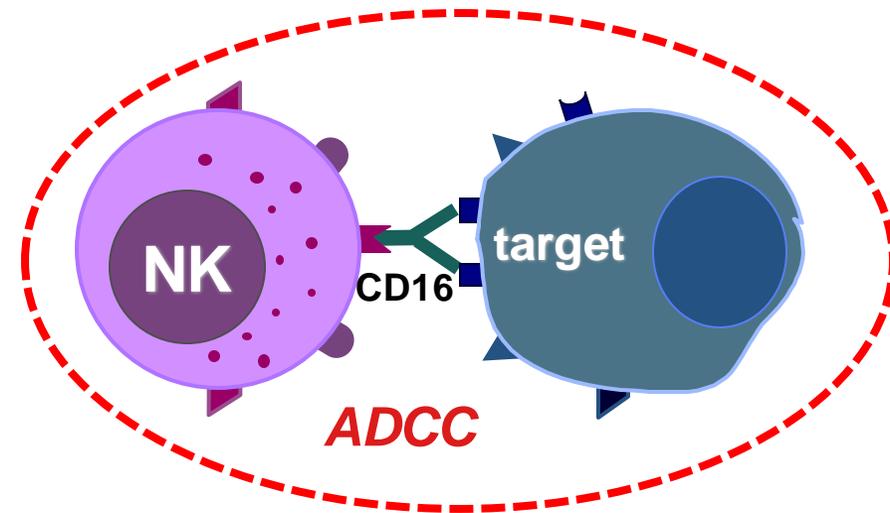
→ cellule tumorali  
cellule infettate da virus  
cellule emopoietiche immature

- **Citotossicità anticorpo-dipendente (ADCC)**

→ cellule ricoperte da anticorpi (IgG)



*Citotossicità naturale*



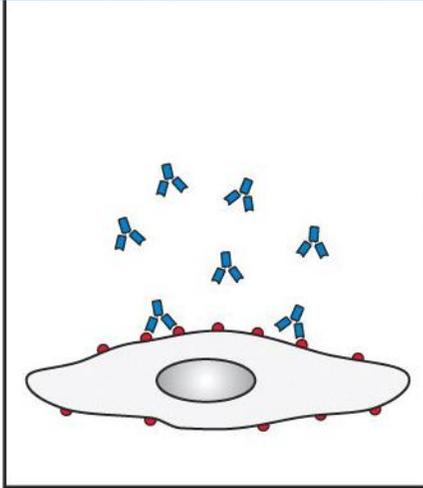
*ADCC*

- **Produzione di mediatori solubili:** le cellule NK producono anche molte citochine e chemochine!  
**IFN- $\gamma$ , TNF- $\alpha$ , IL-5, GM-CSF, TGF- $\beta$ , CCL3/MIP1 $\alpha$ , CCL4/MIP1 $\beta$ , CCL5/RANTES**

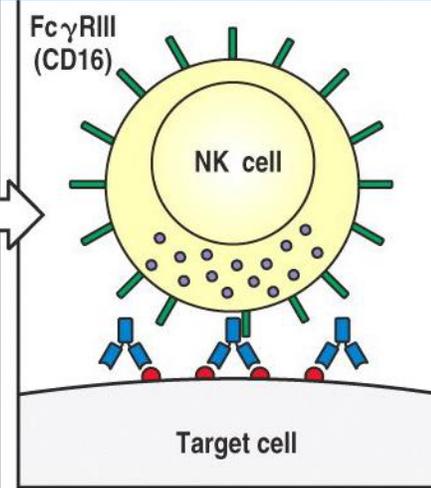
# Gli anticorpi inducono la citotossicità anticorpo-dipendente (ADCC) delle cellule NK...

ADCC: Antibody-Dependent Cell Cytotoxicity

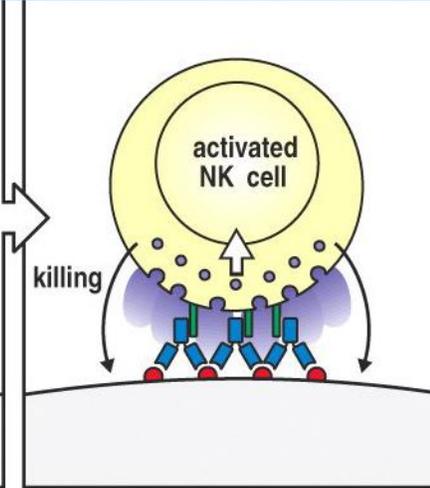
**1** L'anticorpo si lega agli antigeni sulla superficie della cellula bersaglio



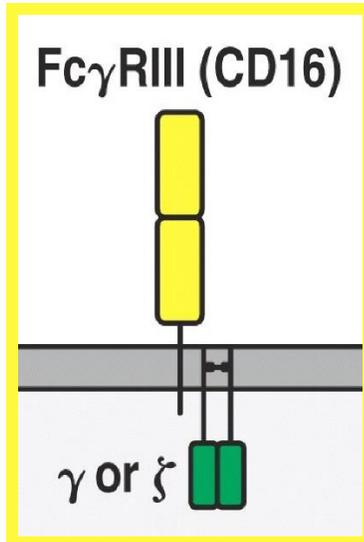
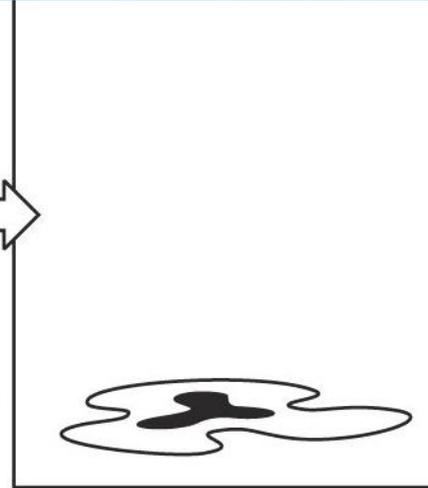
**2** I recettori Fc espressi sulle cellule NK riconoscono l'anticorpo legato



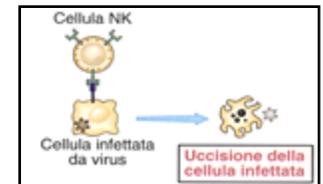
**3** Il crosslinking dei recettori per Fc segnala alle cellule NK di uccidere le cellule bersaglio



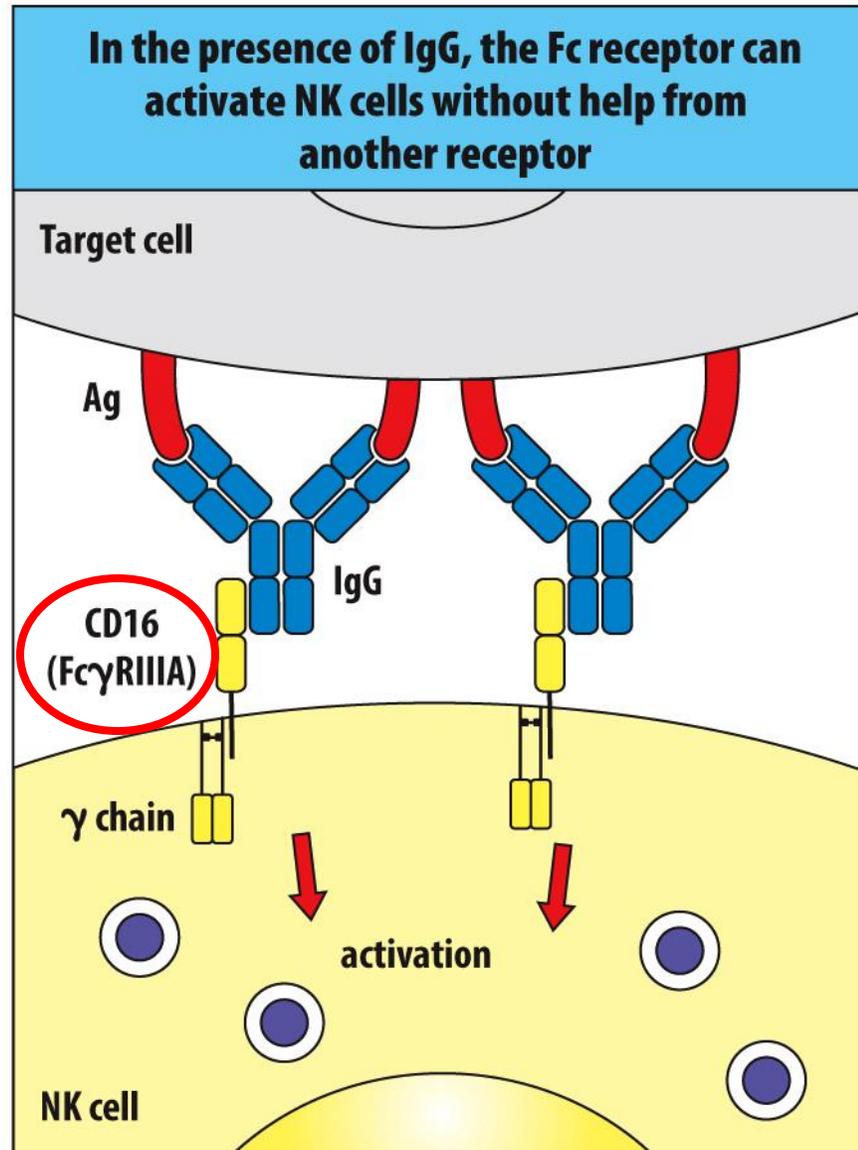
**4** La cellula bersaglio muore per apoptosi e/o danno alla membrana



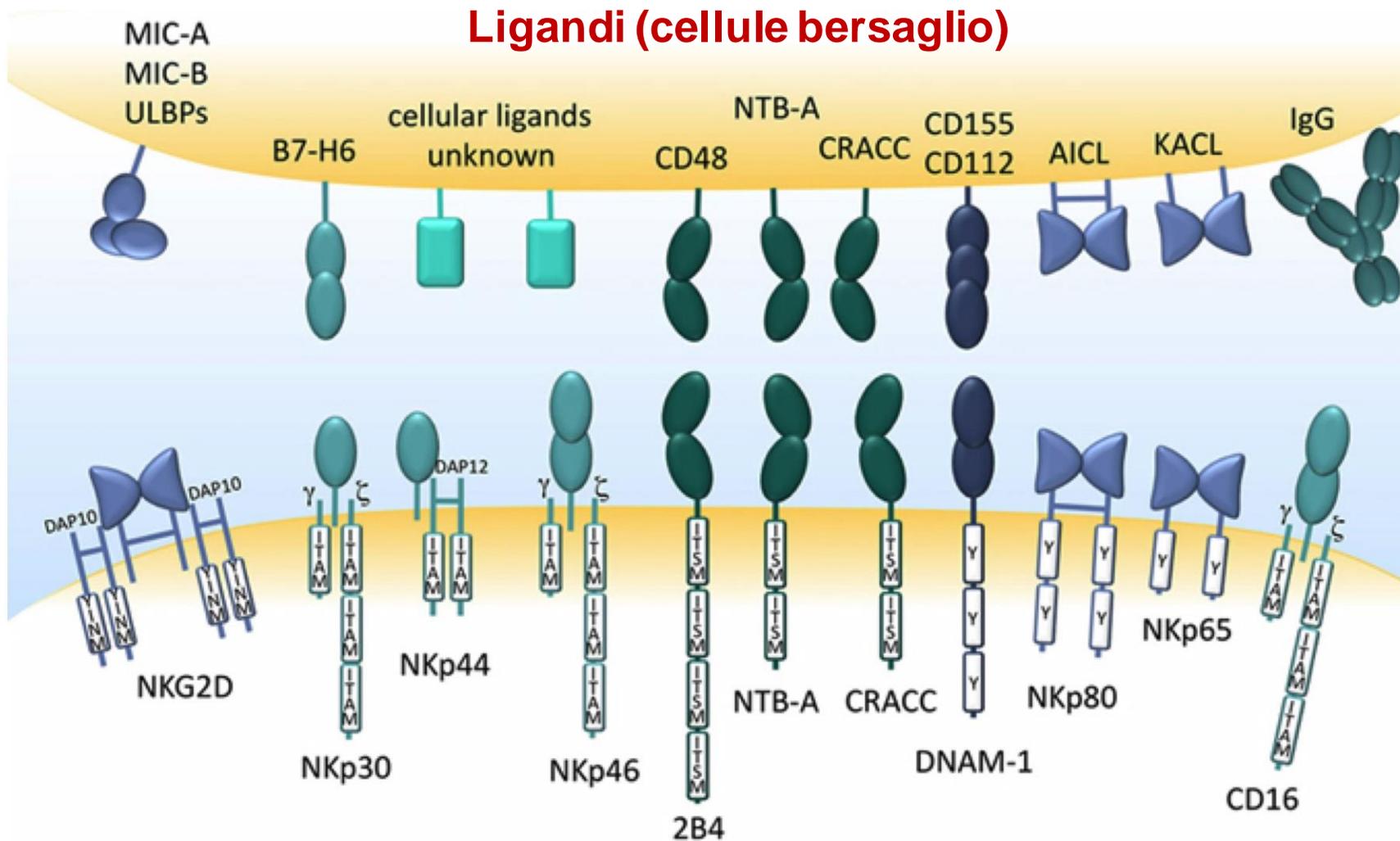
...mediante il CD16, un recettore per le IgG (FcγRIII)



