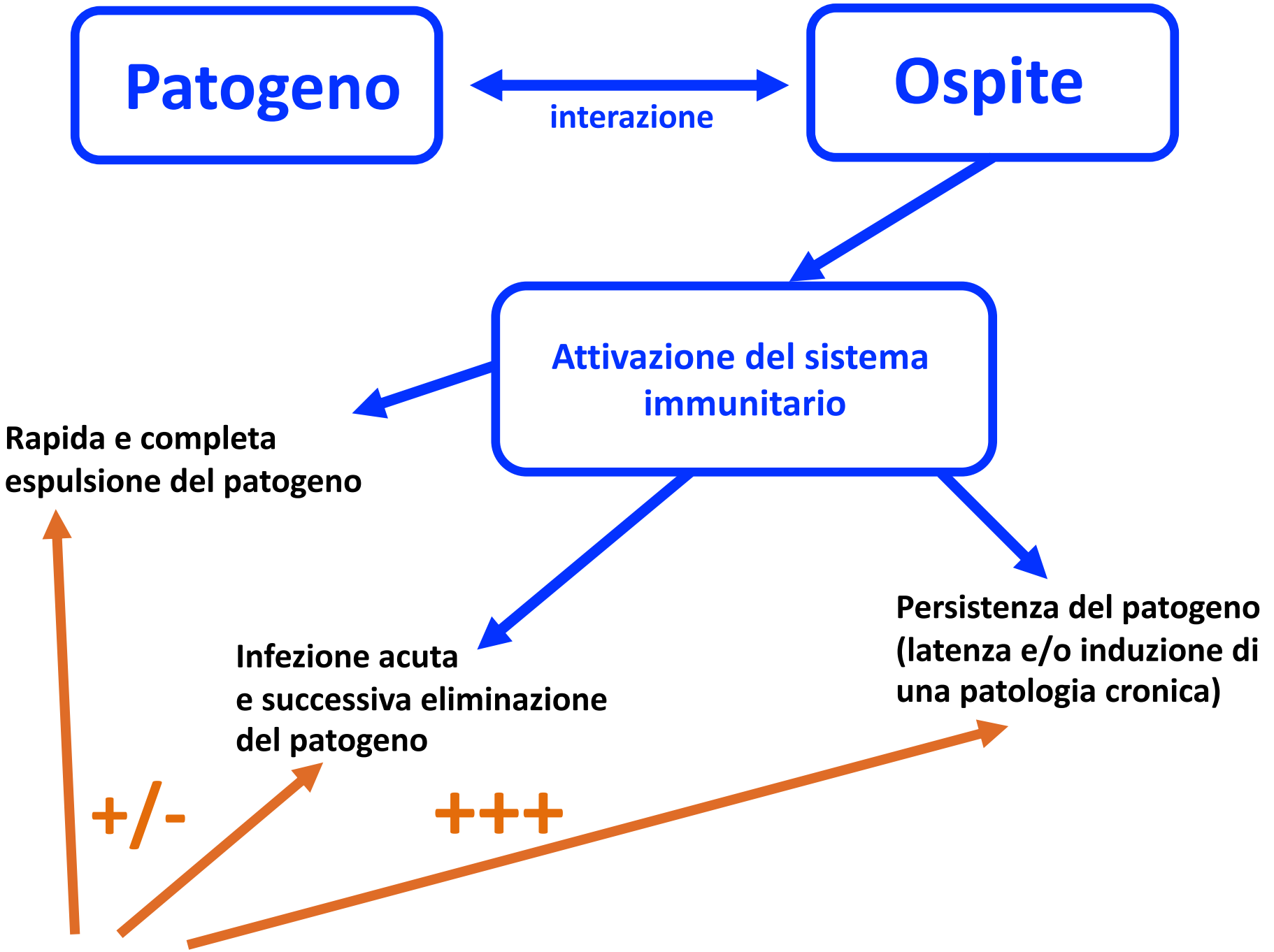
CODICE OPIS
PATOLOGIA MOLECOLARE E IMMUNOPATOLOGIA (1041600)

11JLUJMH

PATOLOGIA MOLECOLARE
(1041600_2)


Patogeno
interazione
Ospite
Attivazione del sistema immunitario
Rapida e completa espulsione del patogeno
Infezione acuta e successiva eliminazione del patogeno
Persistenza del patogeno (latenza e/o induzione di una patologia cronica)
+/-
+++
Pressione selettiva esercitata dal sistema immunitario sul patogeno

Nel corso della co-evoluzione con l'ospite, i patogeni hanno acquisito meccanismi strategici per evadere l'immunità innata ed adattativa

Esempi di strategie:

"Dormancy" per rendersi "poco visibili"

- produzione minima di proteine (herpesvirus)
- quiescenza dei micobatteri
- integrazione del DNA nel genoma dell'ospite (retrovirus)

I patogeni hanno “imparato” ad evadere l’immunità innata ed acquisita nel corso della co-evoluzione con l’ospite

Esempi di strategie:

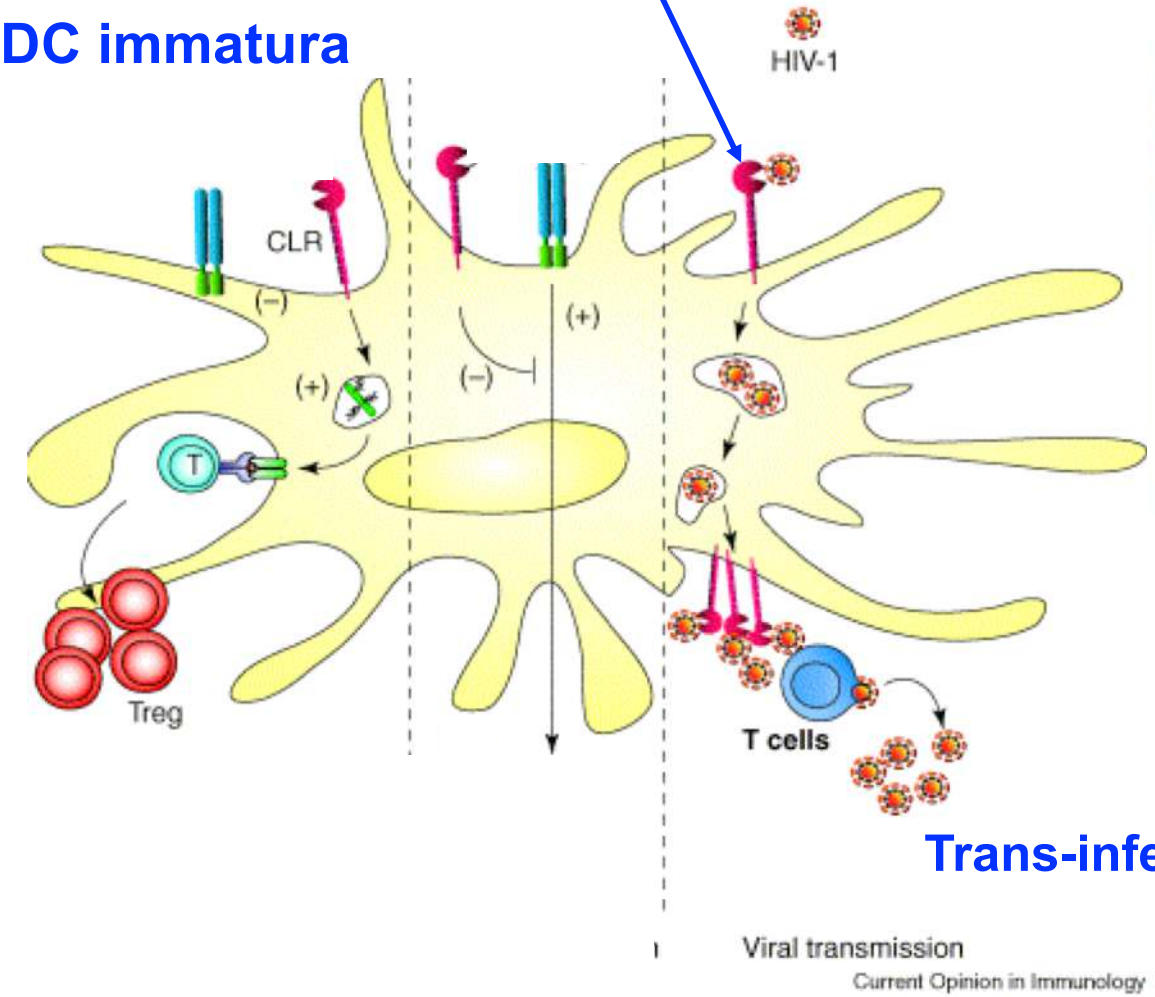
“Sequestration” ovvero occupare “siti speciali”

- invasione dei globuli rossi da parte del Plasmodio della malaria
- colonizzazione della cistifellea da parte di *S. enterica* Typhi
- uso da parte di HIV delle cellule dendritiche per il trasporto dalle mucose ai linfonodi

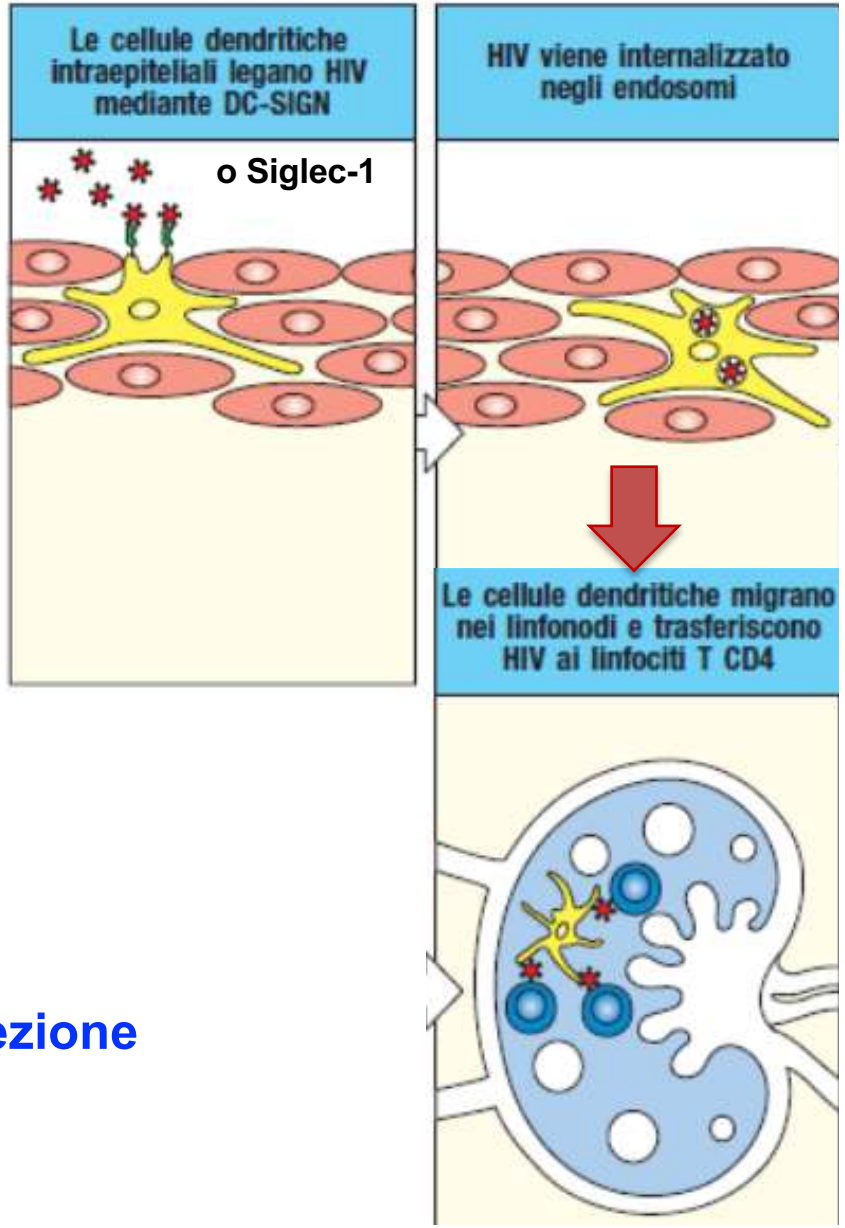
HIV usa le cellule dendritiche come cavallo di Troia per il trasporto dalle mucose ai tessuti linfoidi

Legame di gp120 a recettori lectinici (Siglec-1 e DC-SIGN);
Internalizzazione e persistenza nel compartimento endosomiale