# Il CD16 è in grado di promuovere da solo l'attivazione delle cellule NK



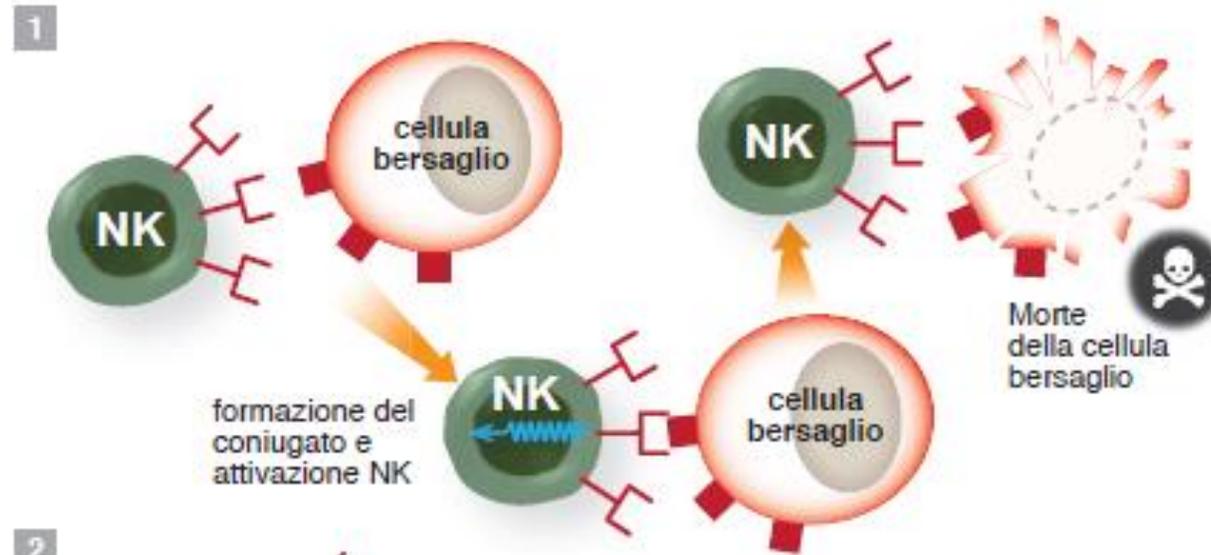
# ... MA I RECCTORI ATTIVATORI DELLE CELLULE NK SONO MOLTI!!!



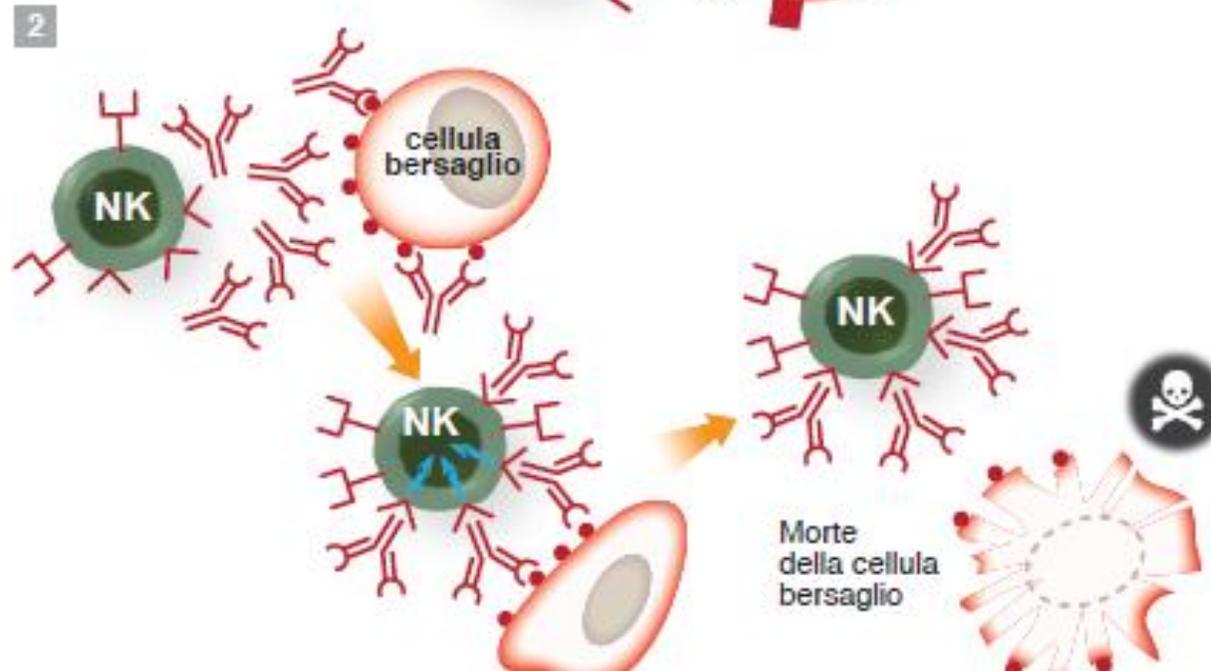
Reccettori (cellule NK)

# LA CITOTOSSICITÀ DELLE CELLULE NK

LA CITOTOSSICITÀ  
“NATURALE”



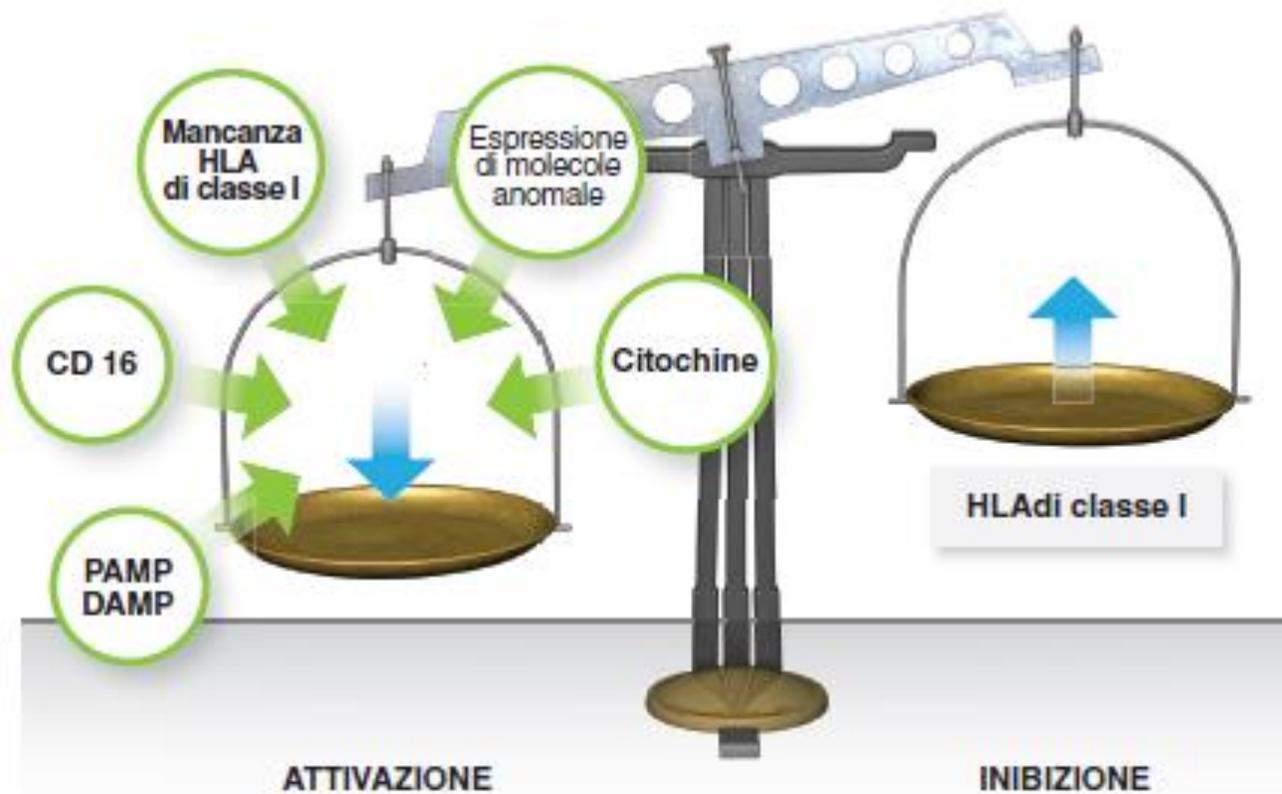
LA ADCC



# LA BILANCIA DELL'ATTIVAZIONE DELLE CELLULE NK

## RECETTORI ATTIVATORI

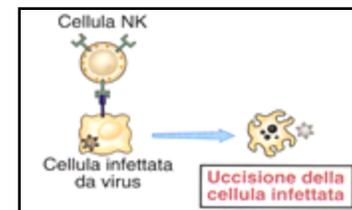
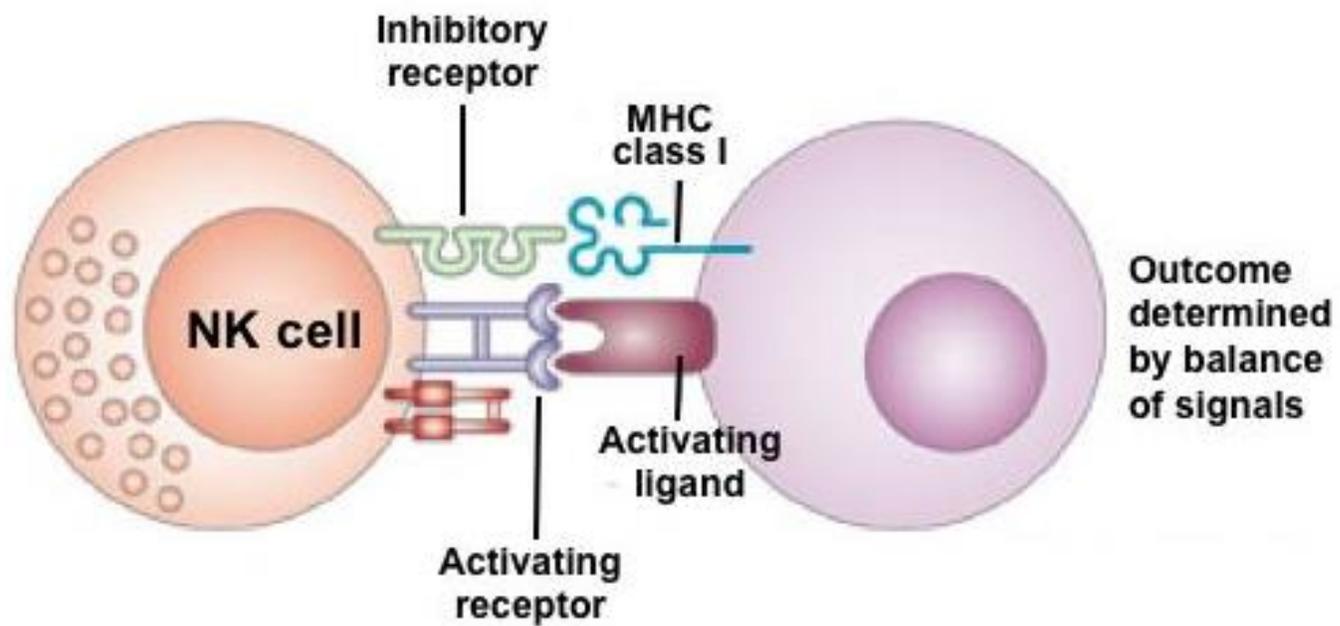
CD16  
NCR  
TLRs  
KIRs  
NKG2D  
DNAM-1  
....



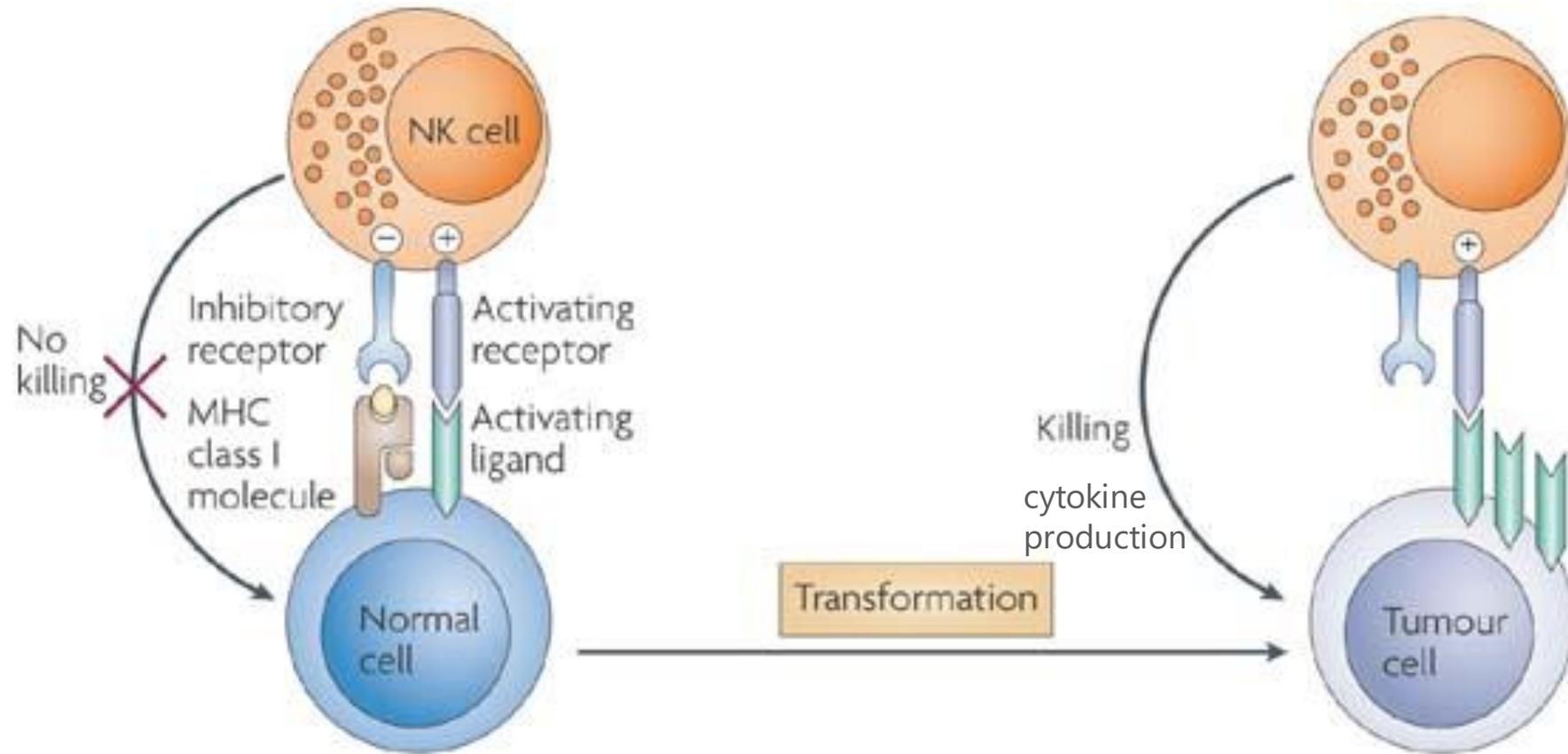
## RECETTORI INIBITORI

KIRs  
CD94/NKG2A  
PD-1  
...

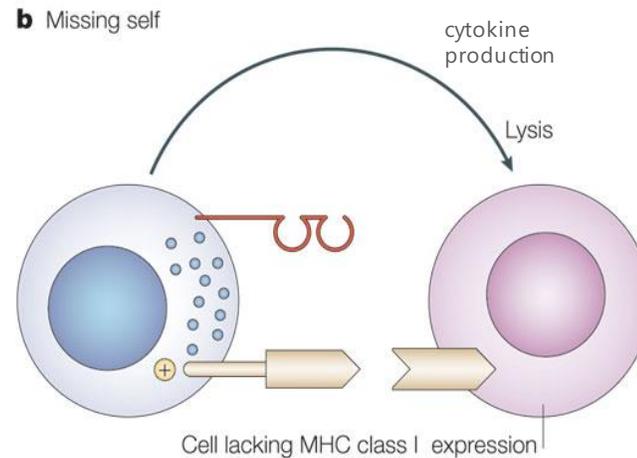
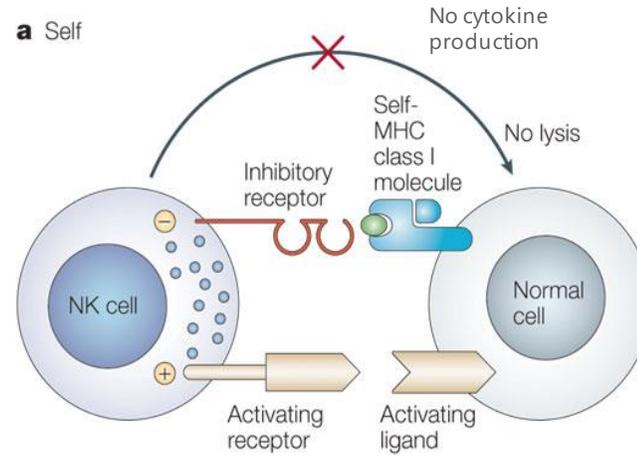
## LE CELLULE NK SONO REGOLATE DA UN EQUILIBRIO DI SEGNALI ATTIVATORI E INIBITORI



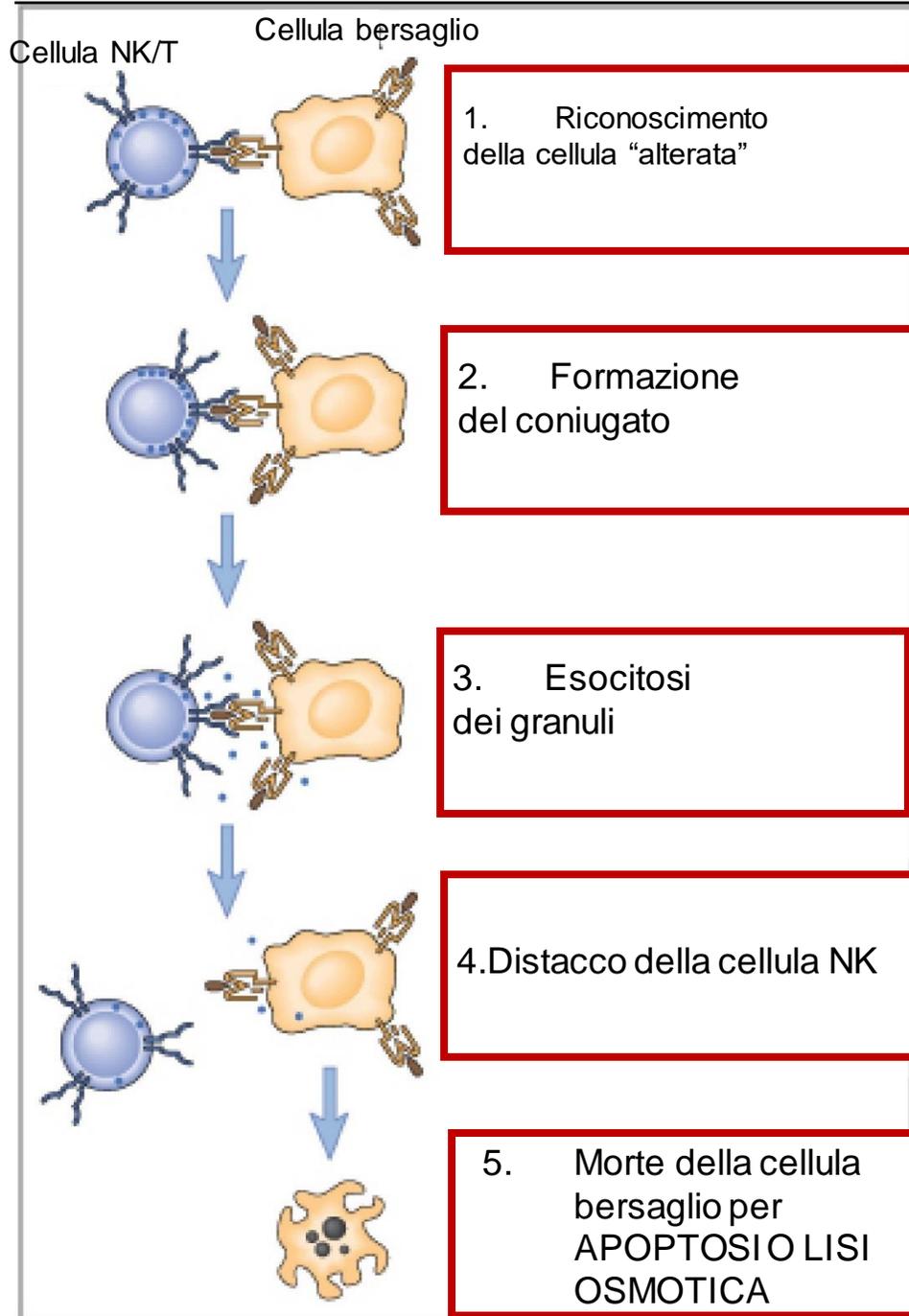
# IL "MISSING-SELF"



# LE CELLULE NK SONO IN GRADO DI DISTINGUERE LA PRESENZA O L'ASSENZA DI MOLECOLE MHC DI CLASSE I SULLA CELLULA BERSAGLIO

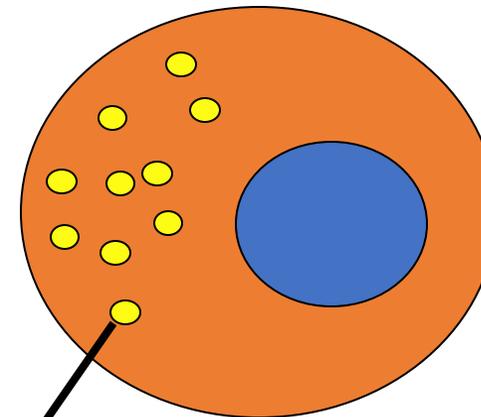


La perdita delle molecole MHC di classe I attiva l'uccisione delle cellule bersaglio (es. infettate o tumorali)