DC immatura



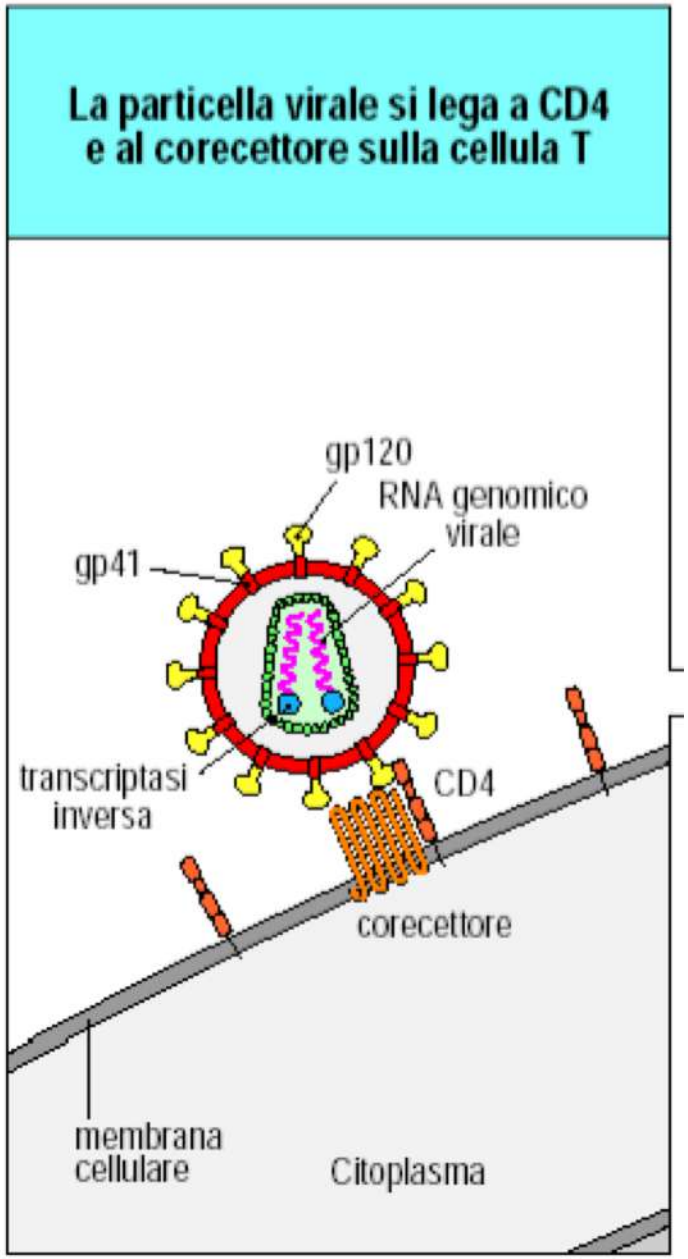
Trans-infezione

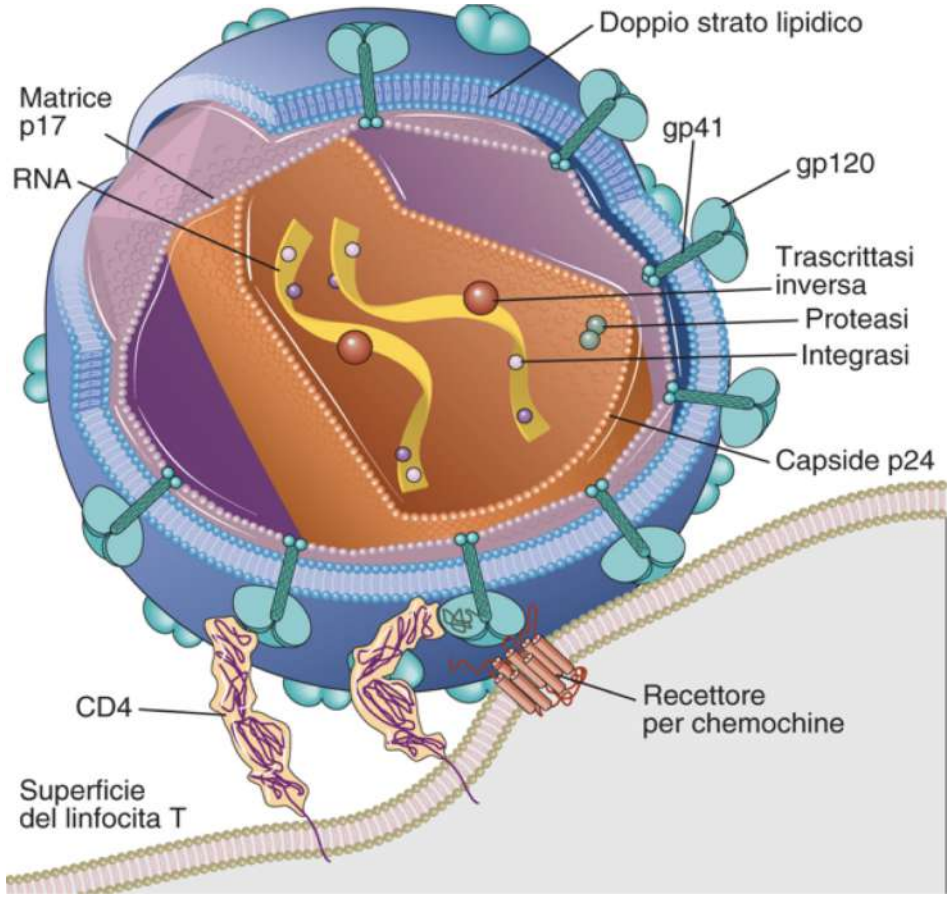


Struttura di HIV ed infezione cellulare

polimorfismi dell'envelope e corecettori utilizzati

Il CD4 funge da recettore di HIV

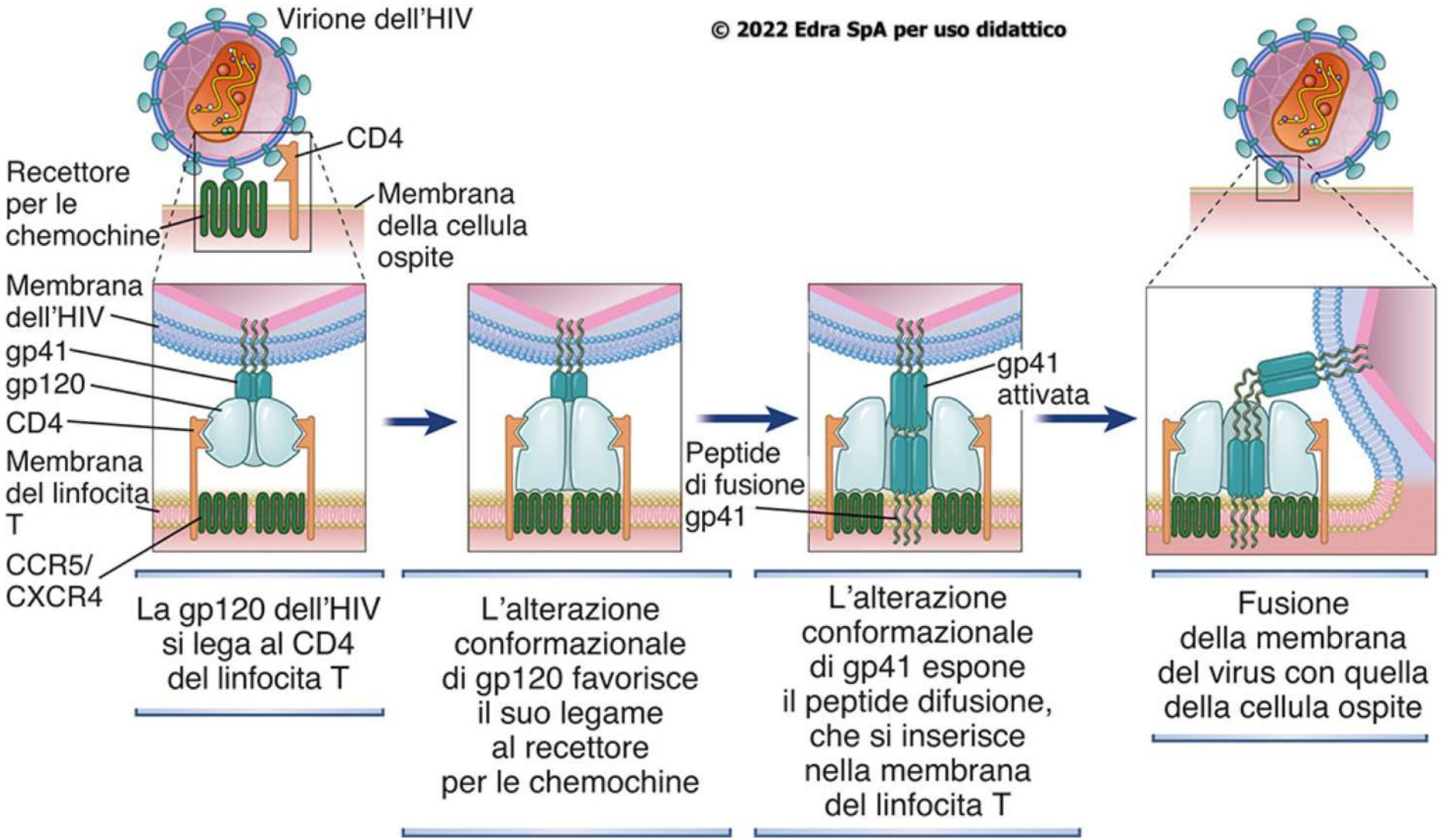




Varianti R5 (M-tropiche) usano come corecettore CCR5 ed infettano DC (iDC), macrofagi e cellule T CD4+

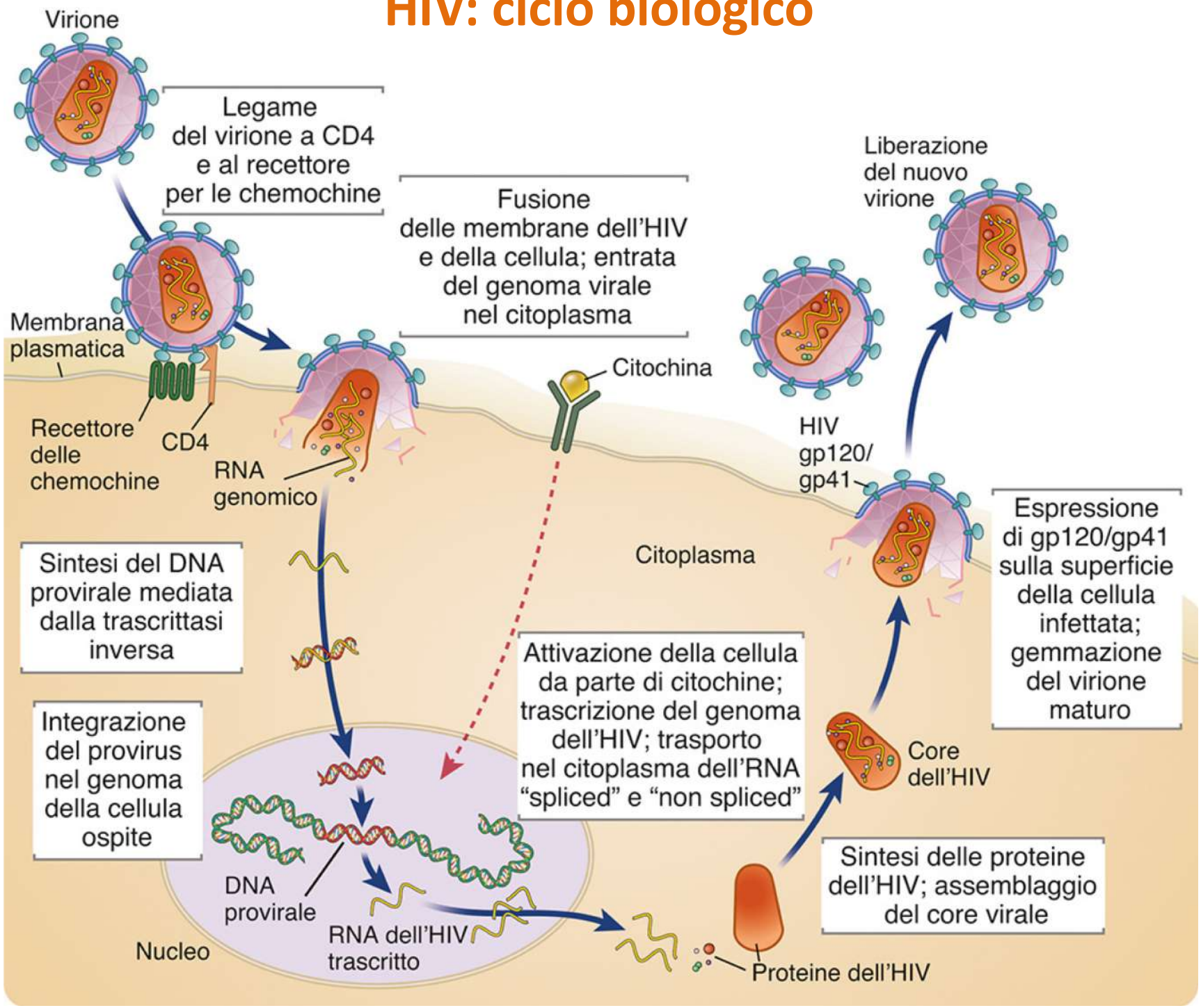
Varianti X4 (T-tropiche) usano come corecettore CXCR4 ed infettano preferibilmente i linfociti T CD4+ (anche le mDC)

HIV: ingresso nella cellula bersaglio



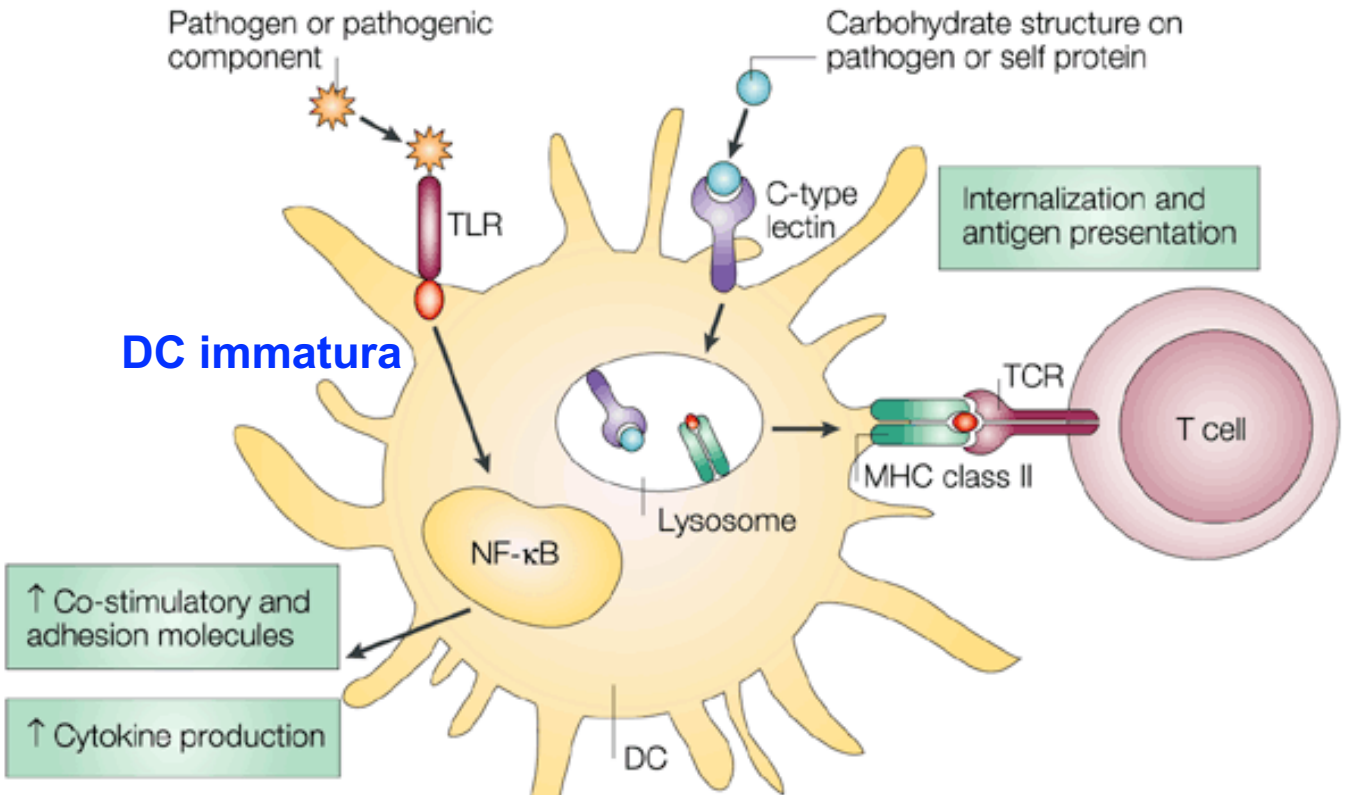
Meccanismo di penetrazione di HIV nella cellula bersaglio. Nel modello presentato si nota come il legame a CD4 induca modificazioni sequenziali della conformazione delle proteine gp120 e gp41. Tali modificazioni promuovono il legame del virus al corecettore (di solito un recettore per le chemochine). Parallelamente, l'attivazione del peptide di fusione della gp41 smaschera i residui aminoacidici idrofobici che vanno a inserirsi nella membrana plasmatica della cellula ospite. Tutto ciò favorisce la fusione della membrana dell'HIV-1 con quella della cellula bersaglio.

HIV: ciclo biologico



L'interazione dei patogeni con le DC è mediato da recettori di vario tipo e promuove la risposta immunitaria adattativa ma alcuni patogeni possono sabotare i pathways di attivazione e maturazione

L'esempio di HIV

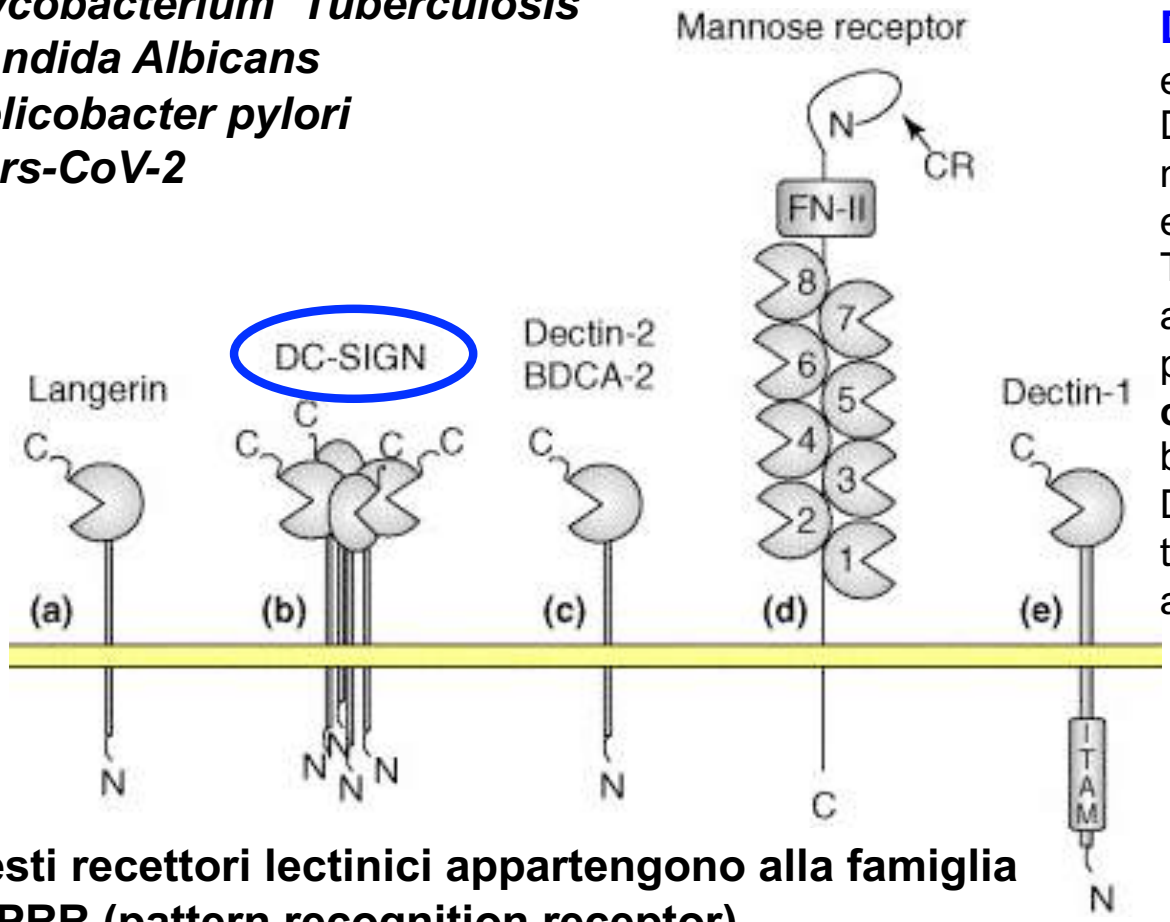


C-type lectins and Toll-like receptors: pathogen receptors on dendritic cells. For the recognition of microorganisms, immature dendritic cells (DCs) express Toll-like receptors (TLRs) and C-type lectins that bind specific pathogen components and carbohydrate structures, respectively. After recognition by TLRs a signal-transduction cascade is induced, which through the activation of nuclear factor-B (NF-B) results in the upregulation of expression of co-stimulatory molecules and adhesion molecules, and the production of cytokines, leading to DC maturation. The recognition of pathogens by C-type lectins leads to internalization of pathogens and intracellular processing for presentation by MHC class I and II molecules to T cells. TCR, T-cell receptor.

DC-SIGN: recettore lectinico di tipo C espresso da DC e macrofagi

Virus che interagiscono con DC-SIGN:

- **HIV (gp120)**
- **Virus Dengue**
- **HCV**
- **Virus Ebola**
- **CMV**
- ***Mycobacterium Tuberculosis***
- ***Candida Albicans***
- ***Helicobacter pylori***
- ***Sars-CoV-2***

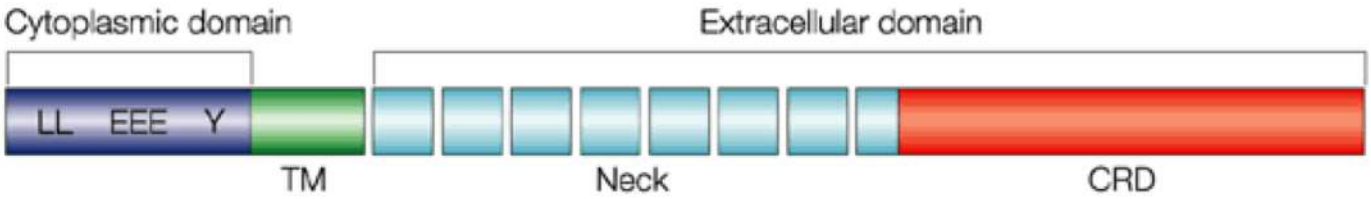


Questi recettori lectinici appartengono alla famiglia dei PRR (pattern recognition receptor)

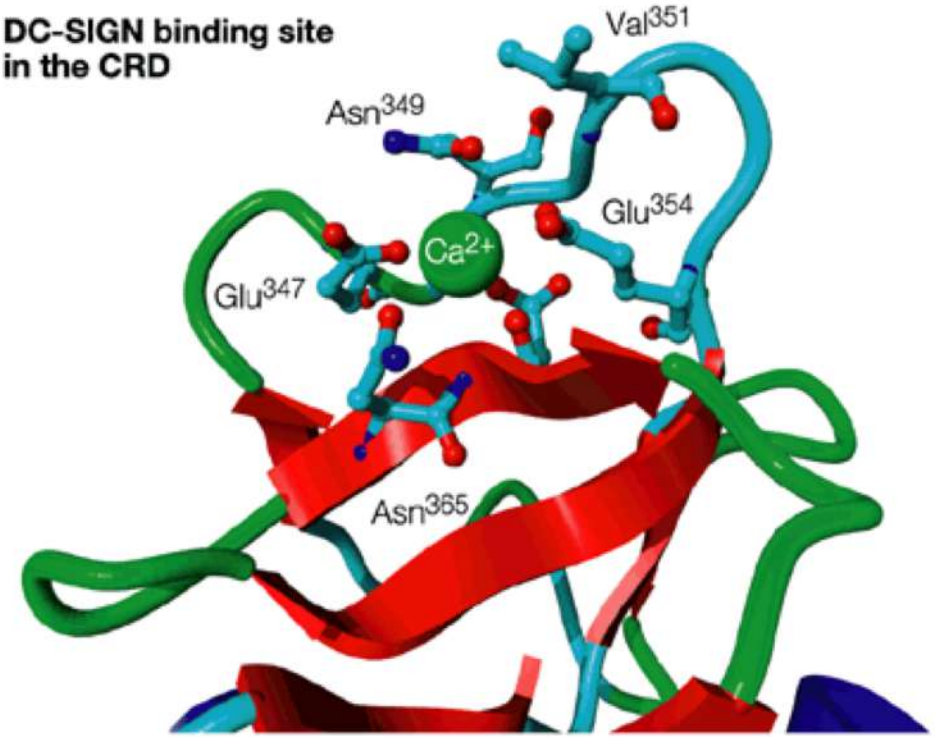
C-type lectin receptors permit interactions with pathogens and endogenous soluble proteins as well as with cell-surface ligands expressed by T cells and anatomically distinct endothelial cells, such as those found in secondary lymphoid organs.