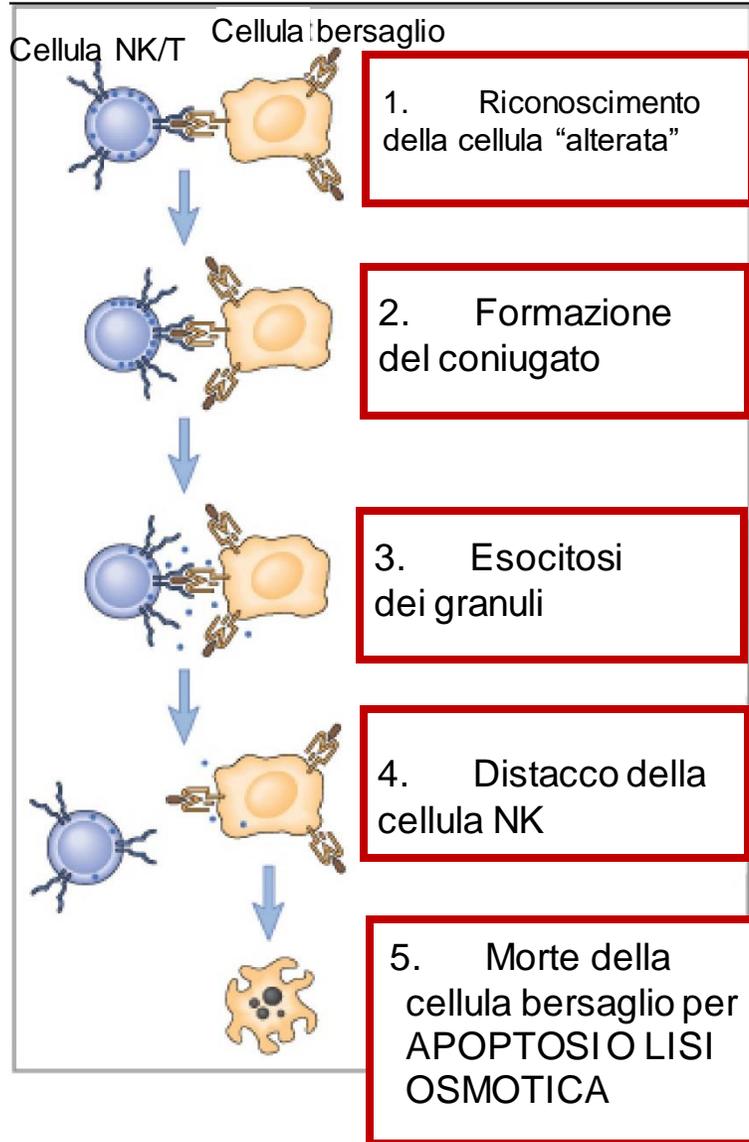
## LE DIVERSE FASI DELLA CITOTOSSICITA' LINFOCITARIA

*CELLULA NK*

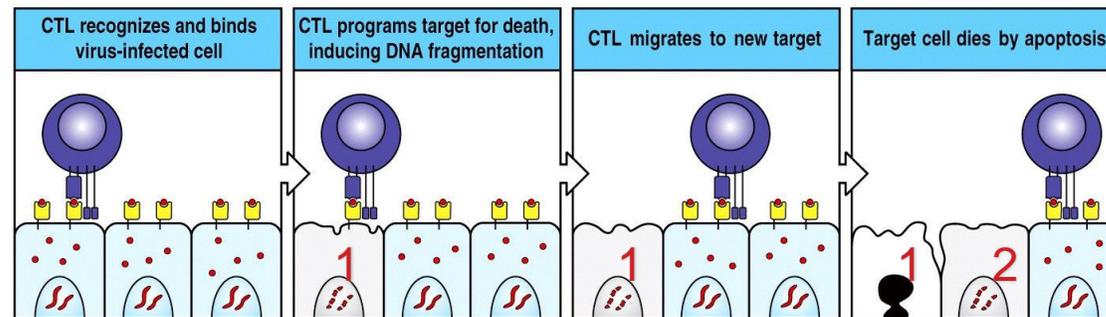


Granuli contenenti perforine e granzimi

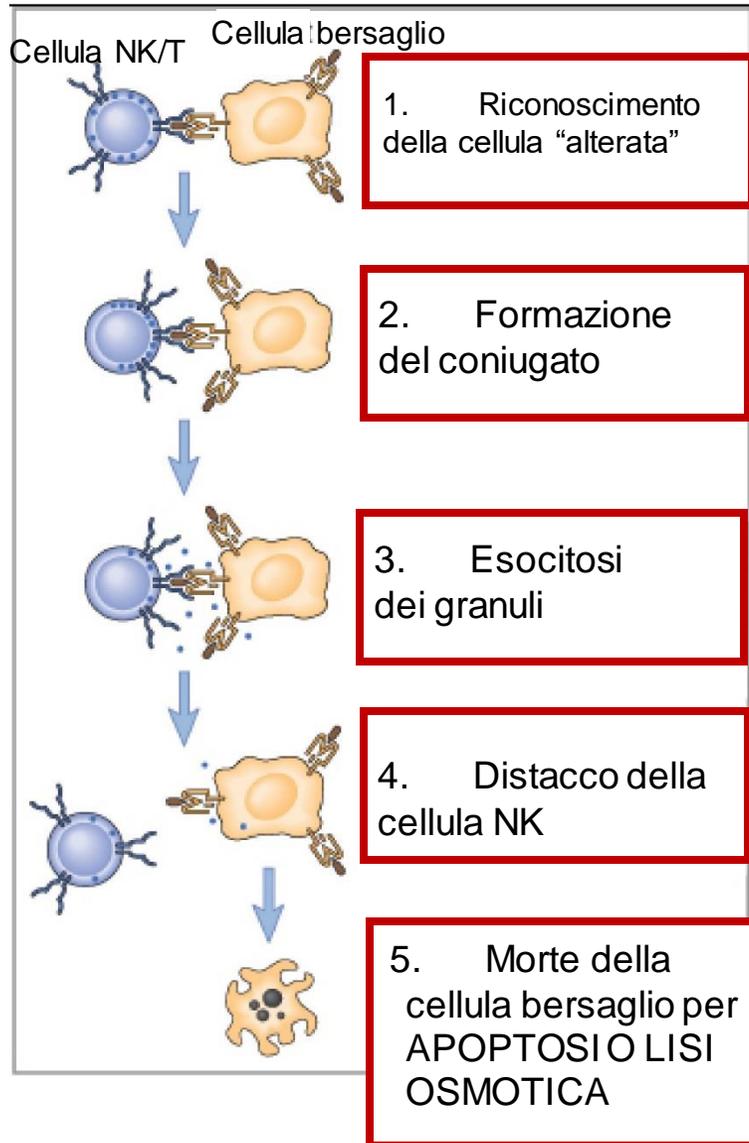
## Le diverse fasi della citotossicità



**Le cellule NK (e i linfociti T citotossici) inducono la morte per apoptosi della cellula bersaglio**



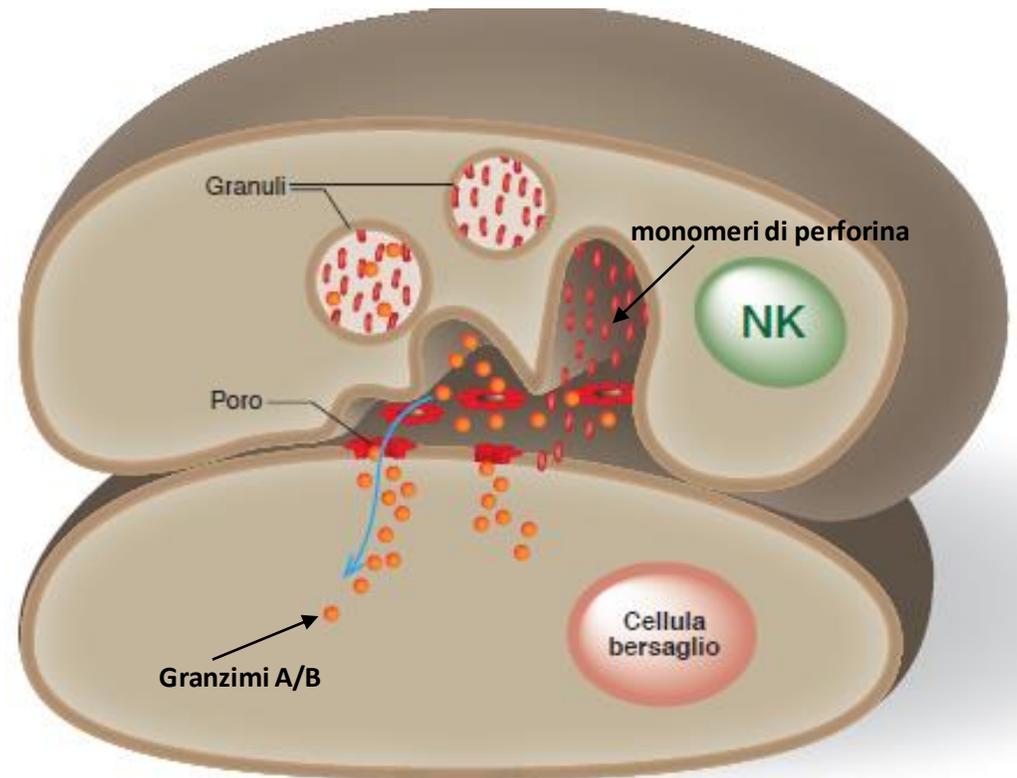
## Le diverse fasi della citotossicità



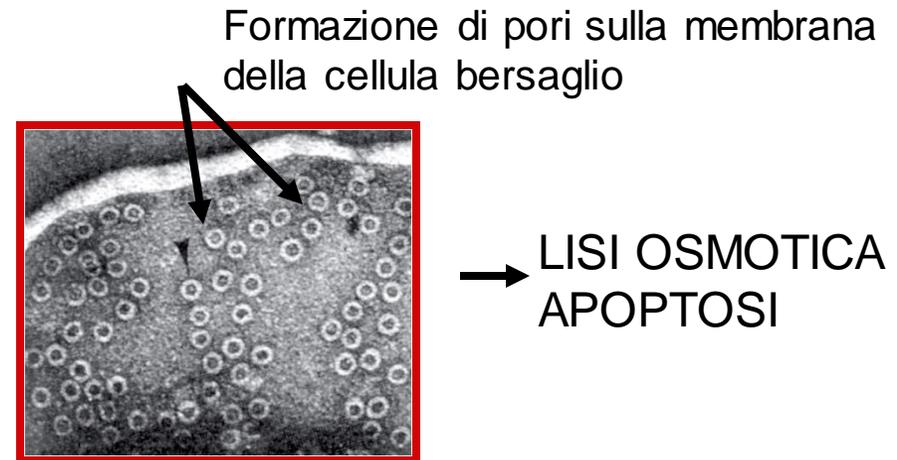
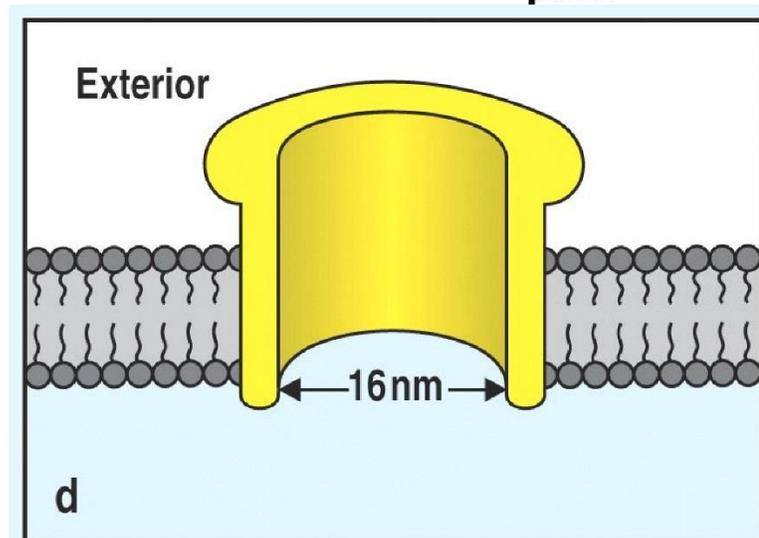
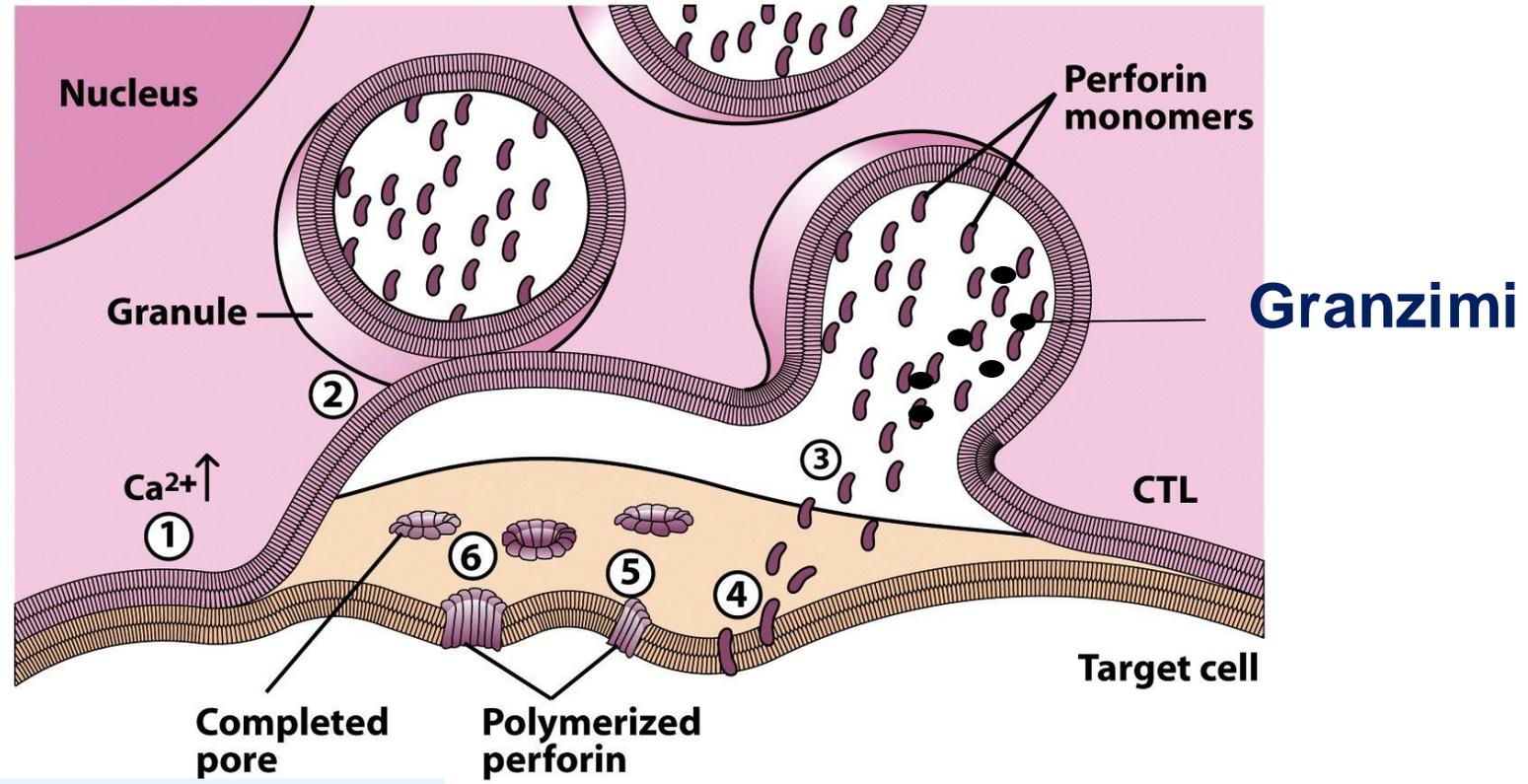
## Che cosa contengono i granuli citotossici delle cellule NK (e dei linfociti T citotossici)?

Protein in lytic granules of T and NK cells	Actions on target cells
Perforin	Polymerizes to form a pore in target membrane
Granzymes	Serine proteases, which activate apoptosis once in the cytoplasm of the target cell
Granulysin	Induces apoptosis

## Perforina e granzimi mediano la morte della cellula bersaglio



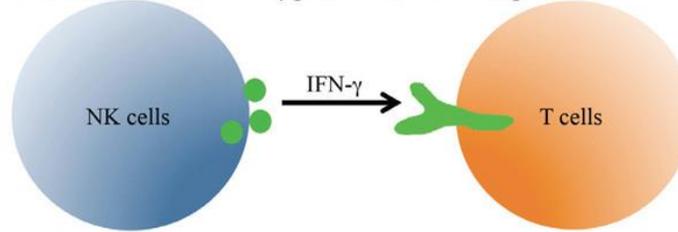
# Sinergismo tra perforina e granzimi nel mediare la morte della cellula bersaglio



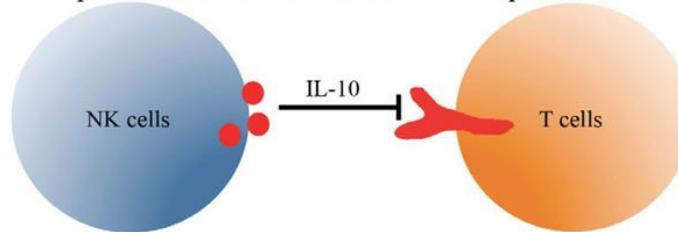
**Le cellule NK istruiscono e modulano  
la risposta immunitaria adattativa**

# Le cellule NK regolano i linfociti T

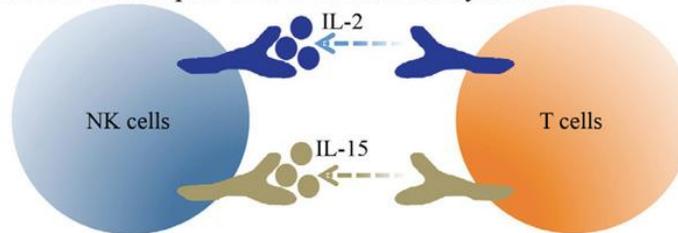
A. NK cell secretion of IFN- $\gamma$  promotes T cell responses



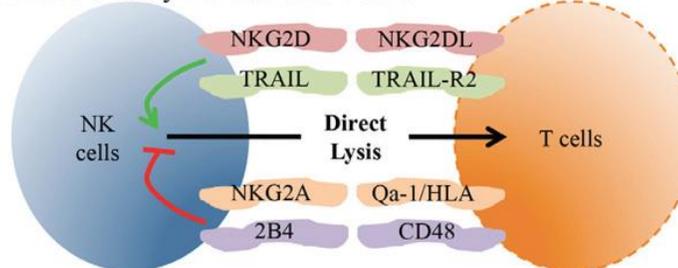
B. NK cell production of IL-10 inhibits T cell responses



C. NK cells outcompete T cells for survival cytokines

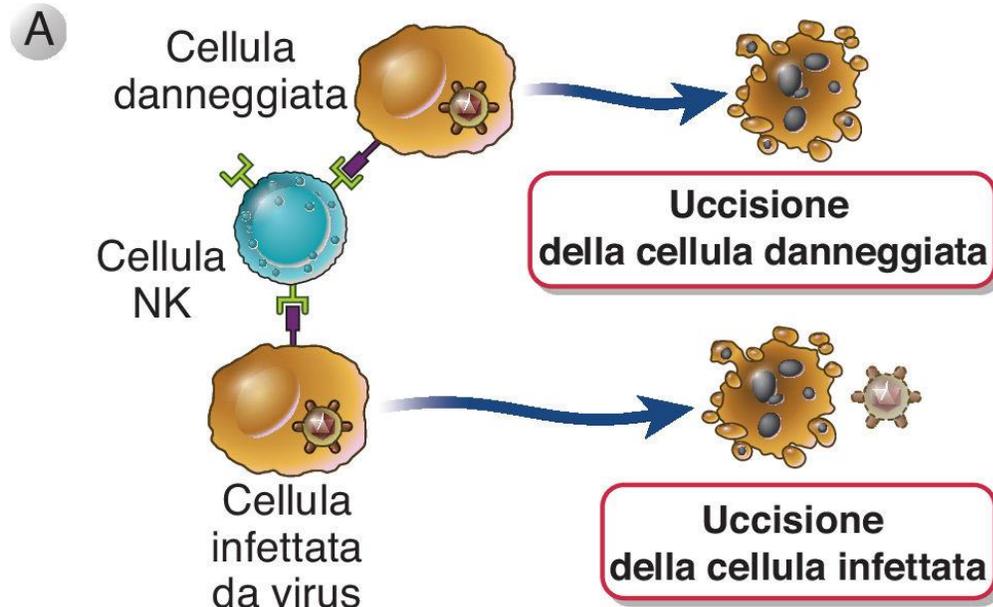


D. NK cells directly kill activated T cells

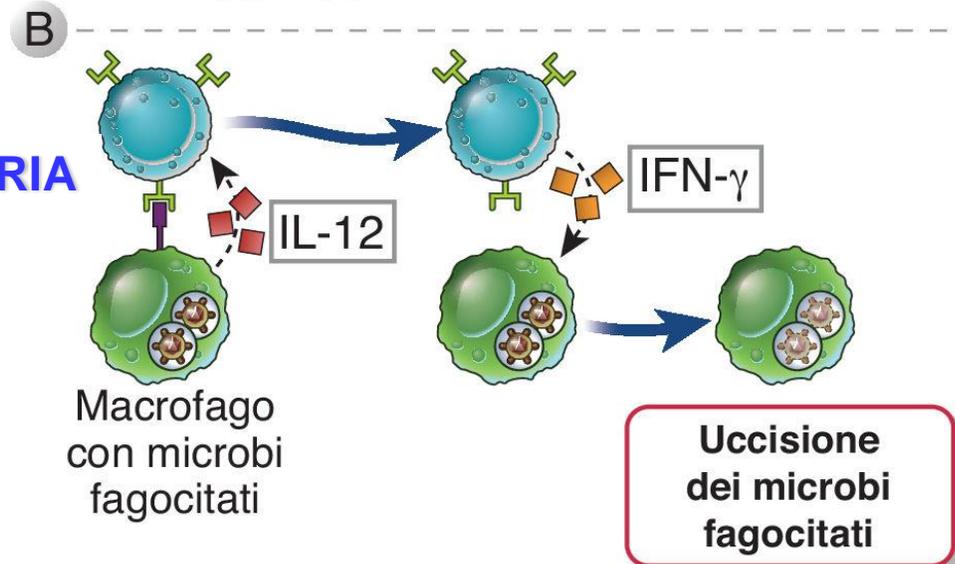


# LE FUNZIONI DELLE CELLULE NK

CITOTOSSICA

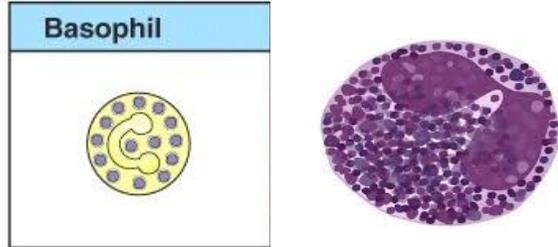
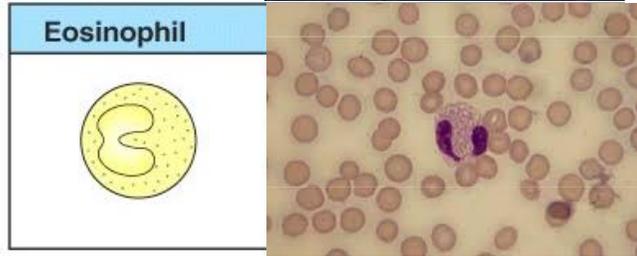
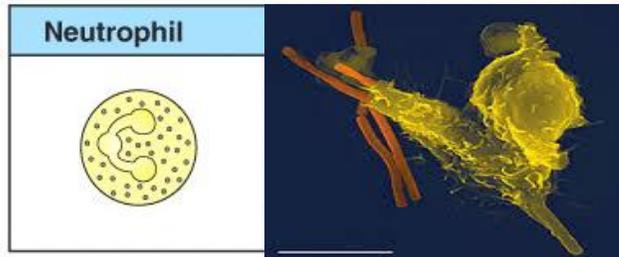


IMMUNOREGOLATORIA

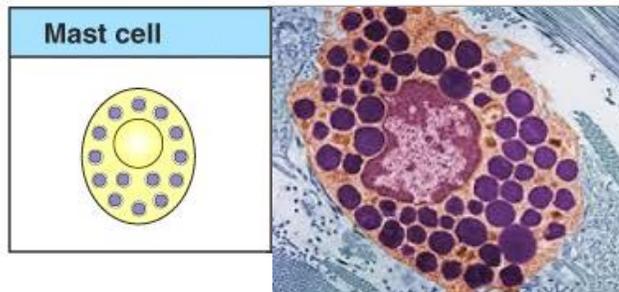


# LE CELLULE DELL'IMMUNITA' INNATA

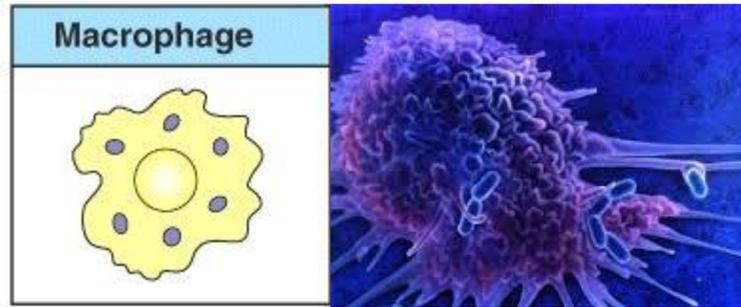
## I GRANULOCITI



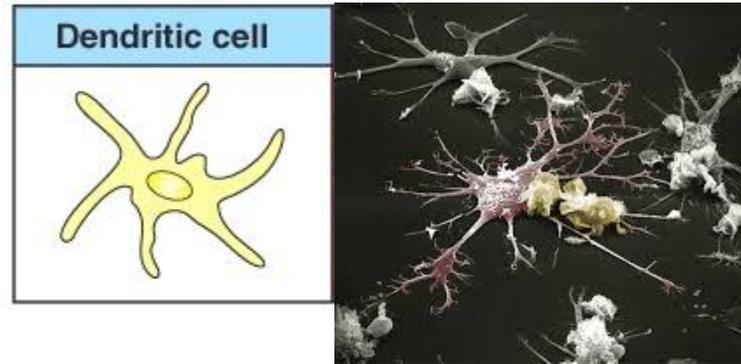
## I MASTOCITI



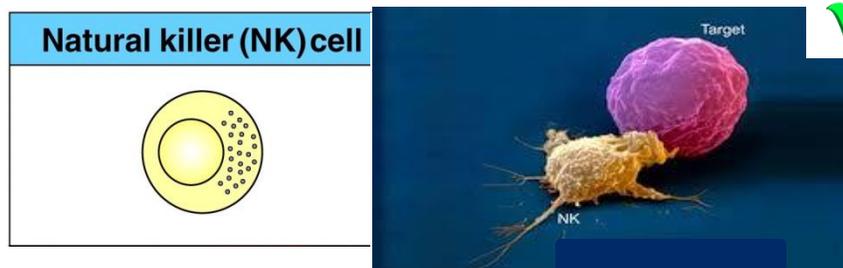
## I MACROFAGI



## LE CELLULE DENDRITICHE



## LE CELLULE NATURAL KILLER (NK)



# INVASIONE MICROBICA: QUALI CELLULE SE NE ACCORGONO ?

Diversi tipi di cellule....

**Mastociti: sentinelle tissutali!**

	Cell		Activated function
<b>Mast cell</b>		Release of granules containing histamine and other active agents	

I granulociti **basofili** sono la controparte circolante dei mastociti (~1% dei leucociti) e condividono con loro recettori e funzioni

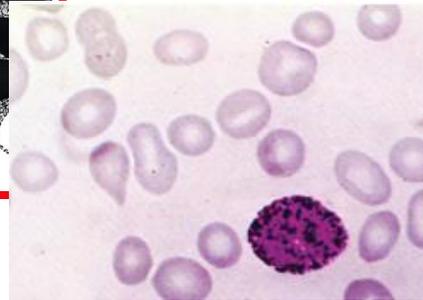
Basophil		Release of histamine
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# I GRANULOCITI BASOFILI (circolanti) e i MASTOCITI (*in situ*)

basofilo



L'ipersensibilità di tipo I è mediata dall'attivazione e successiva degranulazione dei **basofili** e dei **mastociti (*connettivali e mucosali*)**.



Mastociti (Basofili)

## CARATTERISTICHE E FUNZIONI

- Legano le molecole IgE
- Contengono granuli elettron-densi

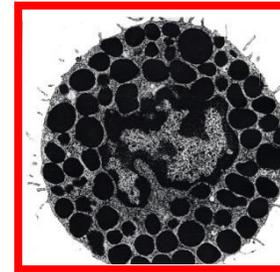
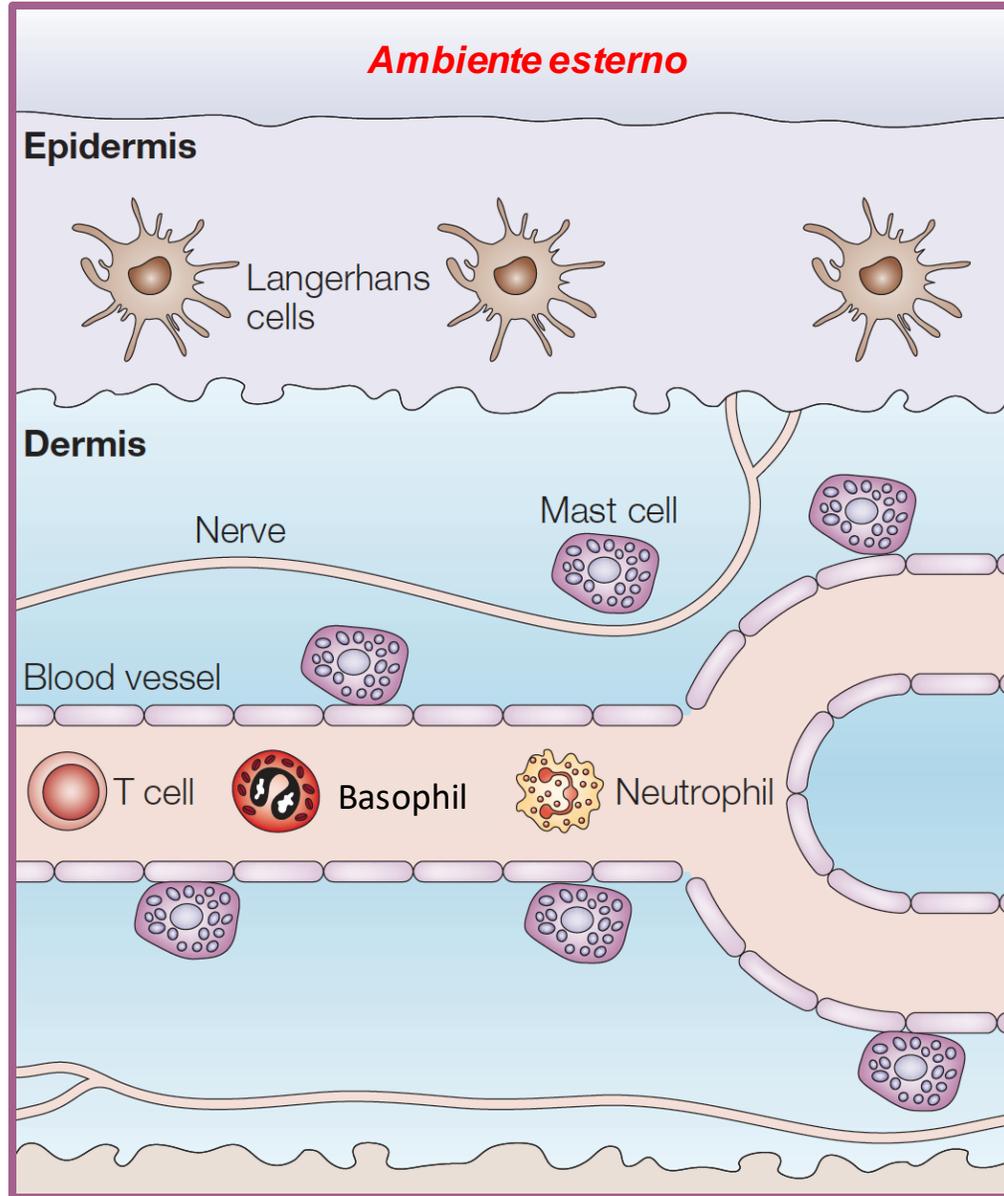
## MEDIATORI PRIMARI DELL'INFIAMMAZIONE