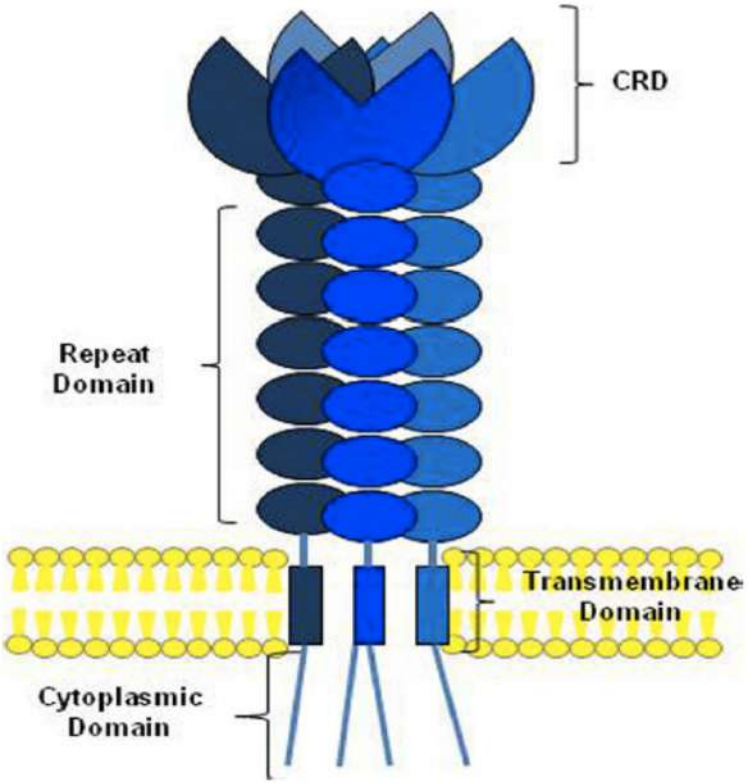
DC-SIGN a type-II C-type lectin is expressed by both macrophages and DCs. It interacts with endogenous molecules, such as ICAM-2, on endothelial cells as well as ICAM-3 on T-cells, mediating intercellular adhesion. In addition, DC-SIGN binds pathogen-associated **mannose-type carbohydrates** found on viruses, bacteria and fungi. Multimerisation of DC-SIGN and other such receptors at the cell surface might facilitate high-affinity ligand binding.

Struttura di DC-SIGN



(carbohydrate recognition domain)

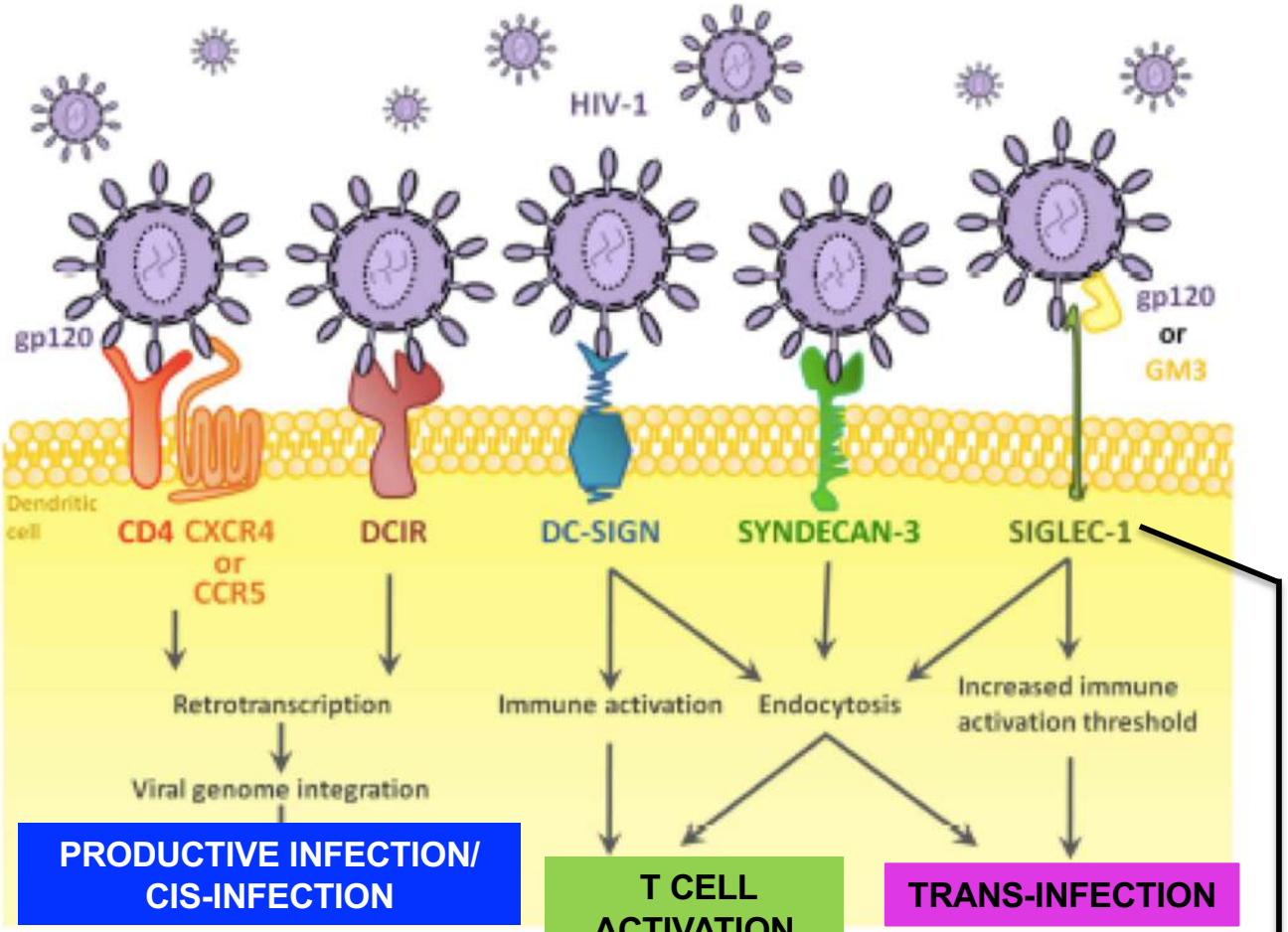




C-type lectins are transmembrane proteins that act as cell-adhesion receptors, are involved in the regulation of signalling pathways and recognize specific carbohydrate structures that are present on pathogens and self antigens.

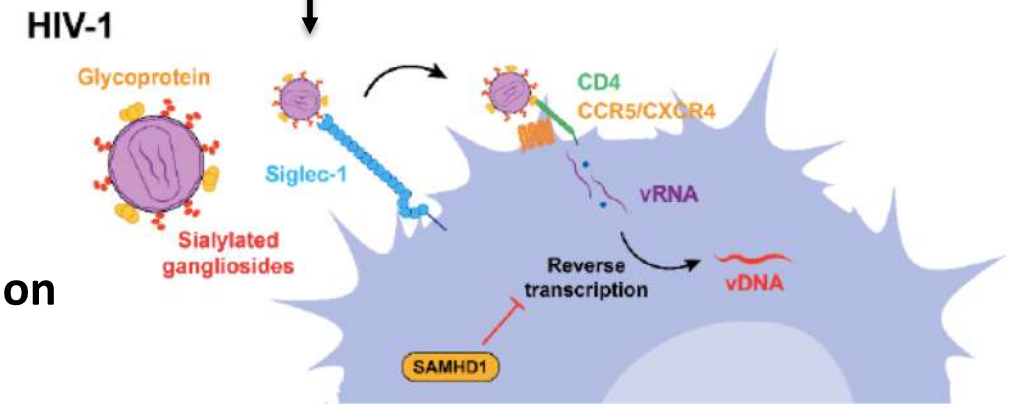
DC-SIGN=Dendritic Cell-Specific Intercellular adhesion molecule-3-Grabbing Non-integrin

Receptors and pathways implicated in the entry of HIV-1 into DCs



HIV-1 binds several different DCs surface receptors, which determines the fate of the virus.

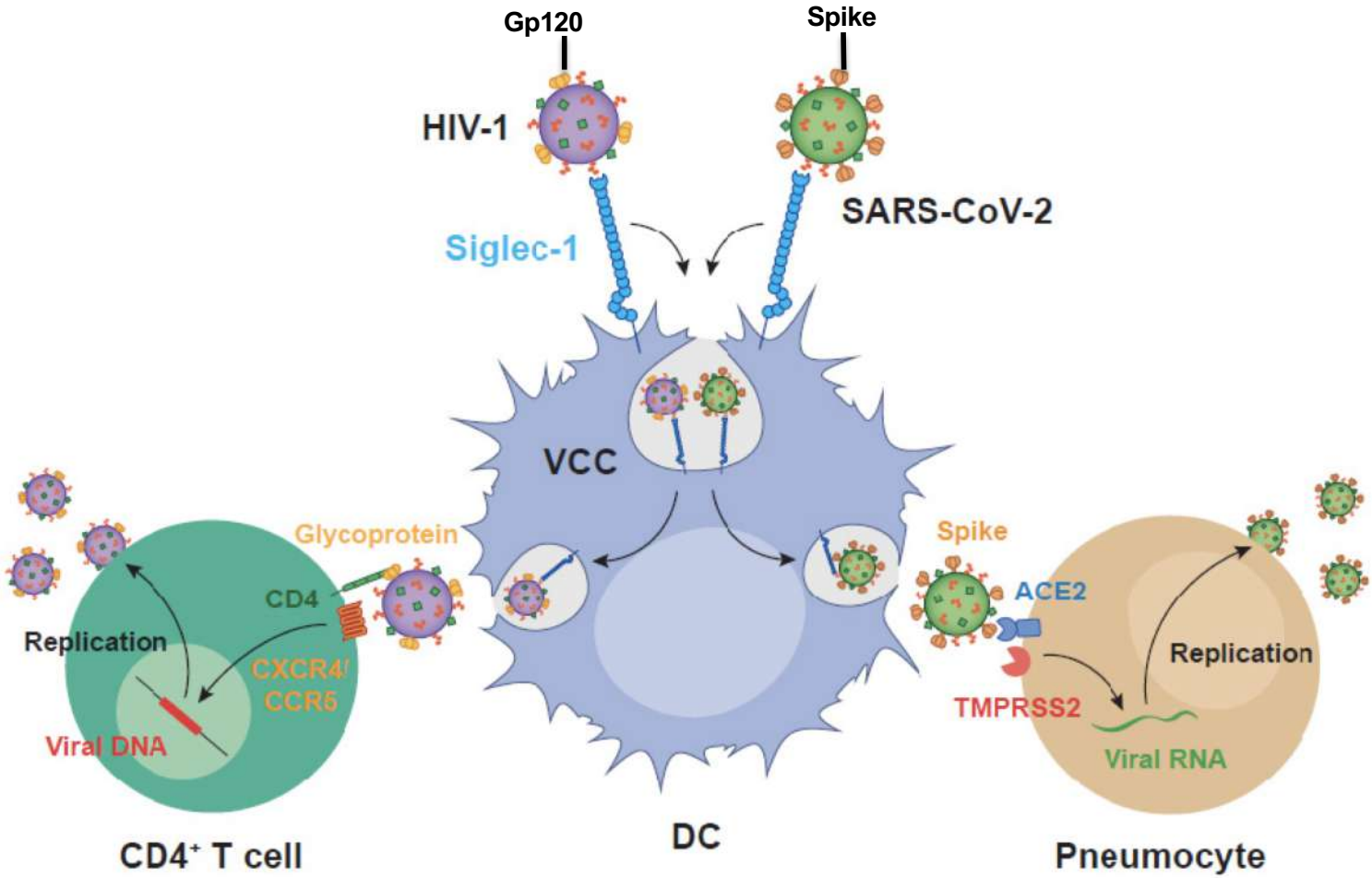
Binding to conventional HIV-1 receptor CD4, or DC-specific DCIR leads to productive infection of the cell in a very small percentage of DCs. In most cases, HIV-1 enters the DC via endocytosis after binding **DC-SIGN** or other receptors, namely, Syndecan-3 or **Siglec-1**. Binding to these receptors can lead to trans-infection or immune recognition and consequent T cell activation.



Siglec-1 >> DC-SIGN

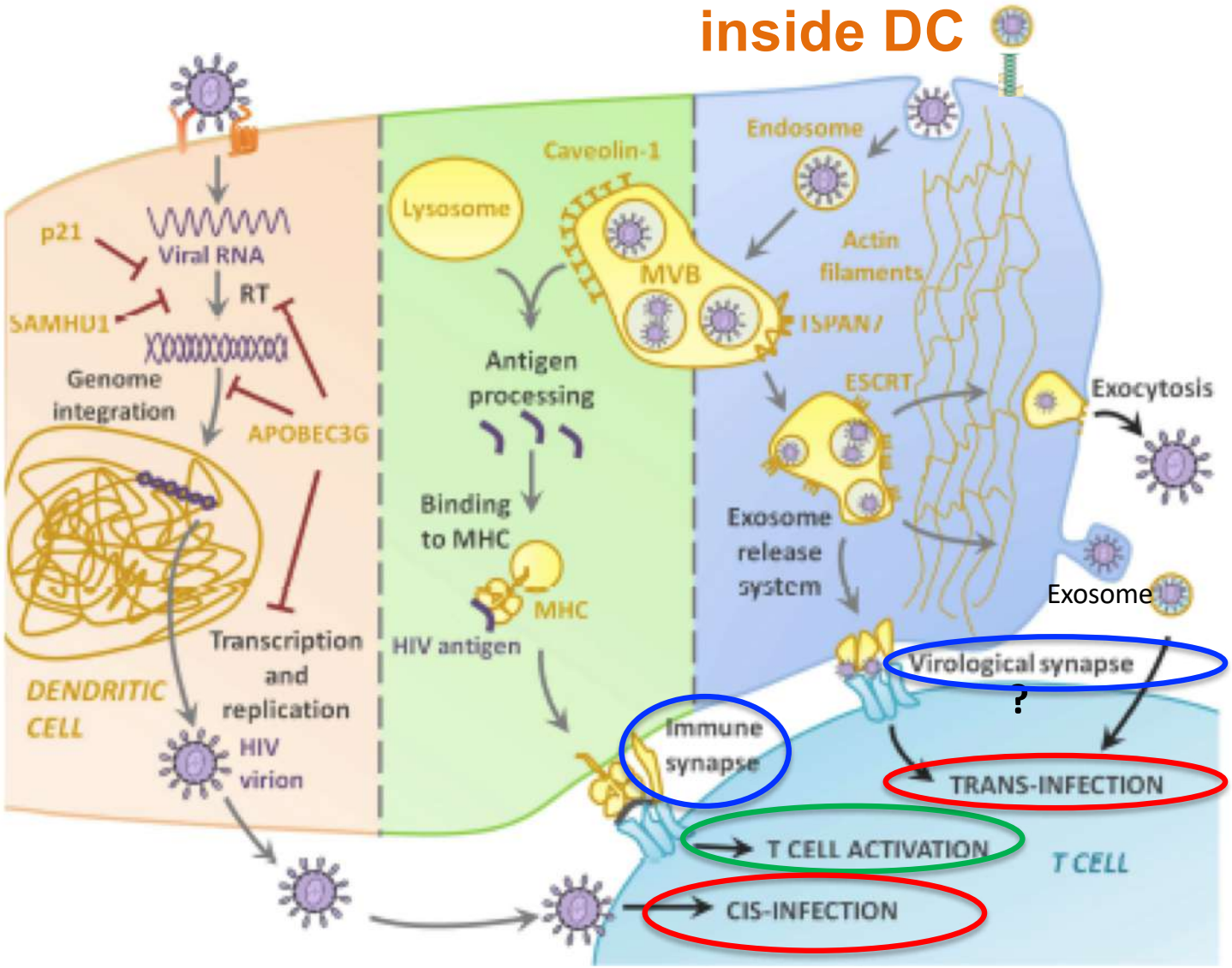
Siglec-1, more than DC-SIGN is the key molecule for DC-mediated HIV-1 trans-infection

DCs mediate trans-infection of HIV-1 and SARS-CoV-2 to target cells via Siglec-1



Viral membrane ganglioside recognition of both HIV-1 and SARS-CoV-2 particles triggers VCC formation and effective transfer to susceptible target cells that become productively infected via viral glycoprotein interaction with CD4+ receptor and coreceptors in the case of HIV-1 and ACE2 and TMPRSS2 in the case of SARS-CoV-2. HIV-1: human immunodeficiency virus type 1; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; **VCC: viral containing compartment;** DC: dendritic cell; CXCR4: CXC chemokine receptor type 4; CCR5: CC chemokine receptor 5; ACE2: angiotensin-converting enzyme 2; TMPRSS2: transmembrane protease serine 2.

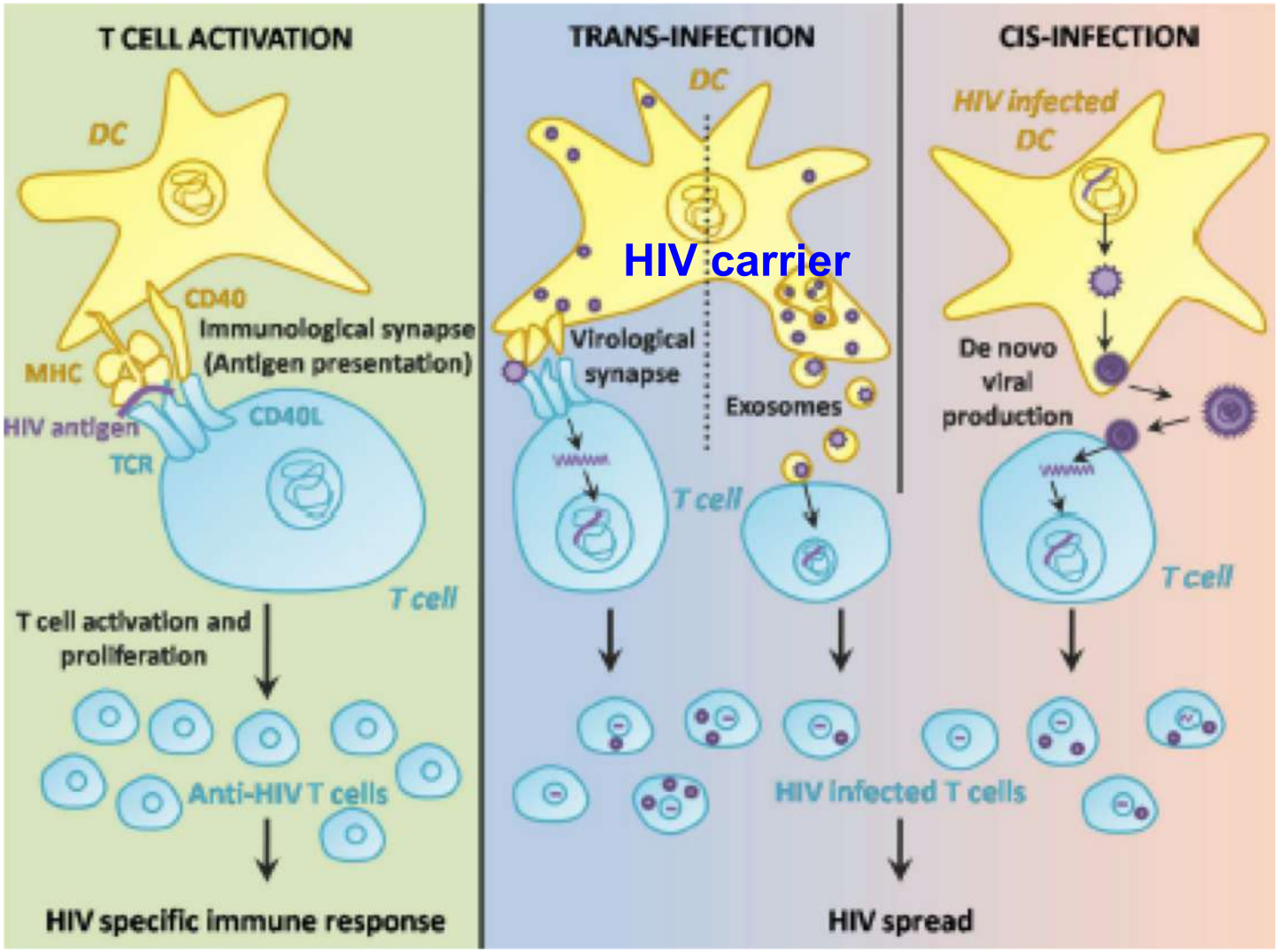
Intracellular pathways and molecular partakers of HIV-1 trip inside DC



Only a small percentage of HIV-DC interactions lead to productive infection, thanks to intracellular molecular defense at different stages of the infection, including and high-lighting the antiviral effect of p21, SAMHD1, and APOBEC3G.
Most of the times, however, the virus enters the DC by endocytosis and accumulates in multi-vesicular bodies (MVB).

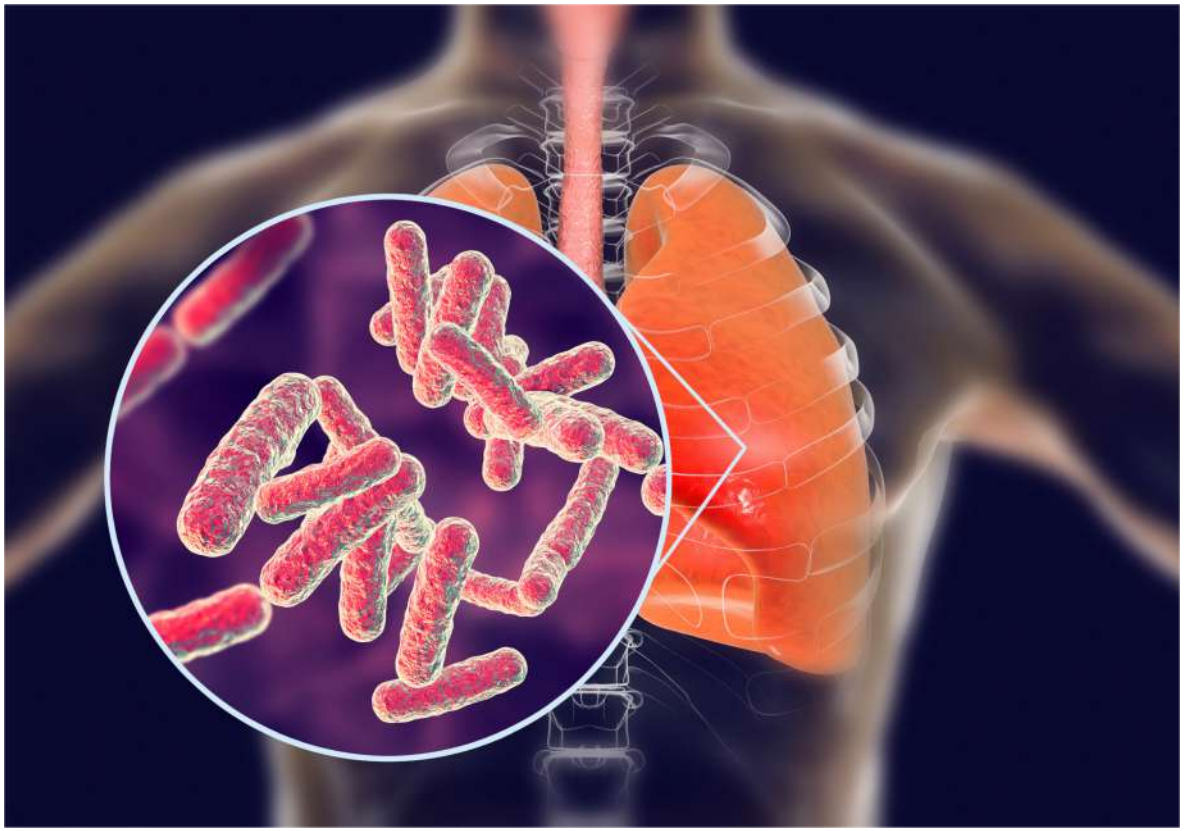
If the MVB fuses with the lysosome, the virus is recognized as antigenic and processed, resulting in viral peptides binding to MHC and showing in the membrane for Ag presentation to T cells. The key and most interesting role of DCs in HIV-1 infection is their function as **Trojan Horse**, as the virus in the endosome can use the cells as a mean of transportation to the lymph nodes, and then be released either through the virological synapse, or via exocytosis.

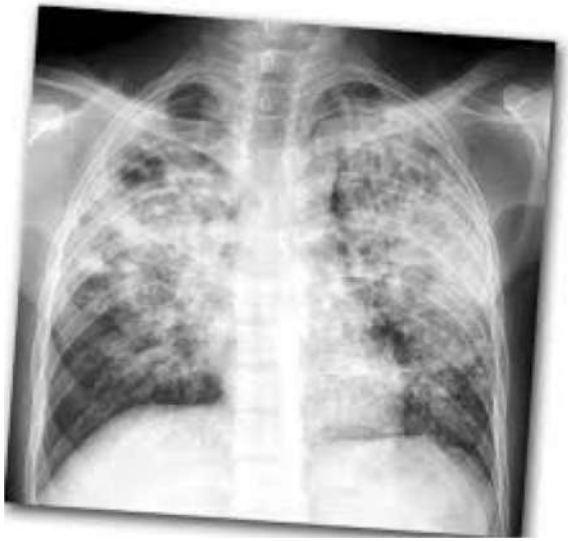
Two-faced role of DCs in HIV-1 infection after T cell contact

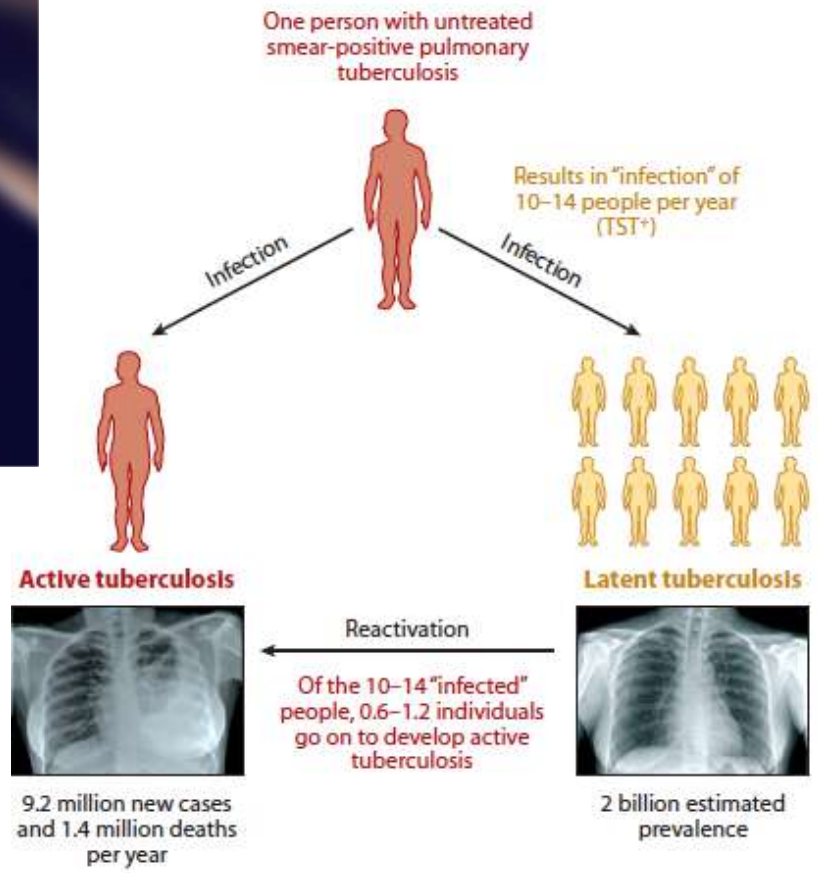


Two-faced role of DCs in HIV-1 infection after T cell contact. DC contact with T cells through the immunological synapse results in Ag presentation and a specific HIV-1 immune response. However, the DC-T cell interaction may also facilitate the transmission of the virus either from an infected DC cell to the surrounding T cells in the lymph nodes, or from a **"carrier"** DC via exosome release or infectious synapse.

Immune evasion by *Mycobacterium tuberculosis*







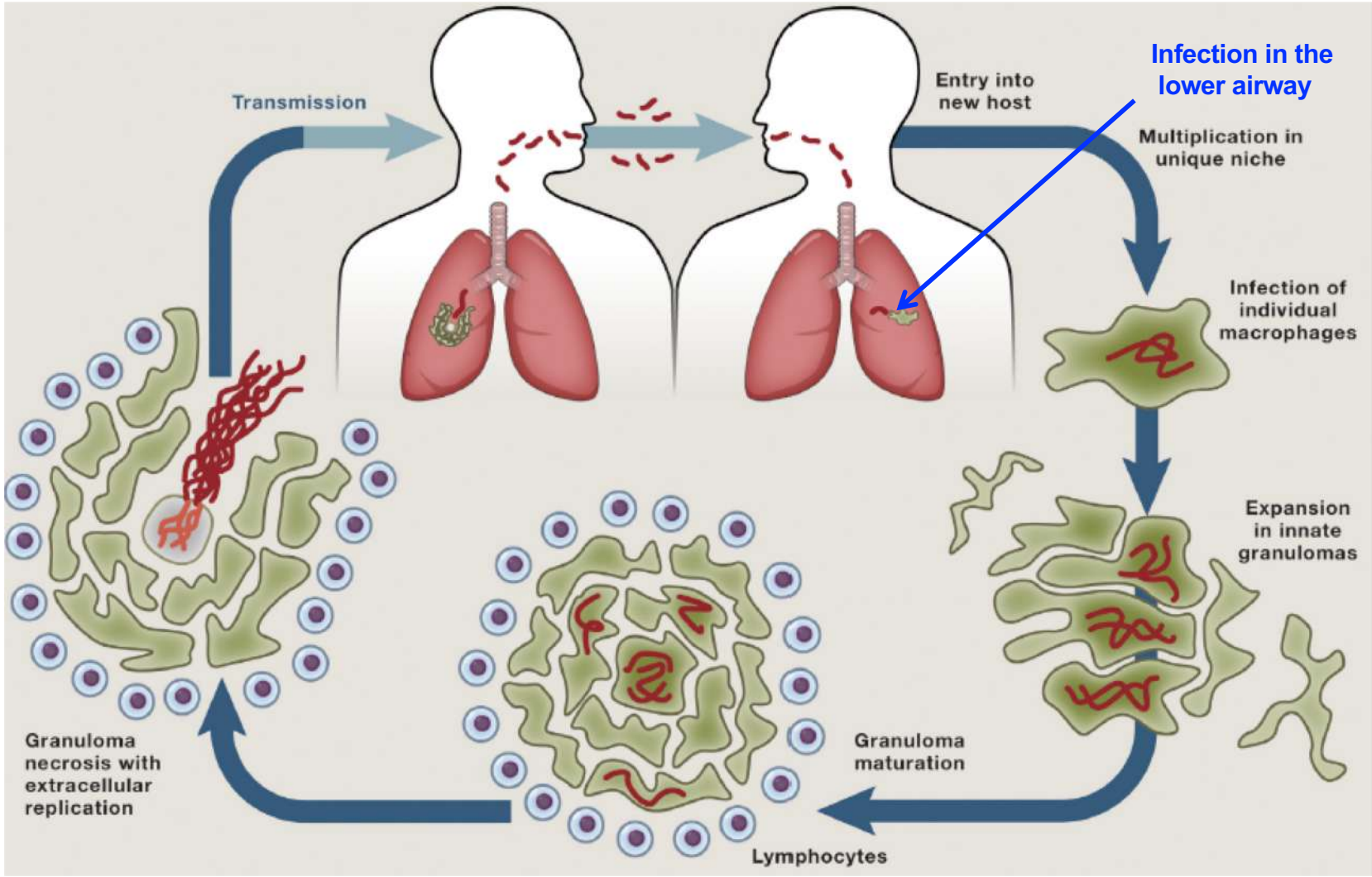
L'infezione da *M. tuberculosis* può causare:

- malattia sintomatica forma attiva
- malattia asintomatica forma latente

1/3 della popolazione mondiale è infettata da *M. tuberculosis*.

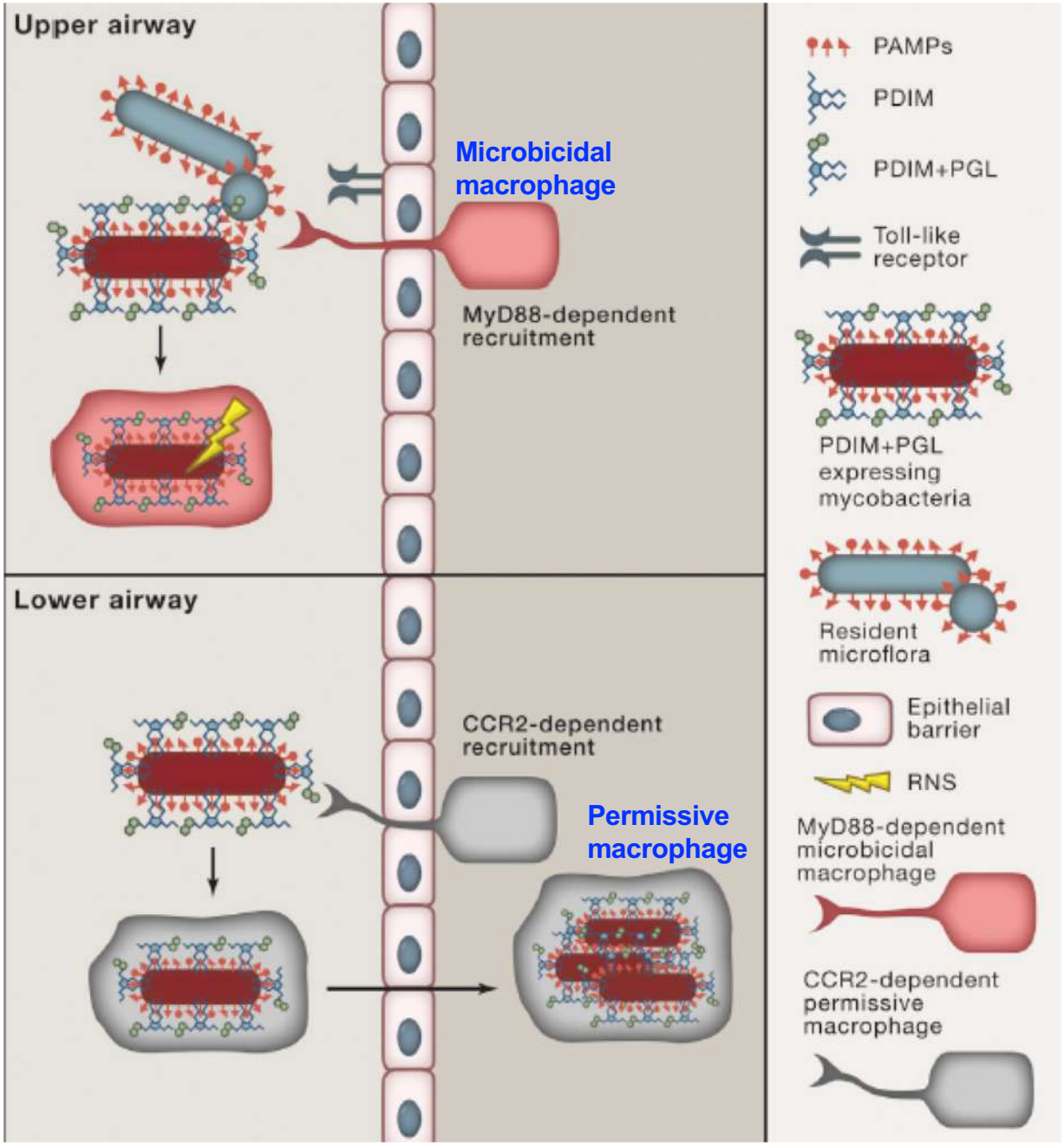
Pathogenic life cycle of *M. tuberculosis*

Invasion of the host, replication and transmission of infection



M. tuberculosis infection initiates when fine aerosol particles containing the bacteria coughed up by an individual with active disease are deposited in the lower lungs of a new host. The bacteria recruit macrophages to the surface of the lung, which become infected, and serve to transport the bacteria across the lung epithelium to deeper tissues. A new round of macrophage recruitment to the original infected macrophage is initiated, forming the granuloma, an organized aggregate of differentiated macrophages and other immune cells. The granuloma in its early stages expands infection by allowing bacteria to spread to the newly arriving macrophages. As adaptive immunity develops, the granuloma can restrict bacterial growth. However, under many circumstances, the infected granuloma macrophages can undergo necrosis, forming a necrotic core that supports bacterial growth and transmission to the next host.