- Istamina
- Leucotrieni (LTC, LTD, LTE)
- Fattore attivante le piastrine
- Fattori chemiotattici per gli eosinofili
- Citochine (ad es. TNF- $\alpha$ , IL-4)

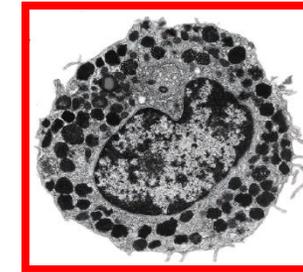
mastocita



# La posizione strategica dei mastociti!

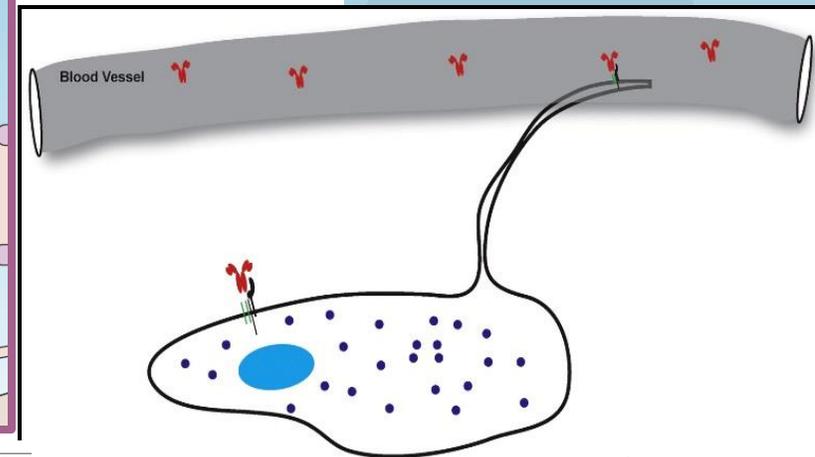


**Basofili  
(circolanti)**

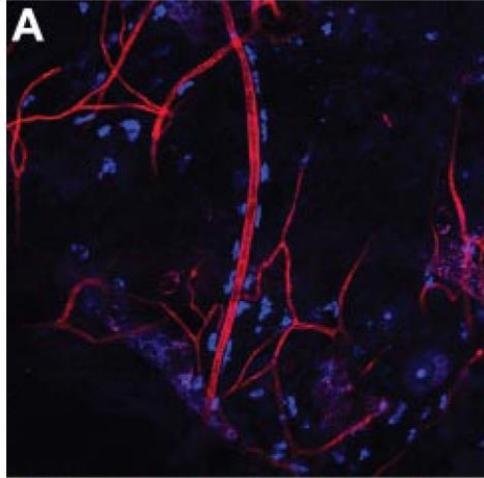


**Mastociti  
(connettivo,  
mucosa)**

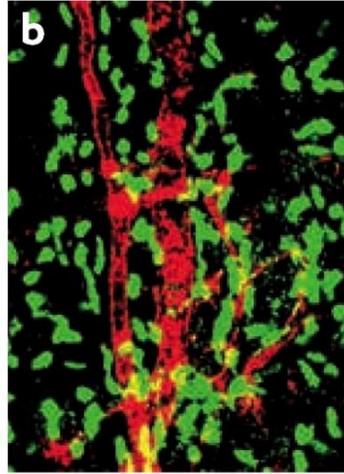
**In cerca IgE  
circolanti!**



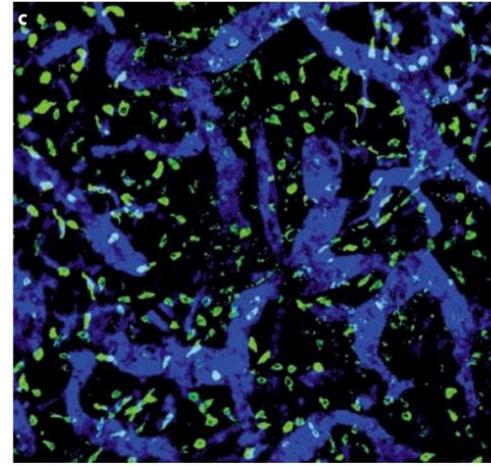
**I mastociti sono la controparte tissutale dei basofili  
e sono le sentinelle tissutali del danno!**



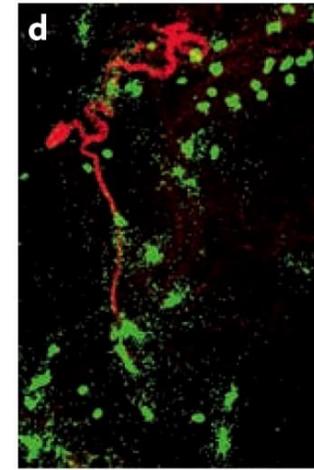
**Mast cells (MC) (blue)**  
**blood vessel (red)**



**MC (green)**  
**blood vessel (red)**



**MC (green)**  
**lymphatic vessel (blue)**



**MC (green)**  
**neuron (red)**

**SI DIVIDONO IN:**

**Connective tissue mast cells**

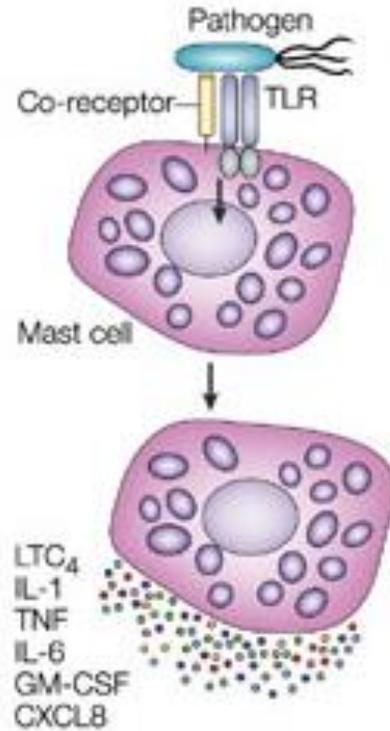
Cute, sottomucosa intestinale

**Mucosal mast cells**

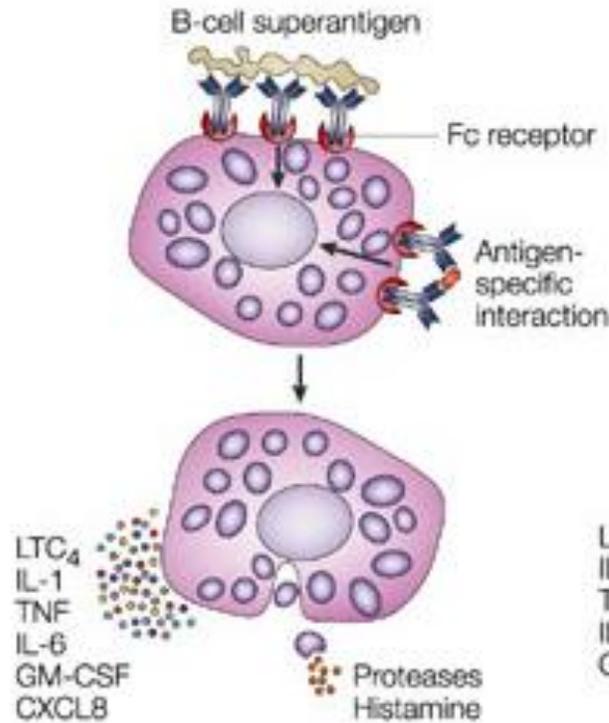
Alveoli, mucosa intestinale

# Non solo fagociti: i mastociti hanno un ruolo sentinella molto importante!

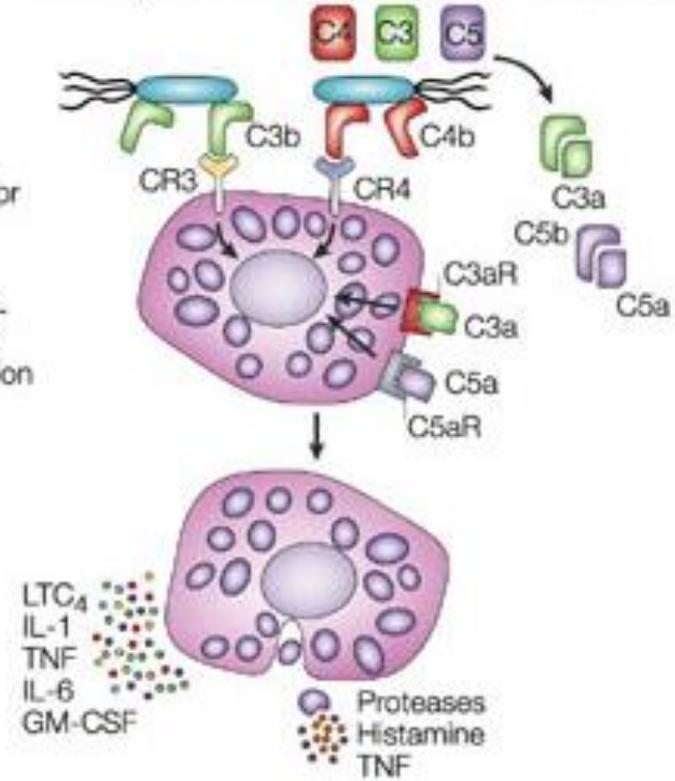
**a Direct interactions**



**b Fc-receptor-mediated activation**

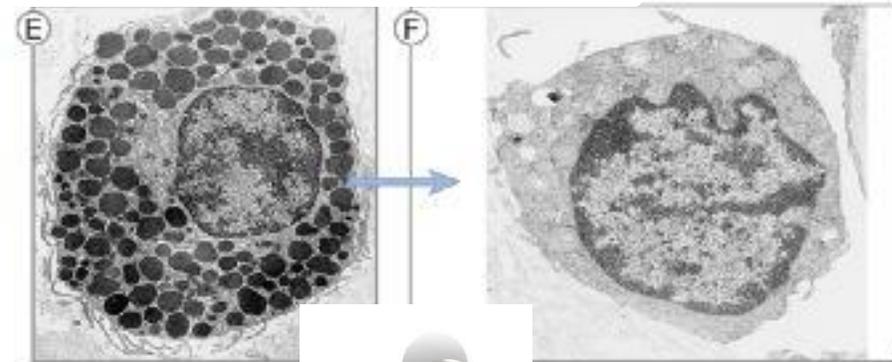
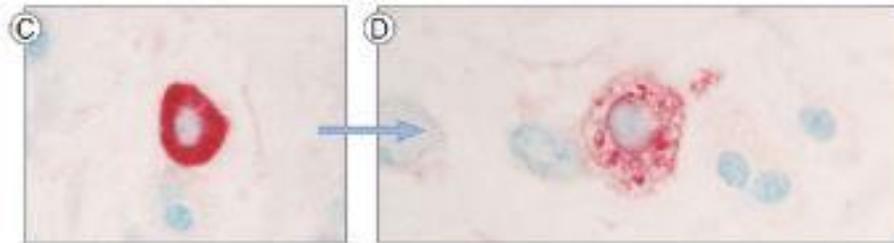
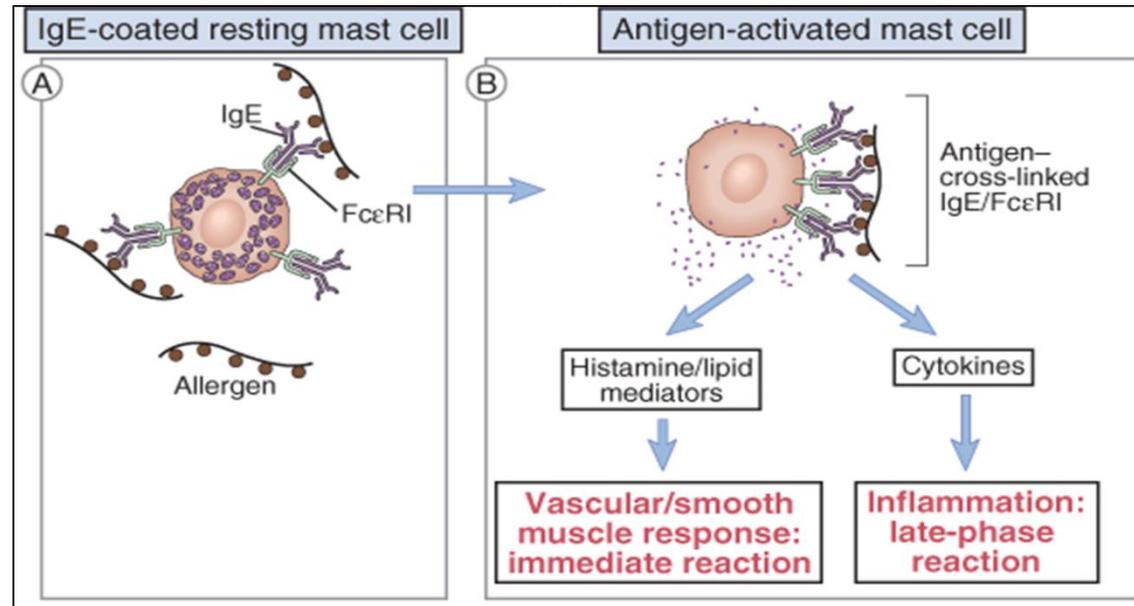


**c Complement-receptor-mediated activation**



# L'attivazione dei mastociti

Ag multivalente:  
Ag con epitopo ripetuto  
Ag con due o più epitopi



## **I mediatori solubili:**



### **Pre-formati**

- Amine vasoattive (istamina)
- Enzimi (serin-proteasi neutre)
- Macromolecole (proteoglicani, eparina, condroitin solfato)
- Alcune citochine

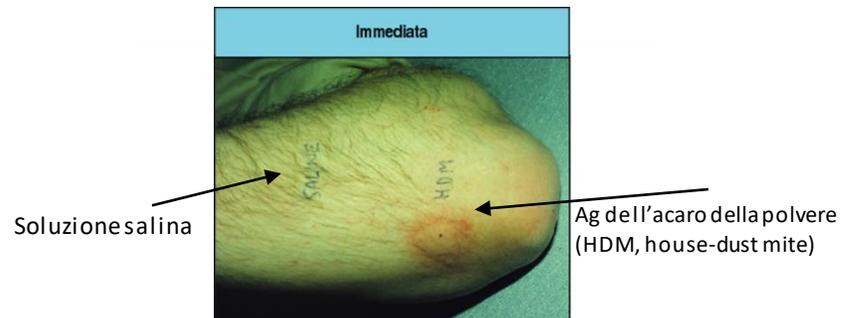
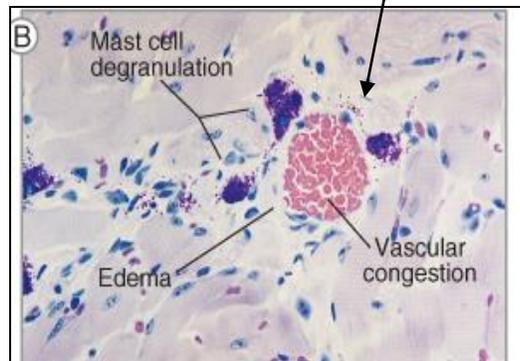
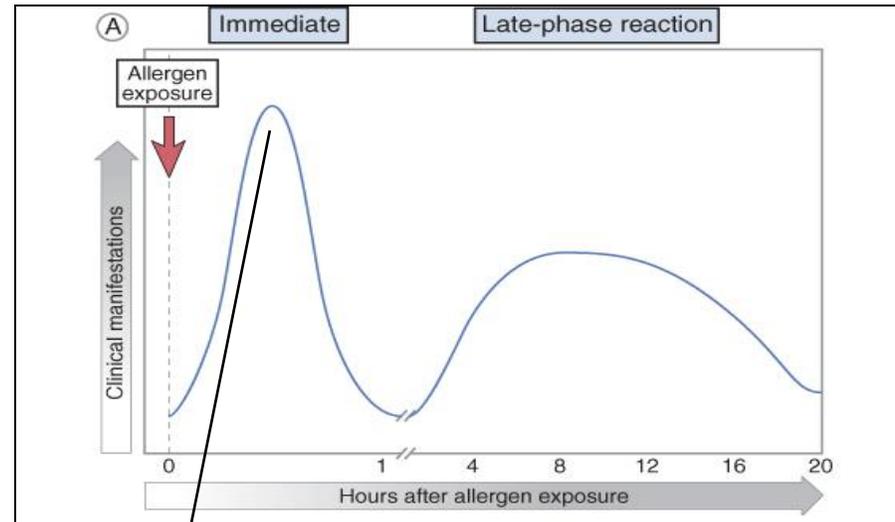
### **Neo-sintetizzati**

- Mediatori lipidici (prostaglandine, leucotrieni, PAF)
- Citochine, chemochine

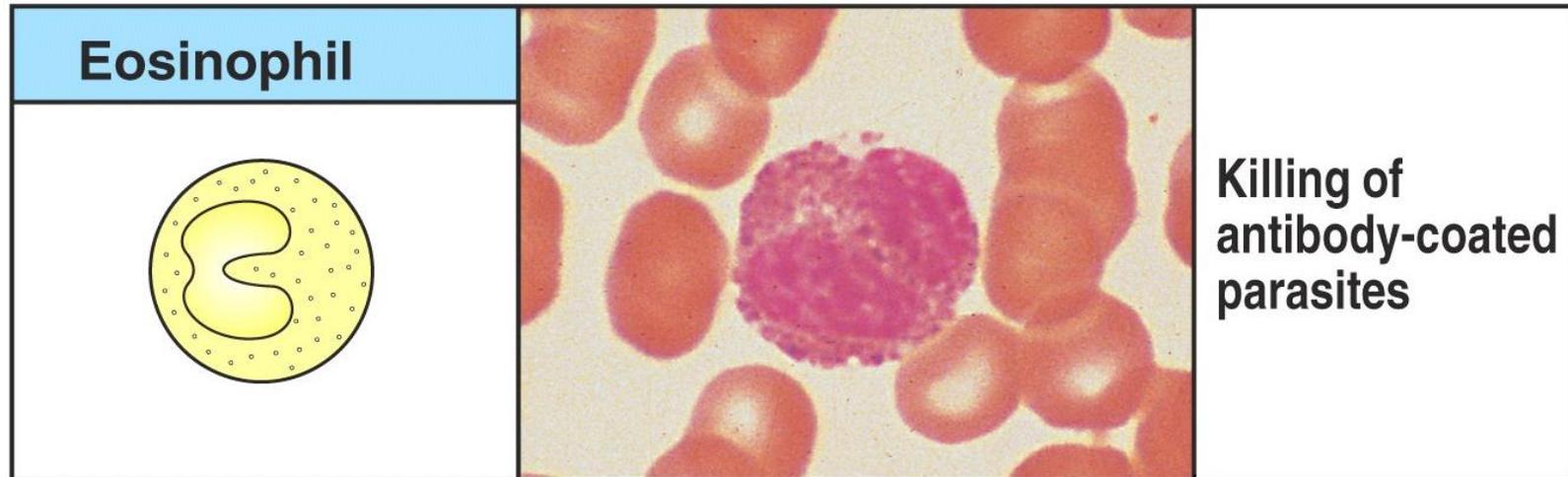
## I mastociti attivati degranulano e producono i mediatori dell'inflammatione

Class of product	Examples	Biological effects
Enzyme	Tryptase, chymase, cathepsin G, carboxypeptidase	Remodel connective tissue matrix
Toxic mediator	Histamine, heparin	Toxic to parasites Increase vascular permeability Cause smooth muscle contraction
Cytokine	IL-4, IL-13	Stimulate and amplify T <sub>H</sub> 2 cell response
	IL-3, IL-5, GM-CSF	Promote eosinophil production and activation
	TNF- $\alpha$ (some stored preformed in granules)	Promotes inflammation, stimulates cytokine production by many cell types, activates endothelium
Chemokine	CCL3 (MIP-1 $\alpha$ )	Attracts monocytes, macrophages, and neutrophils
Lipid mediator	Leukotrienes C4, D4, E4	Cause smooth muscle contraction Increase vascular permeability Stimulate mucus secretion
	Platelet-activating factor	Attracts leukocytes Amplifies production of lipid mediators Activates neutrophils, eosinophils, and platelets

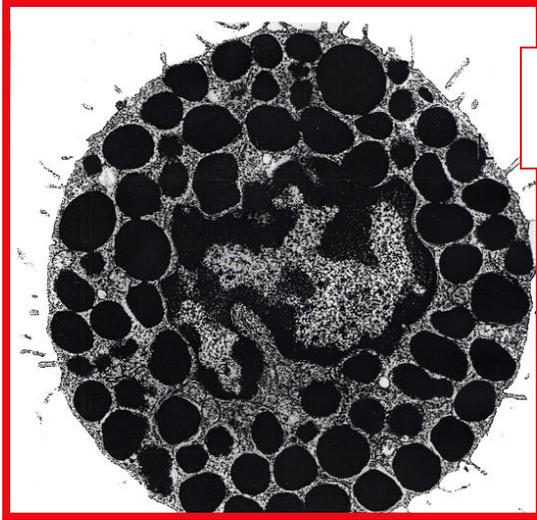
# Un esempio dell'azione di mastociti, basofili ed eosinofili (le fasi immediata e tardiva nelle allergie)



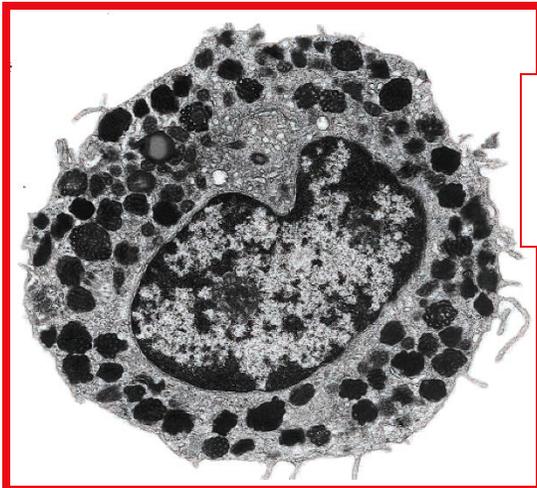
I granulociti **eosinofili** (~ 0,5-3% dei leucociti)  
sono importanti per combattere le infezioni da elminti



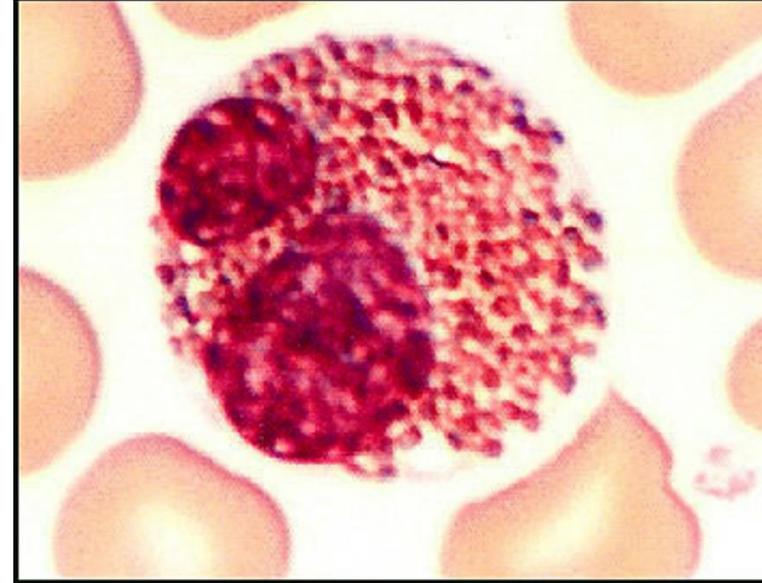
# Basofili (e mastociti nei tessuti) ed eosinofili partecipano alla risposta contro gli elminti



Basofili  
(circolanti)

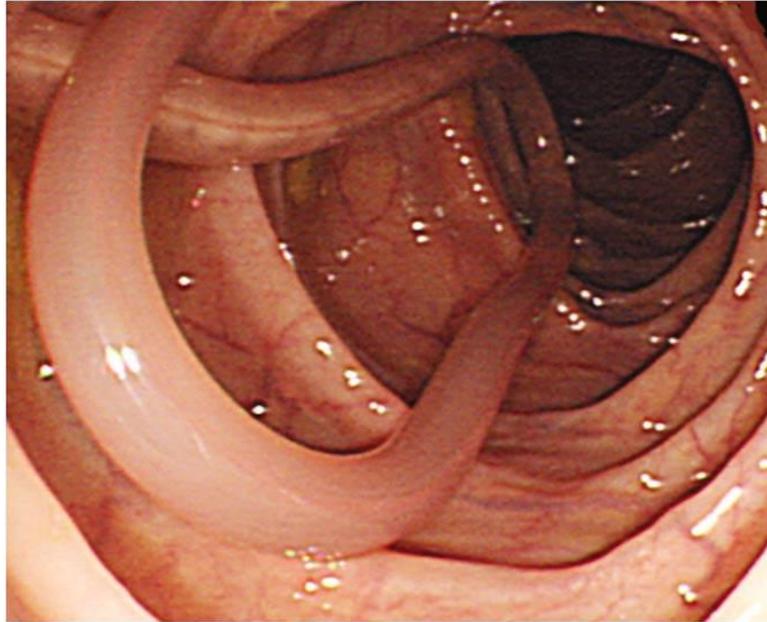


Mastociti  
(connettivali e  
mucosali)