M. tuberculosis Evades Commensal Bacteria to Infect Its Host



Manipulation of macrophage recruitment through coordinated use of membrane lipids

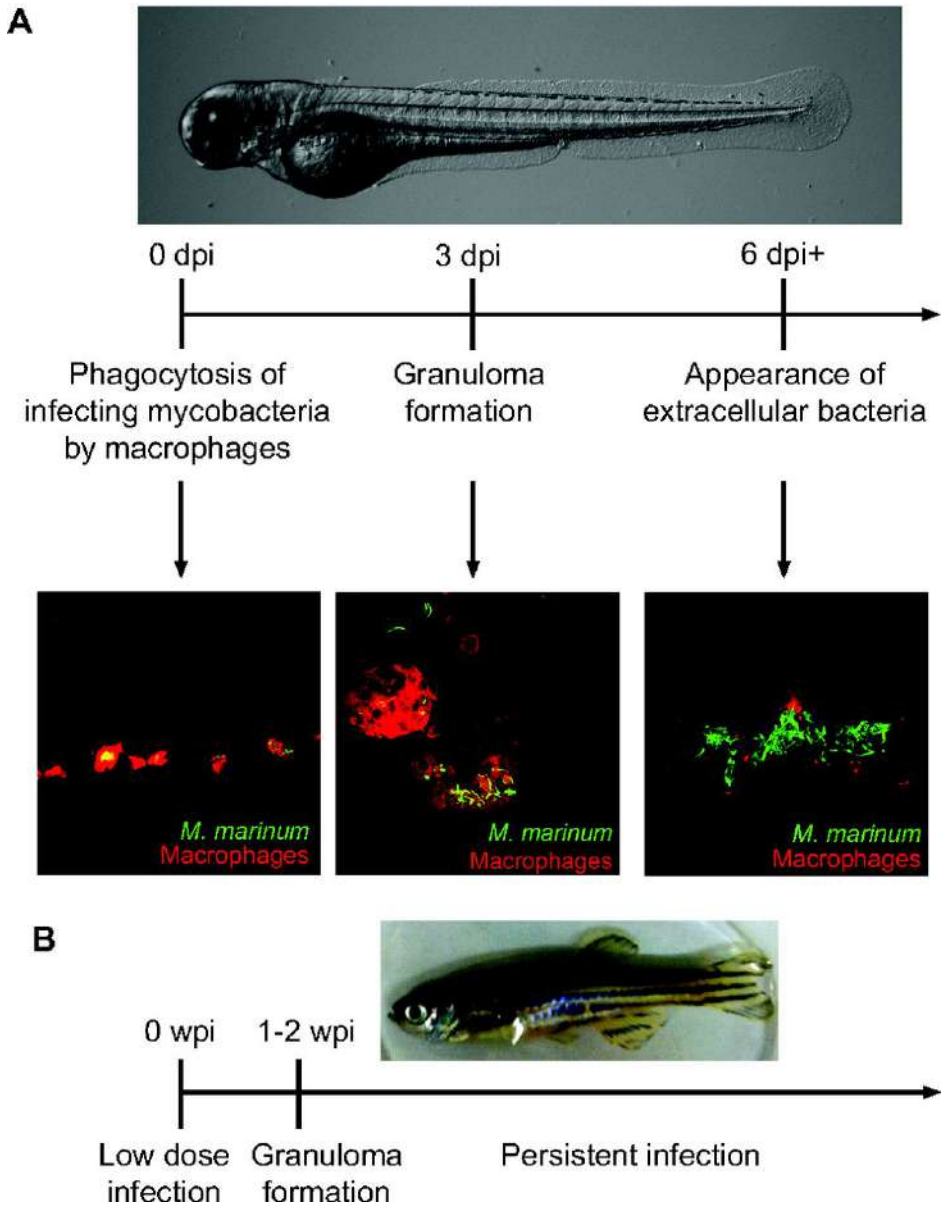
M. tuberculosis avoids the recruitment of microbicidal macrophages to the site of infection by masking its PAMPs with the **PDIM lipid**.

A related surface **lipid PGL** recruits permissive macrophages that can transport the bacteria into deeper tissues. However, the upper airways are colonized by resident microorganisms whose PAMPs recruit microbicidal macrophages. Therefore, this mycobacterial strategy to evade microbicidal macrophages is only effective if infection is initiated in the relatively sterile lower lung.

PMID = phthiocerol dimycoceroserate
PGL = phenolic glycolipid

Fit for consumption: zebrafish as a model for tuberculosis

Modeling mycobacterial infection (*Mycobacterium marinum*) in larval and adult zebrafish

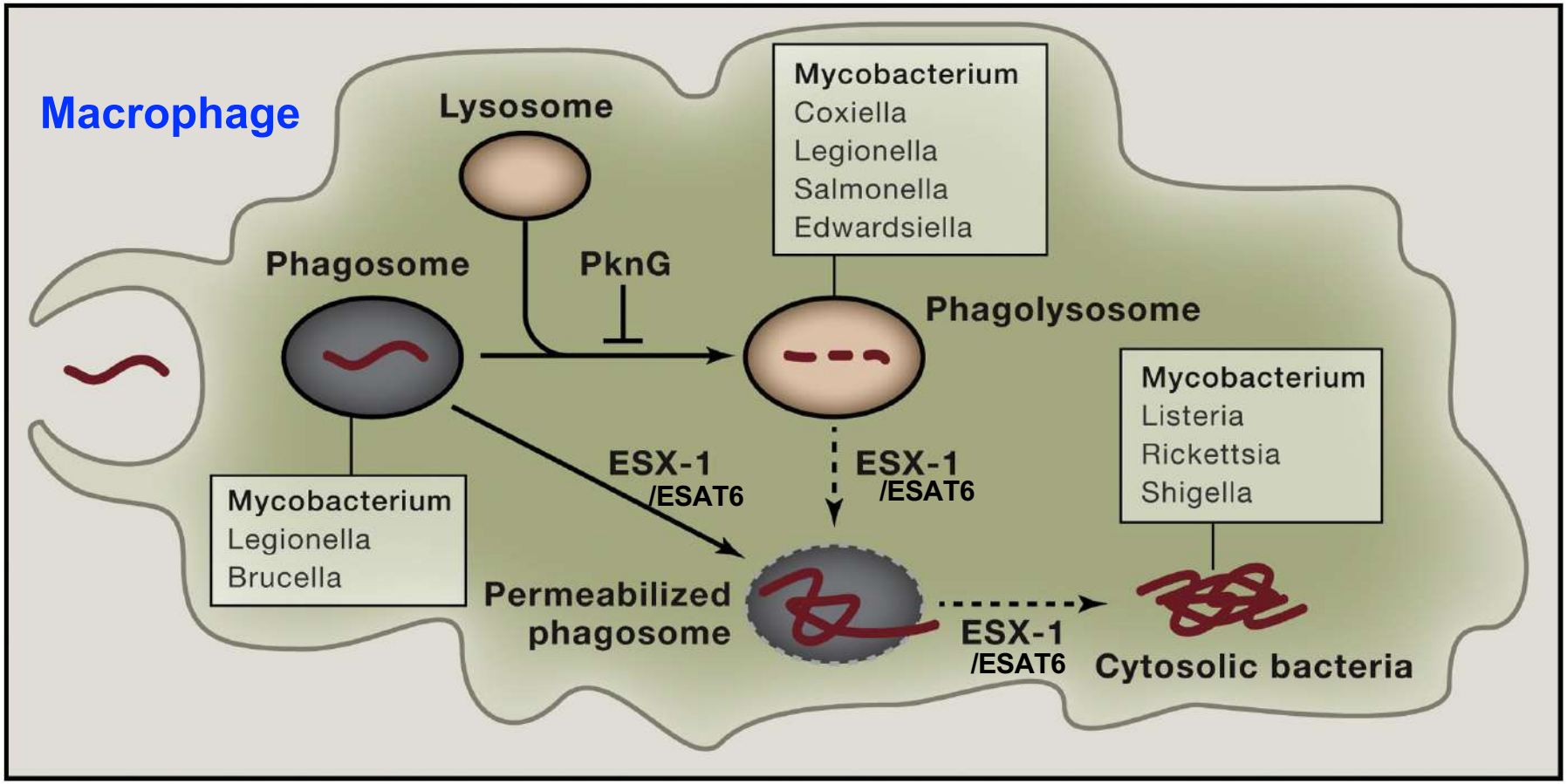


Modeling mycobacterial infection in larval and adult zebrafish. (A) From top – brightfield image of a zebrafish larva. Middle – an approximate timeline of progression of infection in zebrafish larvae, in days post-infection (dpi). Bottom – confocal images of zebrafish lines, with fluorescently labeled macrophages shown in red and infecting fluorescent mycobacteria visualized in green. Representative images display from left – scattered infected macrophages; center – macrophages aggregated into granulomas; right – the appearance of extracellular bacteria as containment fails at isolated granulomas. (B) Top – image of an adult zebrafish. Bottom – a timeline of infection progression in weeks post-infection (wpi) in the adult persistent infection model (based on the work of Parikka et al., 2012).

Mark R. Cronan, and David M. Tobin Dis. Model. Mech. 2014;7:777-784

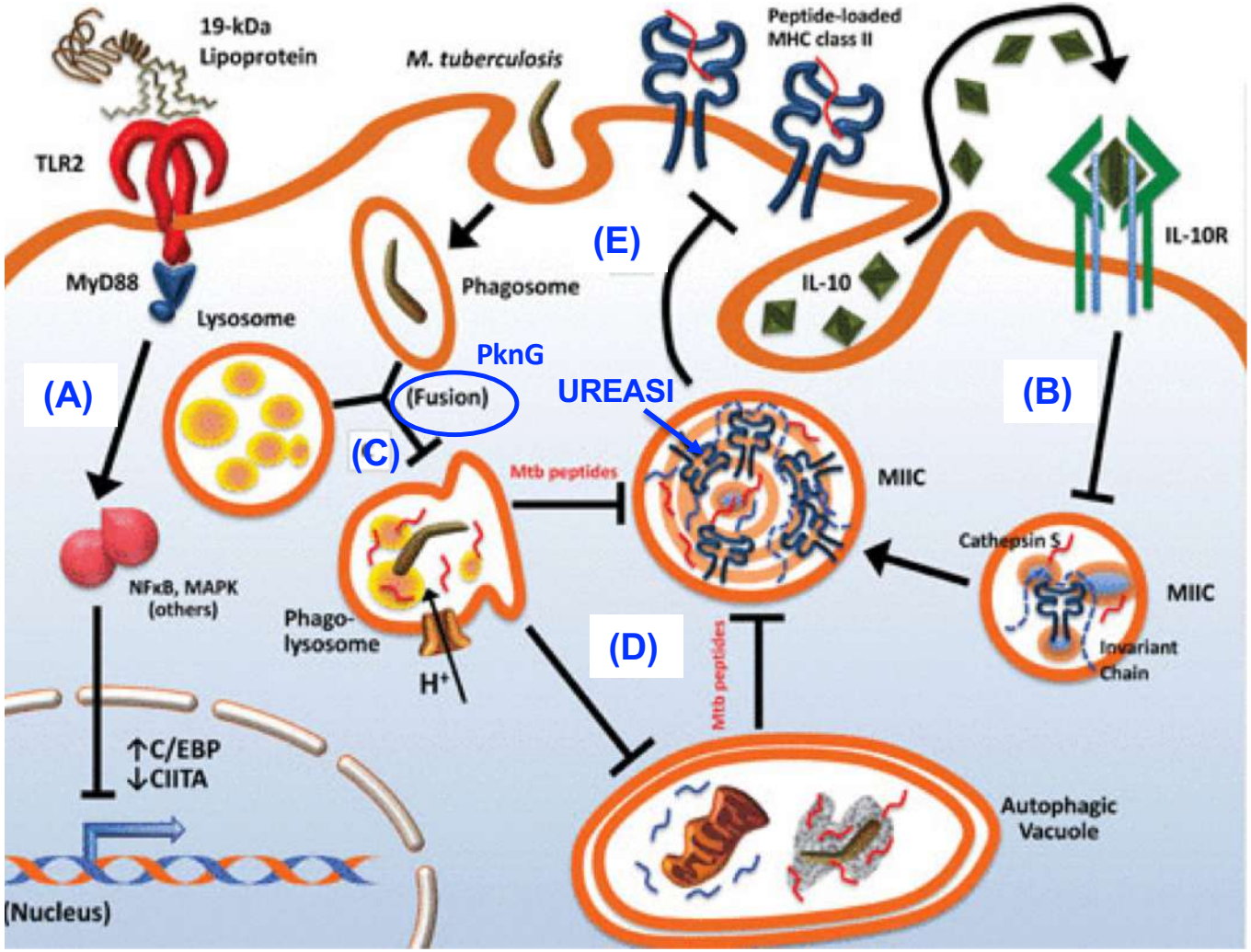
Disease Models & Mechanisms

Intracellular Niches of *M. tuberculosis*



The observed intracellular niches of *M. tuberculosis* within macrophages are shown with other pathogens occupying those niches also listed. Confirmed trafficking pathways are indicated with continuous arrows and putative ones with dashed arrows. Pathways dependent on the mycobacterial ESX1 secretion system are indicated.

M. tuberculosis elude la risposta immunitaria utilizzando diversi meccanismi



Disruption of MHC class II presentation by *M. tuberculosis*. The various steps in the MHC class II processing and presentation pathway that are known or postulated to be influenced by *M. tuberculosis* infection are illustrated.

(A) New synthesis of MHC class II molecules is blocked by TLR2 signaling due to mycobacterial products such as the 19-kDa lipoprotein.

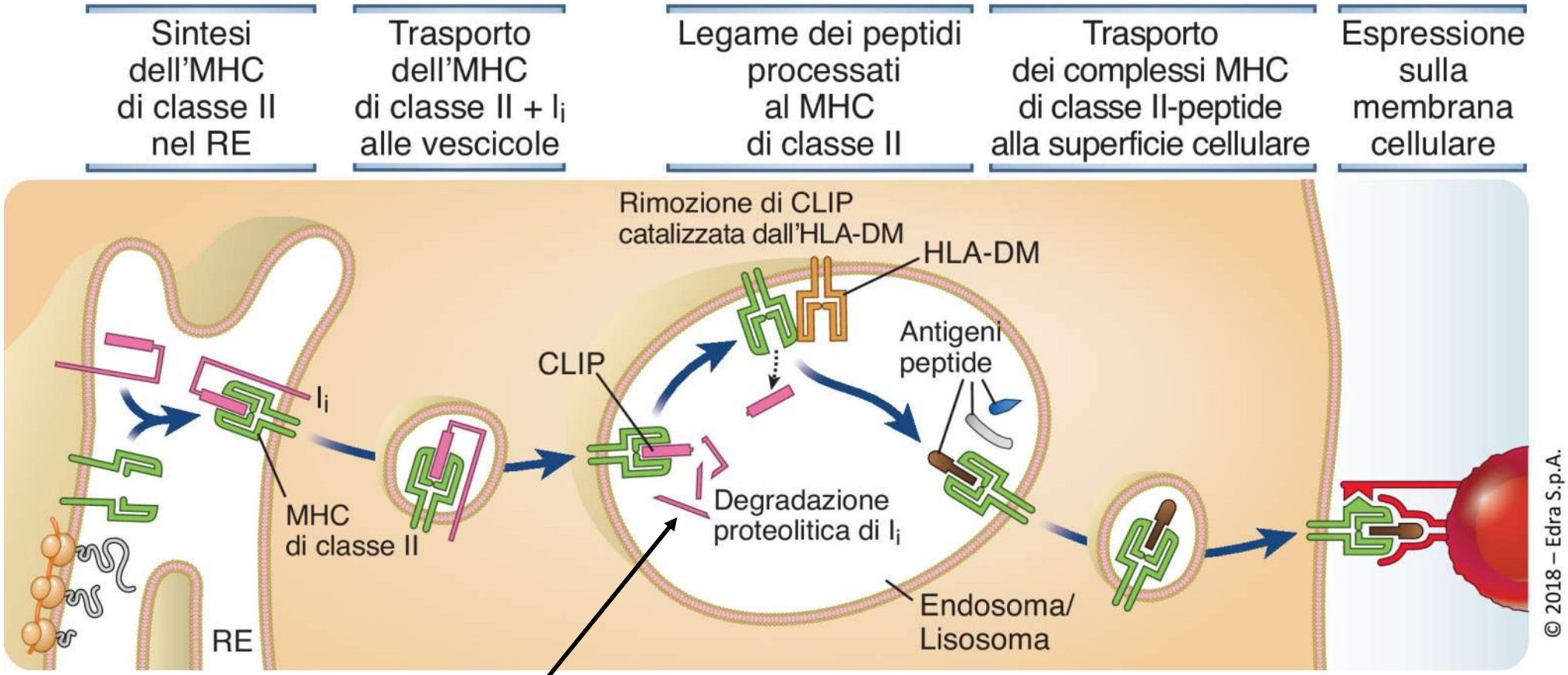
(B) Intracellular trafficking of MHC class II is disrupted by the suppression of cathepsin S, which is due to induction of IL-10 by mycobacterial infection.

(C) Generation of peptide antigens for loading onto MHC class II in relevant endocytic compartments (MIIC) is also inhibited by several effects of mycobacterial infection, including inhibition of phagosome-lysosome fusion, by neutralization of phagosomal pH by bacterial urease, and by blockade of recruitment of the vacuolar proton ATPase.