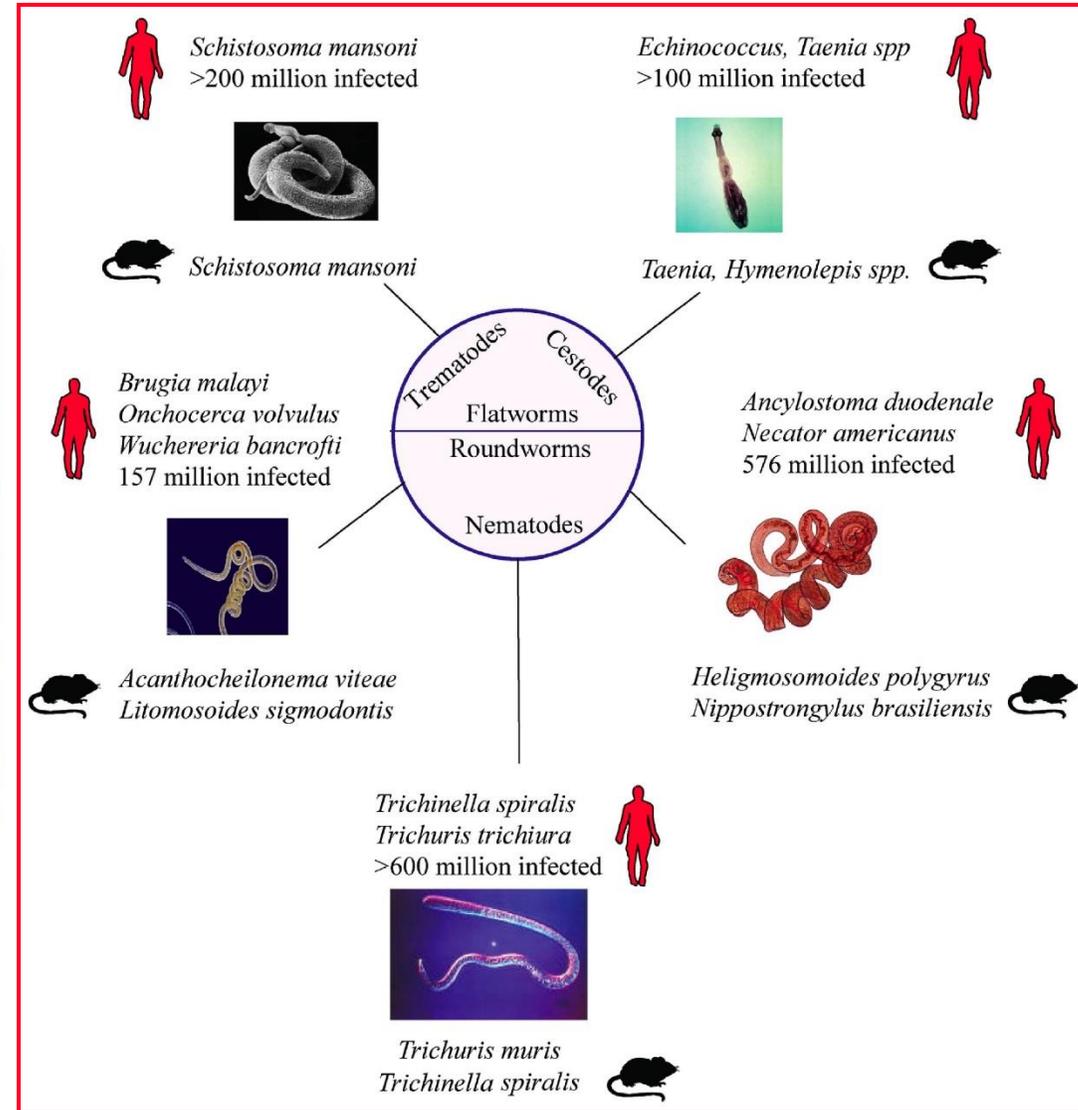


Gli eosinofili contengono  
proteine tossiche che  
danneggiano il tegumento  
degli elminti

# Gli elminti



*Ascaris lumbricoides* (lungo 20 cm)  
nel lume intestinale



Le principali infezioni da elminti  
nell'Uomo

# Gli eosinofili mediano l'ADCC (antibody-dependent cytotoxicity) contro grandi parassiti

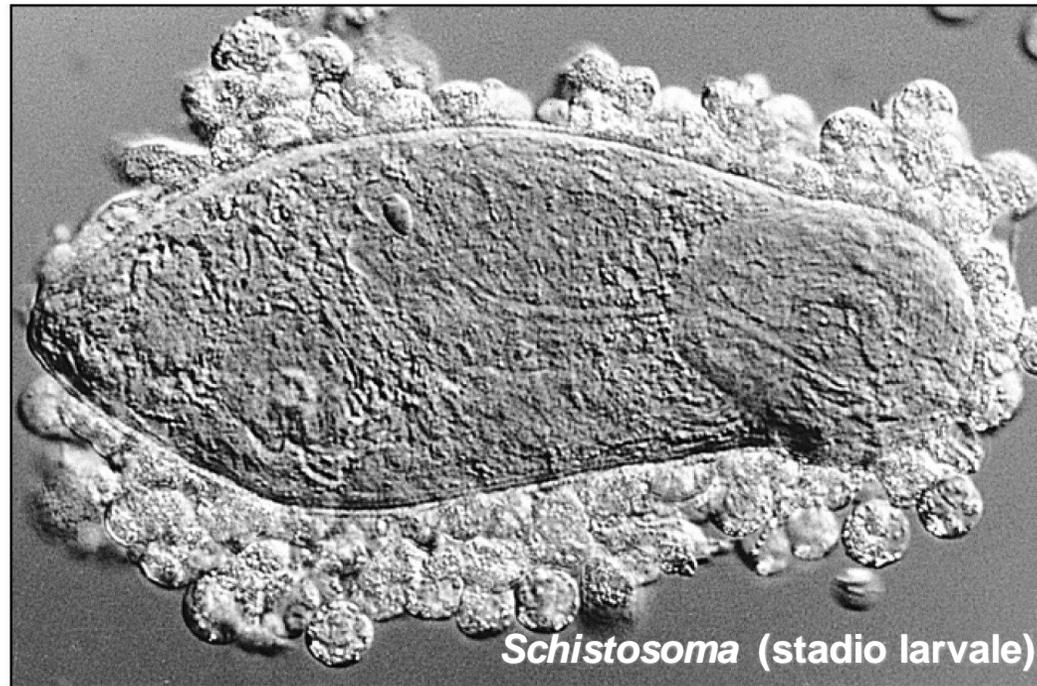
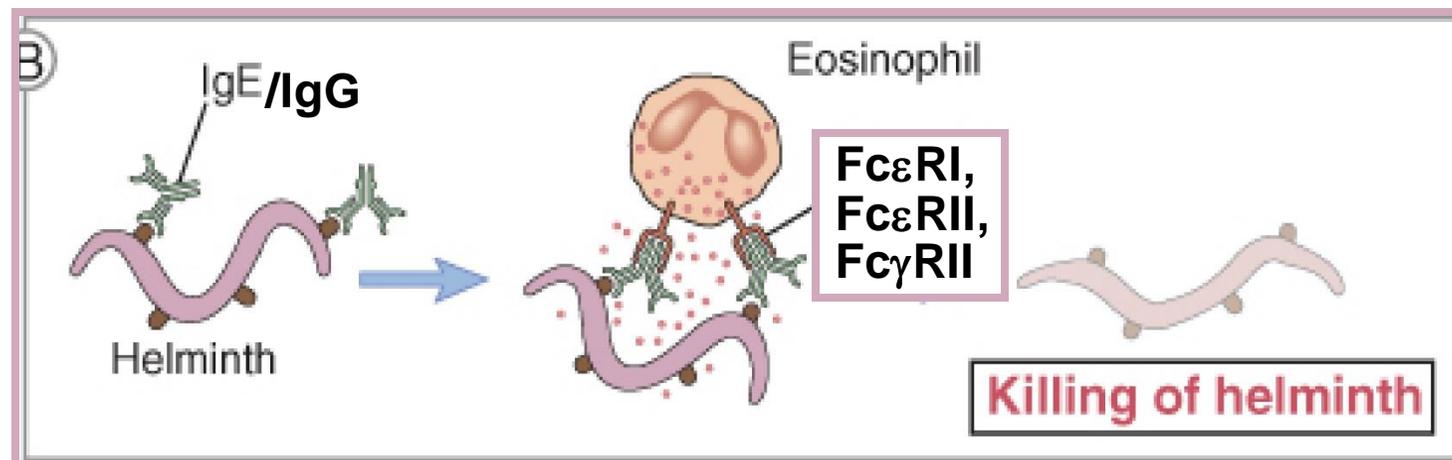


Figure 9-33 Immunobiology, 6/e. (© Garland Science 2005)

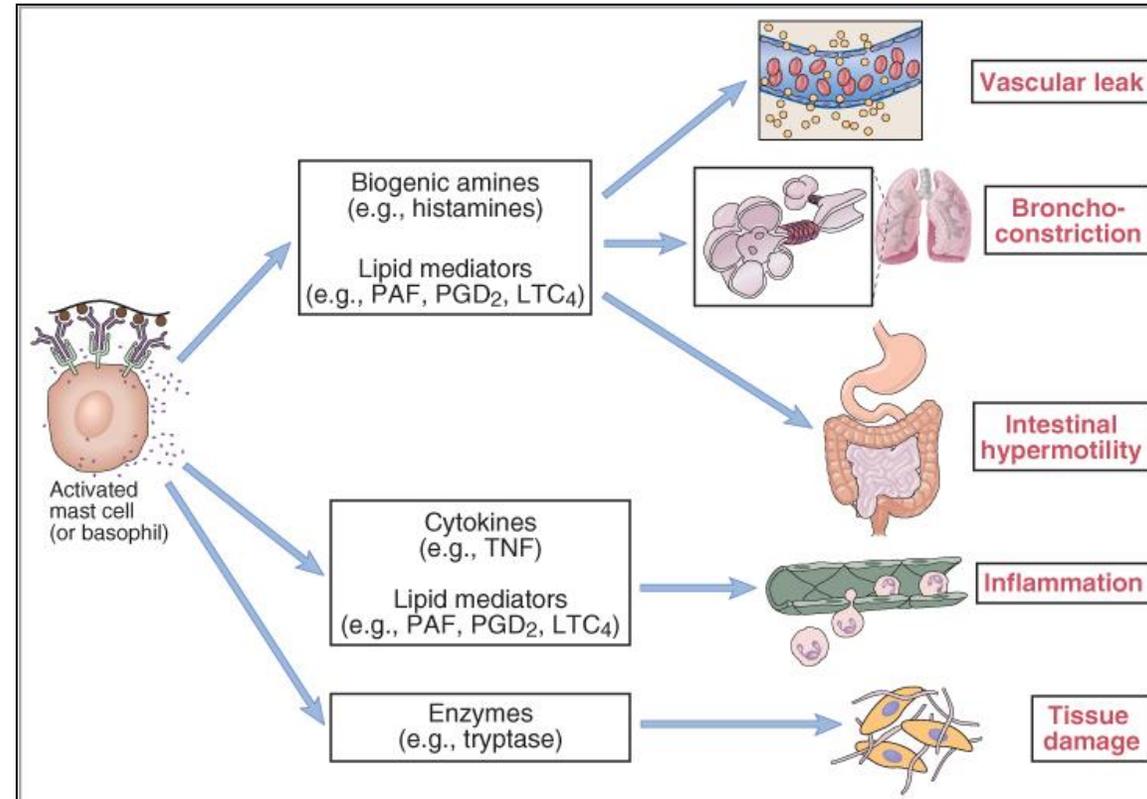


## Gli eosinofili secernono proteine tossiche e altre mediatori contenuti nei loro granuli

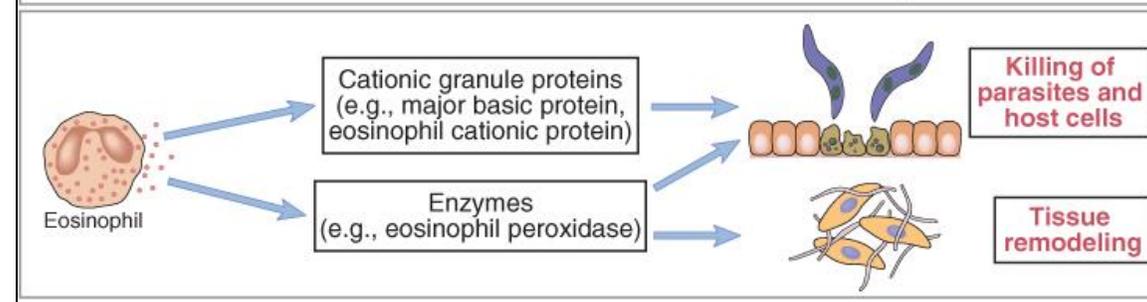
Classe di prodotti	Prodotto	Effetti biologici
Enzimi	Perossidasi degli eosinofili	Tossicità per i parassiti e per le cellule di mammifero mediante catalisi dell'alogeneazione, induzione del rilascio di istamina da parte dei mastociti
	Collagenasi degli eosinofili	Rimodellamento della matrice del tessuto connettivo
Proteine tossiche	Proteina basica maggiore	Tossicità per i parassiti e le cellule di mammifero, induzione del rilascio di istamina da parte dei mastociti
	Proteina cationica degli eosinofili	Tossicità per i parassiti, neurotossicità
	Neurotossina derivata dagli eosinofili	Neurotossicità
Citochine	IL-3, IL-5, GM-CSF	Amplificazione della produzione di eosinofili da parte del midollo osseo, attivazione degli eosinofili
Chemochine	CXCL8	Promozione dell'afflusso di leucociti
Mediatori lipidici	Leucotrieni C <sub>4</sub> , D <sub>4</sub> ed E <sub>4</sub>	Contrazione della muscolatura liscia, aumento della permeabilità vascolare, secrezione di muco
	Fattore di attivazione piastrinica	Chemiotassi per i leucociti, amplificazione della produzione di mediatori lipidici, attivazione di neutrofili, eosinofili e piastrine

# Effetti biologici dei mediatori rilasciati da mastociti, basofili ed eosinofili

## Mastociti e basofili



## Eosinofili



# Un esempio dell'azione di mastociti, basofili ed eosinofili (le fasi immediata e tardiva nelle allergie)