(D) Proposed inhibition of autophagy and autophagic vacuole formation also eliminates a potential source of antigenic peptides that can load MHC class II molecules.

(E) The reduction of peptide antigen availability and incomplete cleavage of MHC class II associated invariant chain (Ii) resulting from cathepsin S suppression result in a reduced transport of stable peptide-loaded MHC class II molecules to the APC surface.

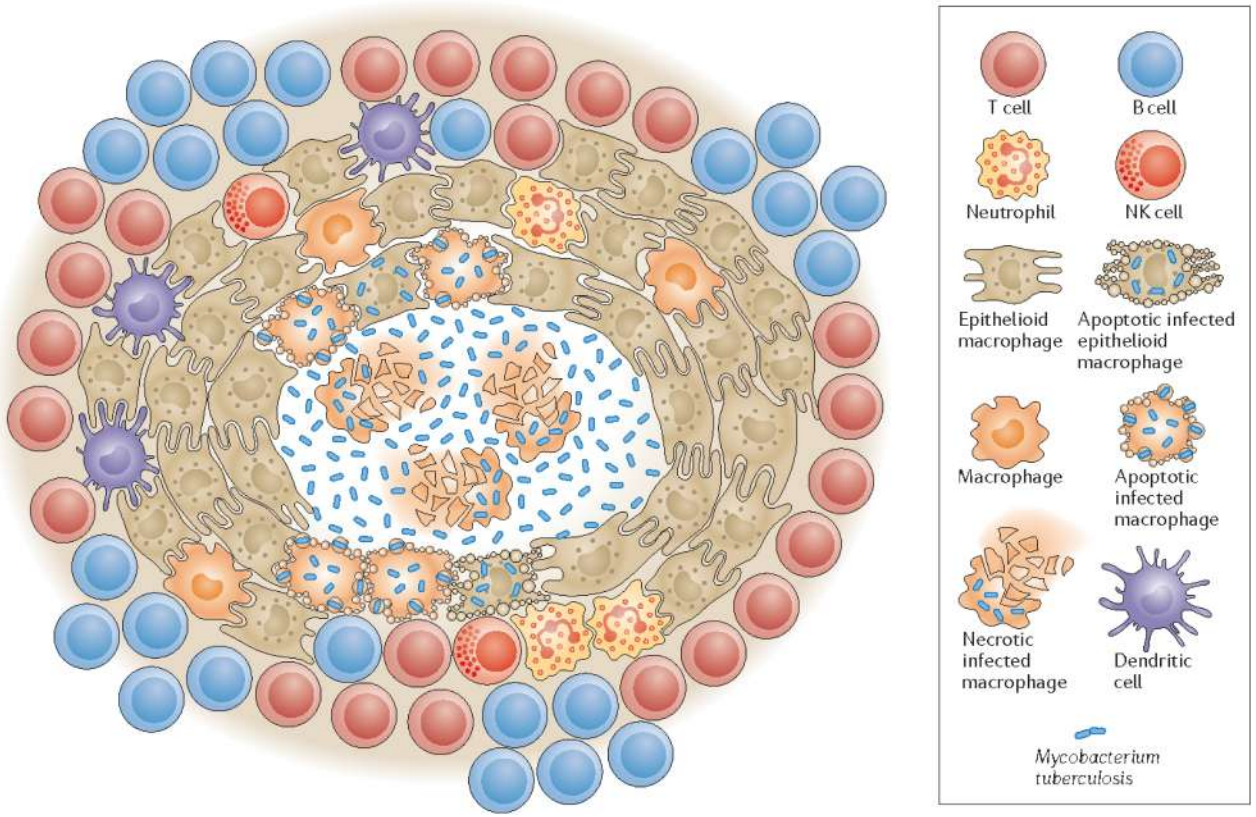
La catepsina S media gli step finali della degradazione della catena invariante (I_i) fino alla produzione del CLIP

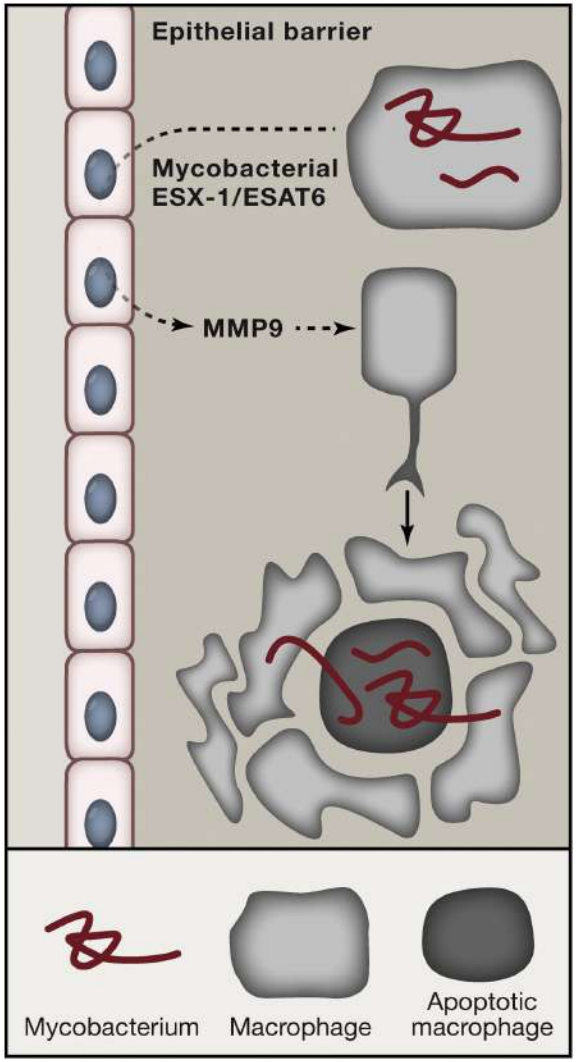


catepsine (fondamentale l'azione della catepsina S)

Mycobacteria Exploit the Granuloma to Expand Their Numbers in early infection

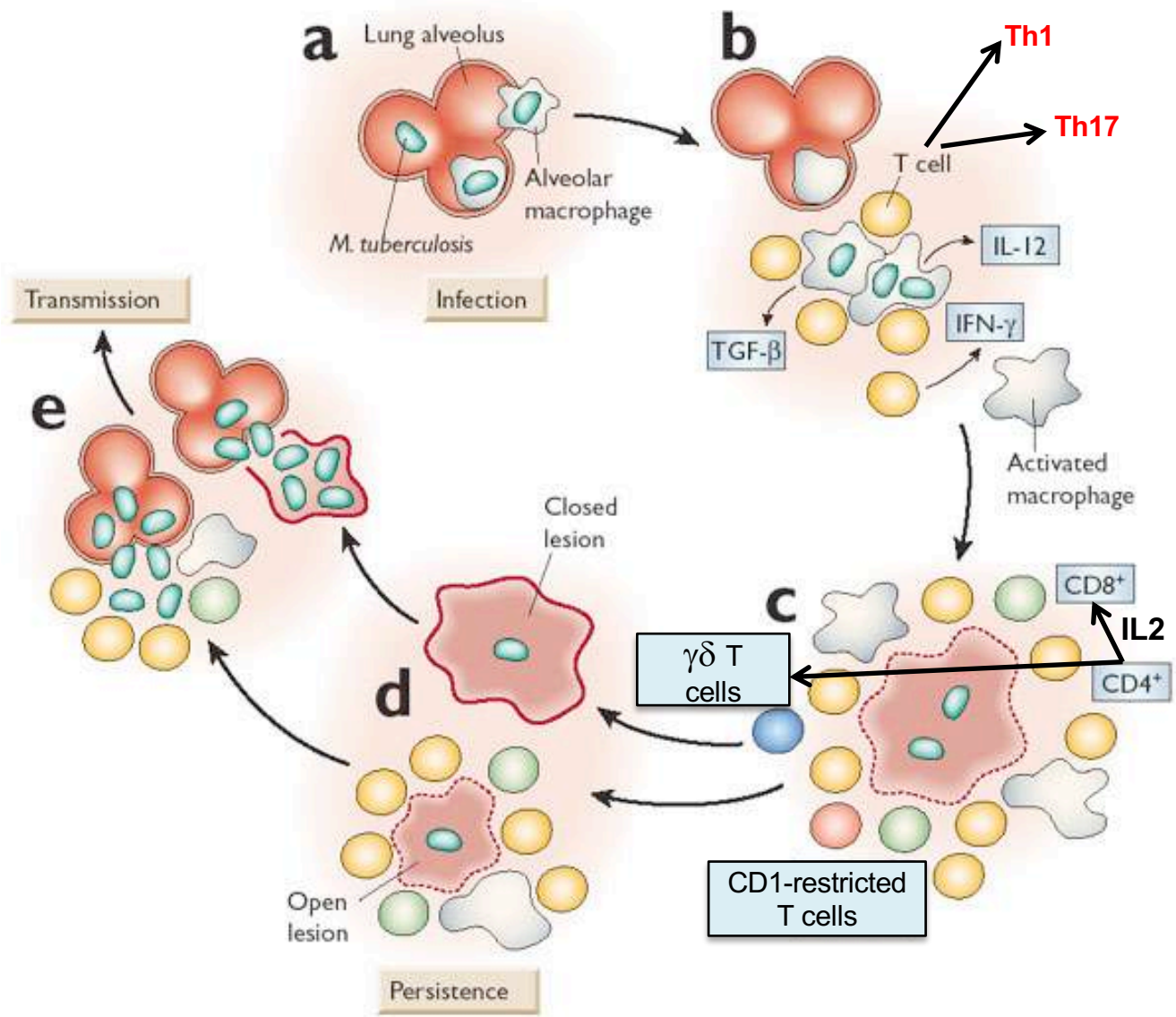
Mycobacteria within infected macrophages induce in an ESX1-dependent fashion MMP9 expression in epithelial cells surrounding the nascent granuloma.
MMP9 stimulates the recruitment of new macrophages to the granuloma. Multiple new arrivals phagocytose the bacterial contents of a given dying infected macrophage, thus spreading the bacteria to new macrophages and providing them new expansion niches.





A classical tuberculosis granuloma. The hallmark tuberculosis granuloma is a highly organized collection of immune cells that aggregate around a central necrotic core.

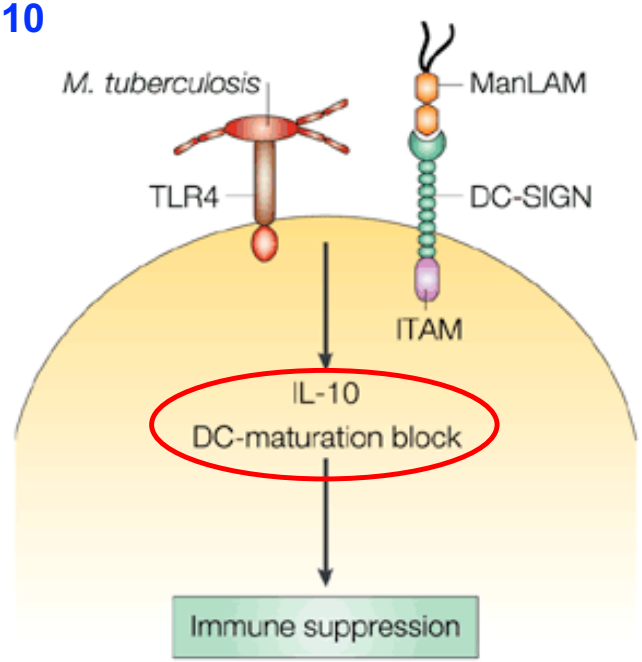
Persistenza del *mycobacterium tuberculosis*



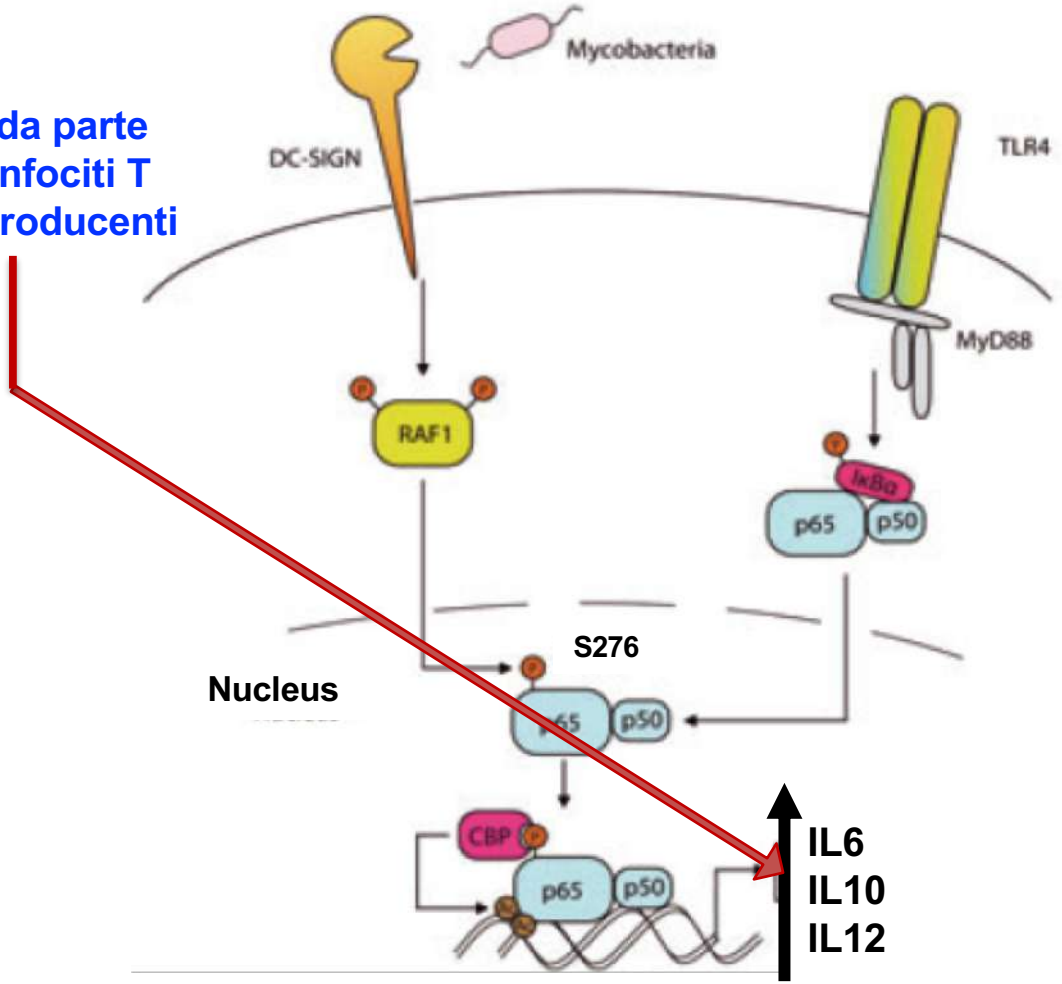
(**a**) Infecting mycobacteria taken up by alveolar macrophages in the lung resist killing by subverting phagosome maturation and by the protective effect of their lipid-rich cell wall.
(**b**) Inflammatory signaling in response to mycobacterial components results in the recruitment of TH1 T cells. T cell-mediated activation of macrophages enhances their ability to control mycobacteria.
(**c**) Remaining viable mycobacteria are sequestered within a granuloma made up of macrophages and a variety of T cell subsets.
(**d**) *M. tuberculosis* is able to persist in an asymptomatic form within the host over many decades. It is likely that mycobacteria are held in a nondividing or slowly dividing state in open lesions or in closed lesions walled-off by fibrosis.
(**e**) Reduced immunity is associated with reactivation disease. Lack of immunosurveillance may stimulate renewed replication of mycobacteria in open lesions. For mycobacteria in closed lesions, liquefaction and breakdown in the absence of immune surveillance will result in disease. Once the disease process is underway, immunopathology contributes to tissue damage and ultimately efficient aerosol transmission to a new host.

DC-SIGN signaling modulates TLR signaling during *M. tuberculosis* infection

La produzione simultanea di IL-10 e IL-12 da parte delle DC disturba il differenziamento dei linfociti T naive verso il fenotipo Th1 ed induce Th producenti IL10



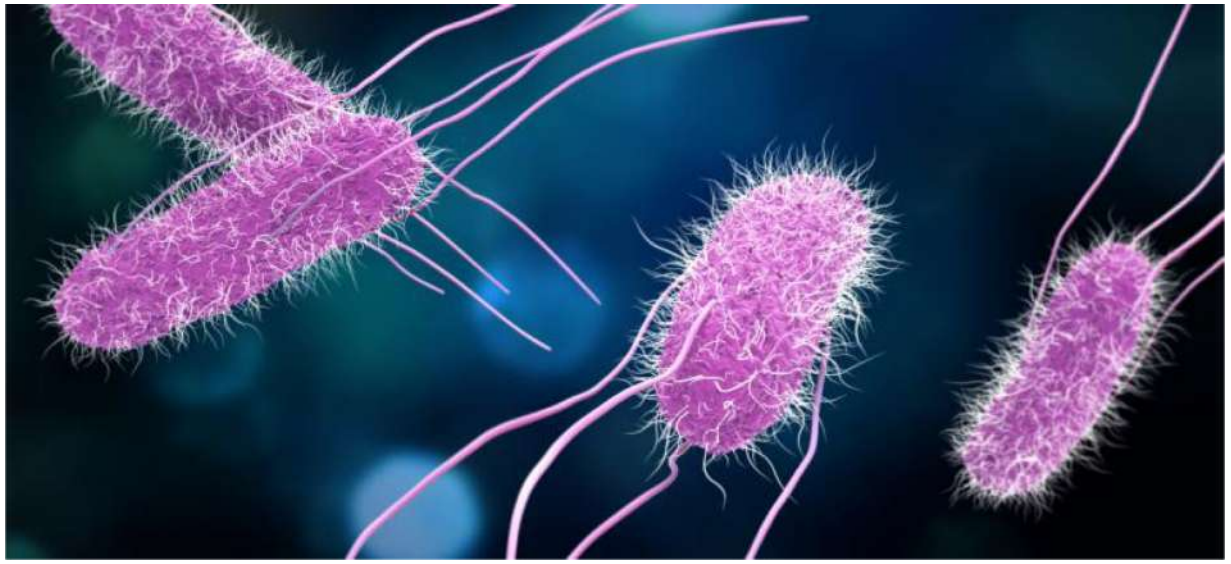
DC-SIGN and TLR4 might inhibit each other after the recognition of *Mycobacterium tuberculosis*. Ligation of DC-SIGN by the *M. tuberculosis* virulence factor mannose-capped cell-wall component lipoarabino-mannan (ManLAM) reduces TLR4-triggered DC maturation and enhances the production of interleukin-10 (IL-10) disfavouring a TH1-cell-mediated response and survival of the pathogen.



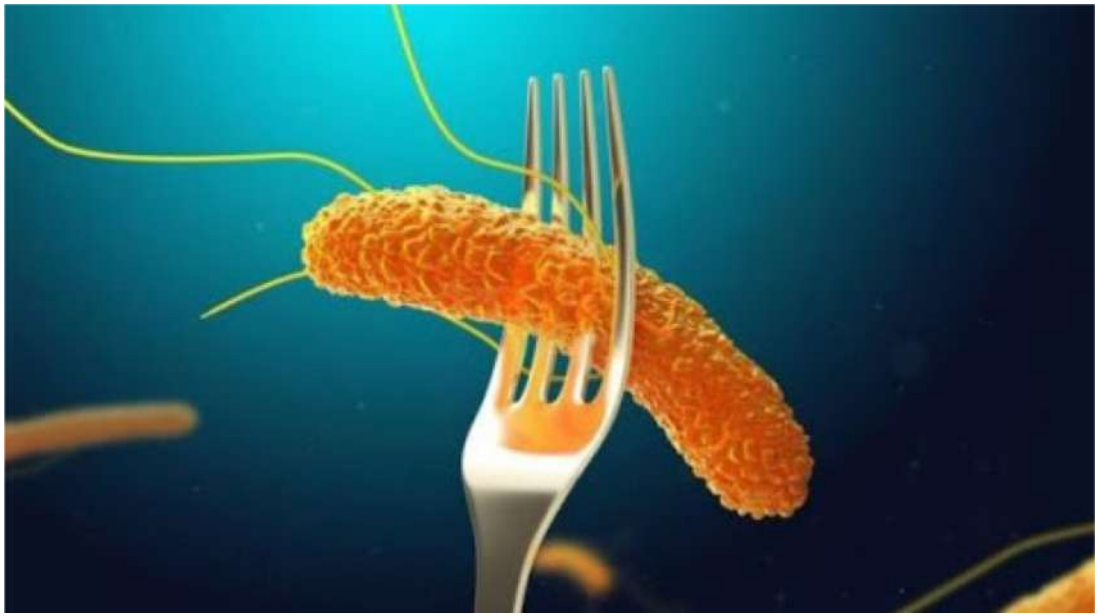
DC-SIGN binds ligands such as mycobacteria. DC-SIGN induces RAF1 phosphorylation, which modulates TLR induced NF-κB activation. Upon TLR stimulation the canonical NF-κB subunit p65 is released from its inhibitor and translocates to the nucleus. Phosphorylated RAF1 induces p65 phosphorylation at Ser276 that functions as a binding site for the histone acetylase CBP. Acetylation of p65 induces enhanced and prolonged *Il6, Il10* and *Il12ab* transcription. CBP, CREB binding protein; DC-SIGN, DC-specific ICAM3-grabbing nonintegrin; MyD88, myeloid differentiation primary response protein 88; NF-κB, nuclear factor κ B; IκBα, inhibitor of NF-κBα; TLR, Toll-like receptor.

Meccanismi di immunoevasione e immunosoppressione del *Mycobacterium tuberculosis* mediante:

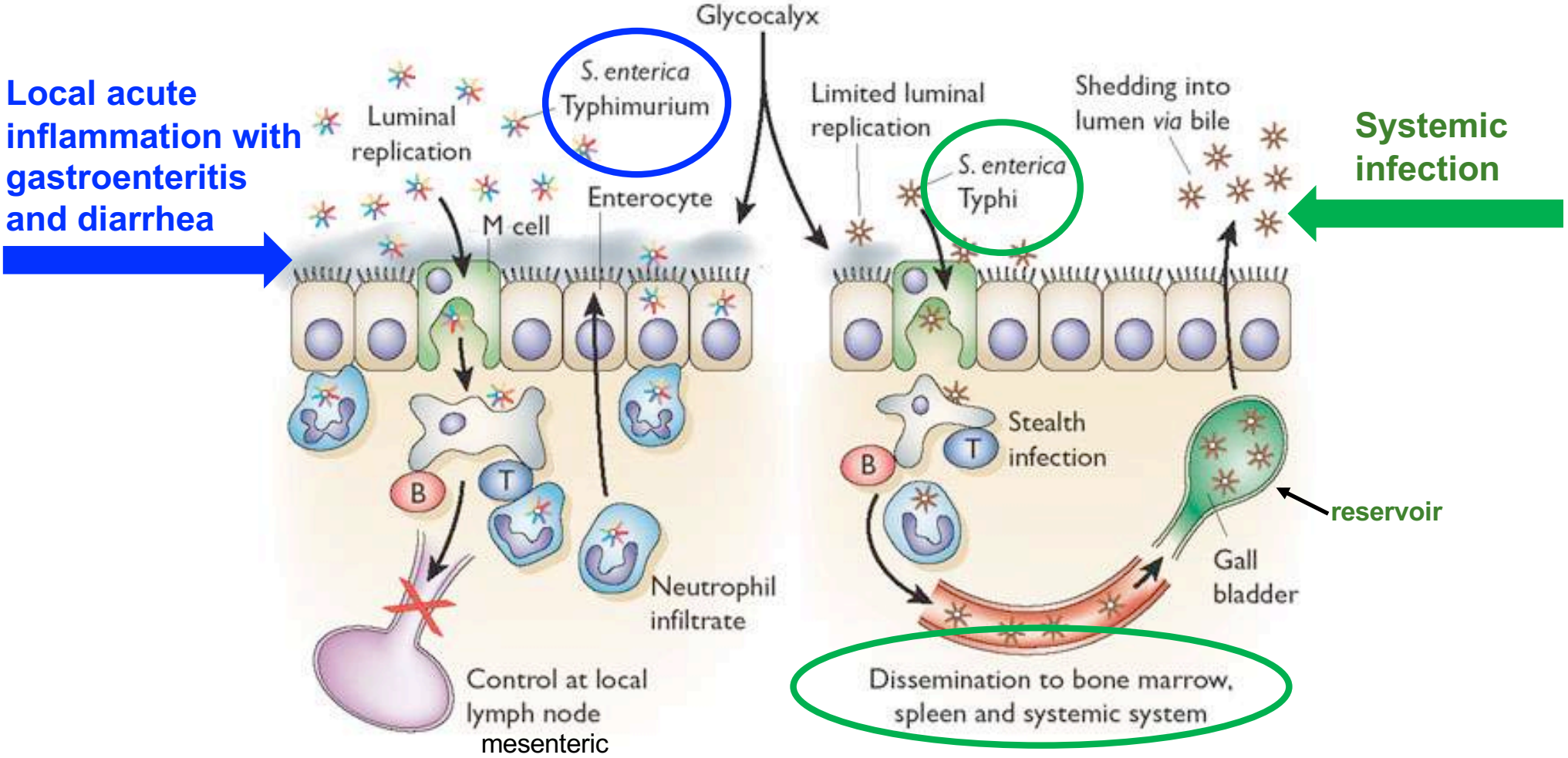
- Evasione dal signaling dei TLRs, dei NLRs e dei recettori lectinici di tipo C
- Inibizione delle funzioni effettrici dei macrofagi e blocco della maturazione e dell'acidificazione del fagolisosoma
- Interferenza con la presentazione antigenica mediata dalle molecole MHC di classe II e classe I
- Manipolazione dell'apoptosi e dell'autofagia
- Inibizione della maturazione delle cellule dendritiche
- Inibizione della produzione di ROS (specie reattive dell'ossigeno) e resistenza all'ossido nitrico e agli intermedi reattivi dell'azoto



S. enterica Typhi
versus
S. enterica Typhimurium

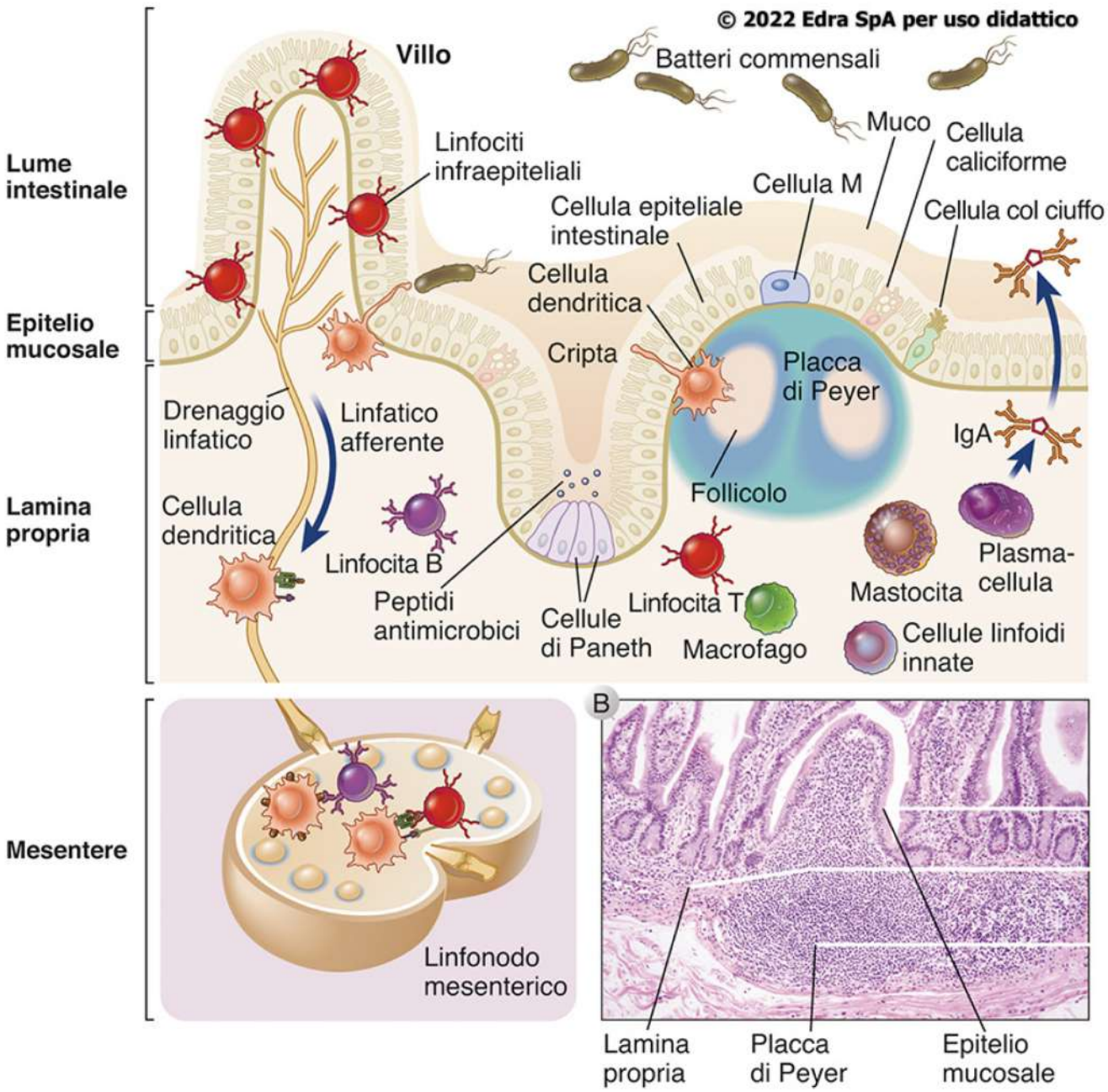


Strategie adottate da *S. enterica* Typhi per la persistenza nell'ospite e differenze con *S. enterica* Typhimurium



On the left of the figure, the acute infection (human gastroenteritis) mediated by *S. enterica* Typhimurium involves bacteria replicating freely in the intestine lumen and using **multiple attachment factors (represented by multicolored tips or fimbriae).** ***S. enterica*** **Typhimurium targets both enterocytes and M cells for invasion**, but is stopped at the mesenteric lymph nodes. Neutrophils are quickly attracted to the invasion site and inflammation follows, leading to diarrhea. ***S. enterica*** **Typhi has dispensed with many attachment and shedding factors** and may preferentially target a limited number of host cell types that favor dissemination to deeper tissues. ***S. enterica*** **Typhi can persist in the bone marrow for extended periods and in the gall bladder for life**. T, T cell; B, B cell.

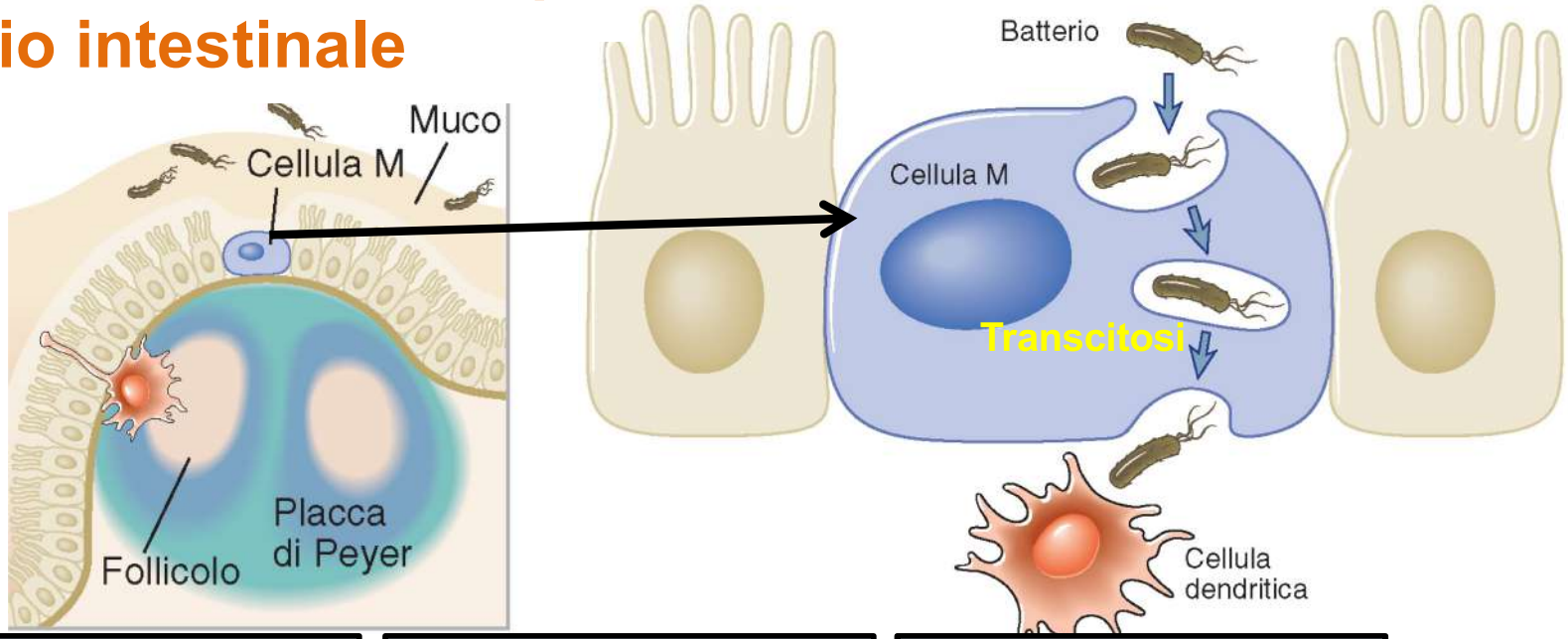
Tessuti linfoidi associati alle mucose: placche del Peyer nell'intestino tenue

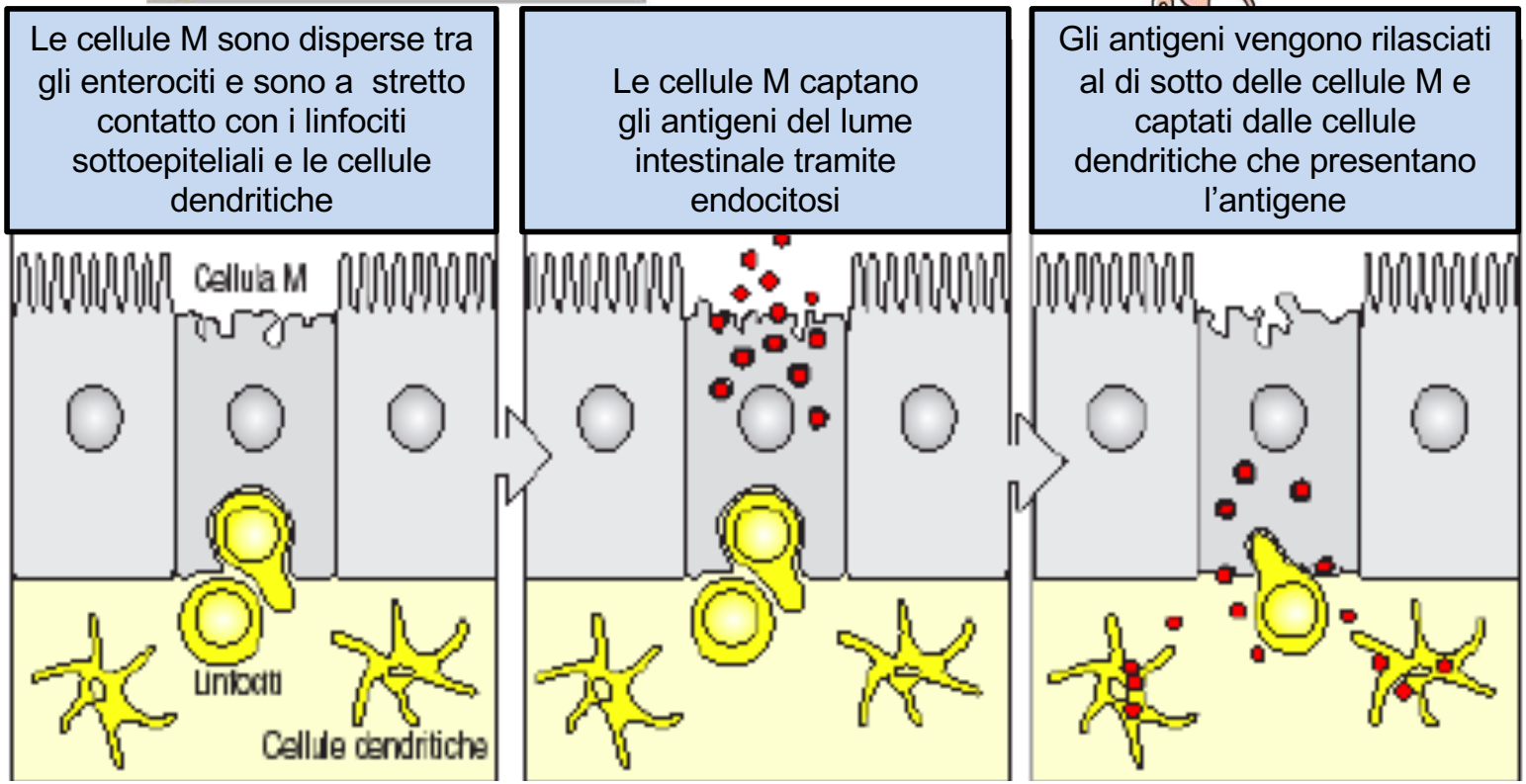


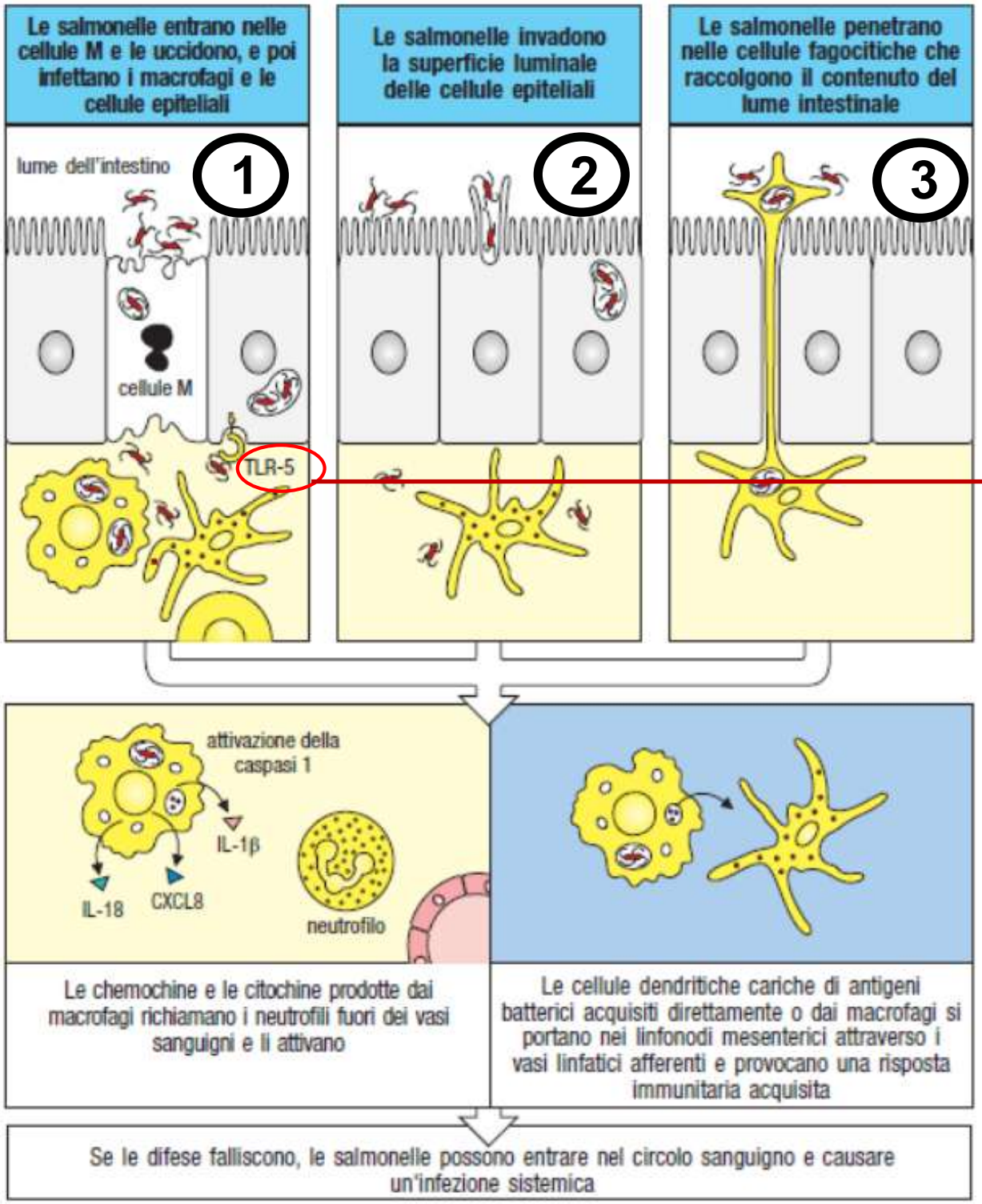
Il sistema immunitario gastrointestinale. **A**, Raffigurazione schematica delle componenti cellulari del sistema immunitario mucosale nell'intestino. Le caratteristiche principali consistono in una barriera epiteliale rivestita da muco secreto, DC e cellule M che campionano antigeni, cellule col ciuffo che rispondono agli elminti secernendo citochine, varie cellule sentinella innate e linfociti presenti nella lamina propria al di sotto dello strato epiteliale, tessuti linfoidi organizzati associati alla mucosa al di sotto della barriera epiteliale, come le placche di Peyer, il drenaggio dei linfonodi mesenterici e delle plasmacellule al di sotto dell'epitelio che secernono le immunoglobuline A (IgA), che vengono trasportate nel lume. I dettagli del campionamento dell'antigene da parte delle DC e delle cellule M, la struttura delle placche di Peyer, la migrazione dei linfociti tra la mucosa e i linfonodi mesenterici e la secrezione e il trasporto di IgA sono tutti descritti in dettaglio in questo capitolo.

B, Microfotografia del tessuto linfoide mucoso nell'intestino umano. Tali aggregati di tessuto linfoide sono presenti in tutto il tratto gastrointestinale.

Le cellule M sono cellule specializzate dell'epitelio intestinale







***S. enterica* Typhimurium oltrepassa la barriera intestinale utilizzando più vie e determina una forte risposta infiammatoria locale**

TLR → **NF-kB**

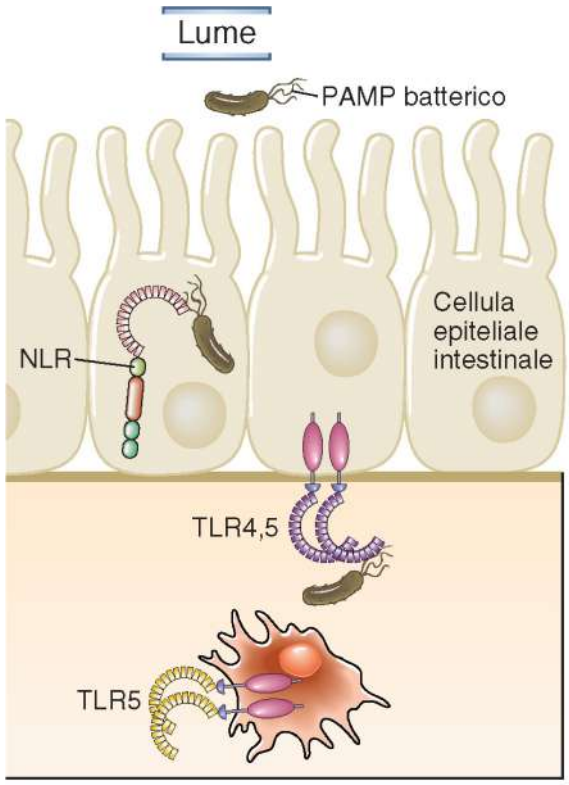
NOD

↓

Chemochine per il reclutamento leucocitario (IL8) e citochine che sostengono un processo infiammatorio acuto

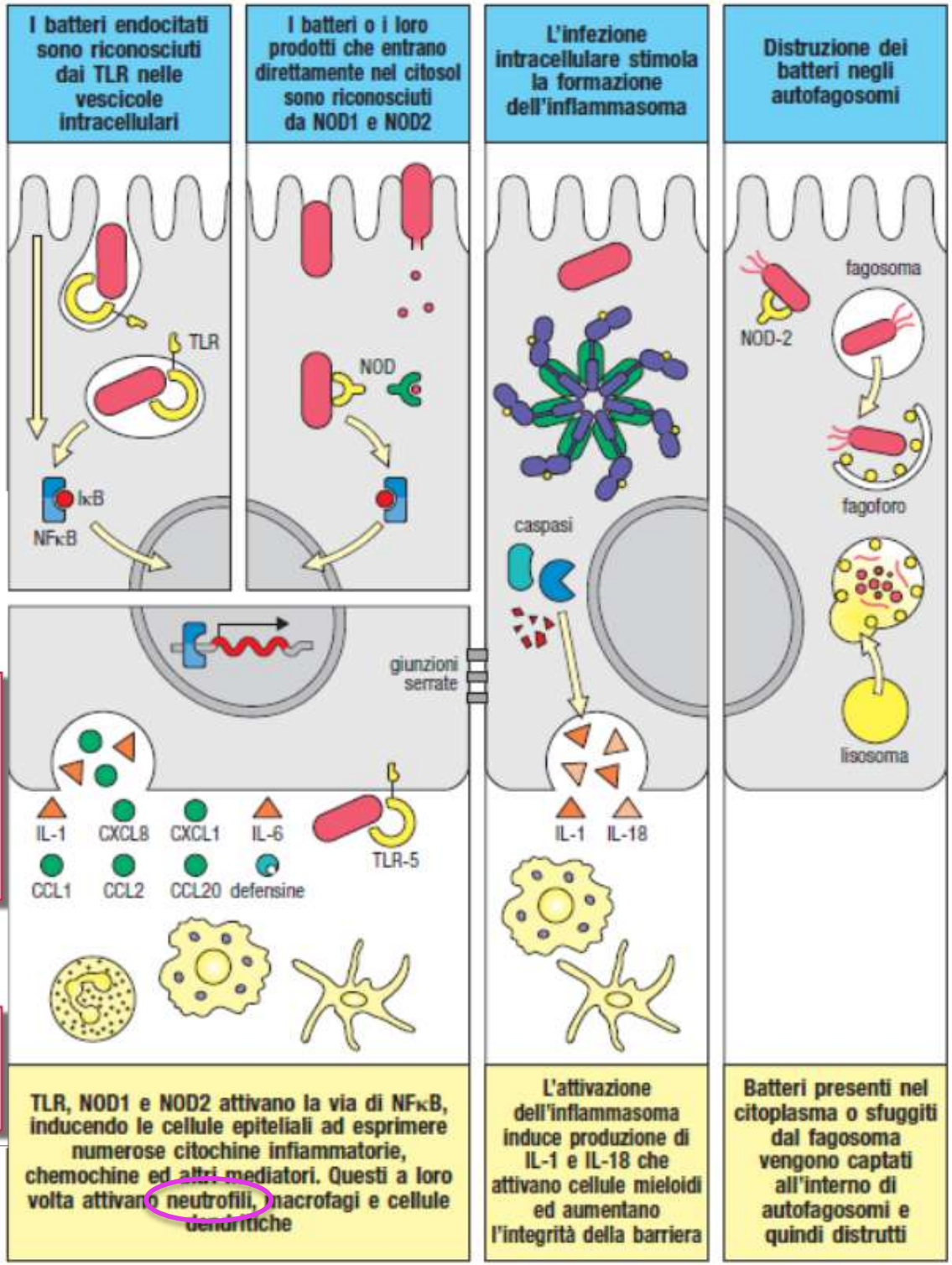
***Invece, S. enterica* Typhi non infetta gli enterociti e non provoca una risposta infiammatoria locale ma supera i linfonodi mesenterici e causa infezione sistemica**

Le cellule epiteliali giocano un ruolo cruciale nella difesa innata contro i patogeni



I recettori dell'immunità innata per i PAMP batterici sono espressi nel citoplasma e sulla membrana basolaterale ma non su quella luminale

Le cellule dendritiche nella lamina propria esprimono bassi livelli di TLR



Variazioni antigeniche

Meccanismo adottato da virus (HIV, virus dell'influenza, Sars-CoV-2), batteri (streptococco, stafilococco, meningococco, *borrelia burgdorferi*, *bordetella pertussis, neisseria gonorrhea*) e protozoi (tripanosomi)

Effetti:

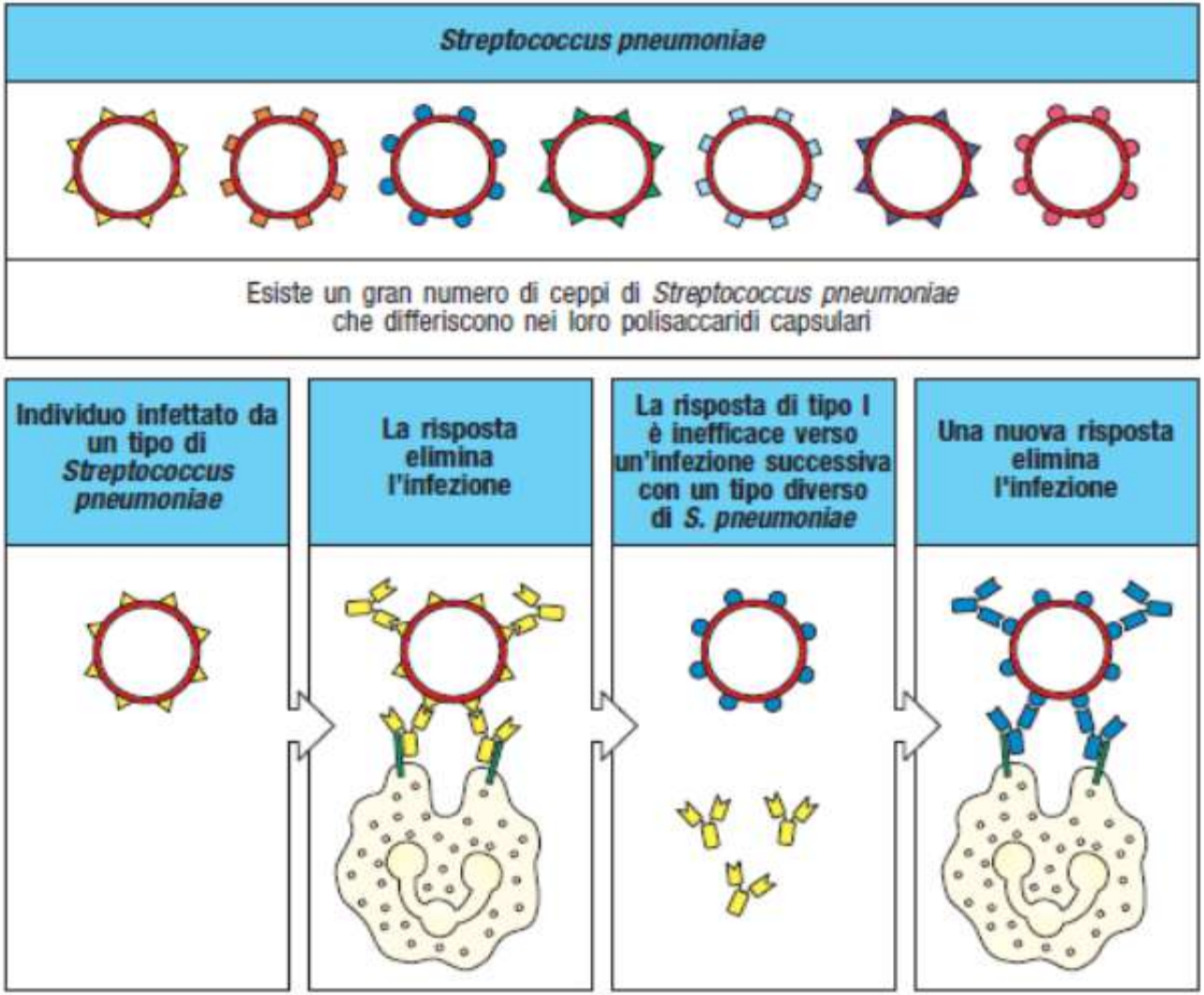
Evasione della risposta umorale per

-mancato riconoscimento da parte degli anticorpi

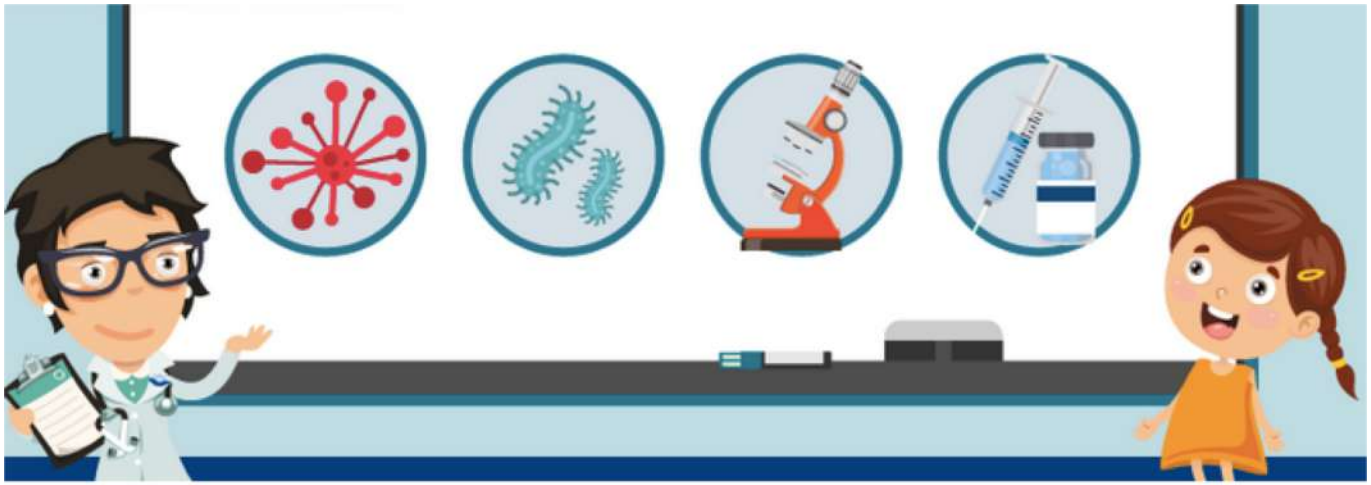
Evasione della risposta cellulo-mediata per

-interferenza con la proteolisi e con la generazione dell'antigene
-mancato legame alle molecole MHC
-mancato riconoscimento da parte del TCR dei linfociti T
-creazione di APL (altered peptide ligand)

Molteplici sierotipi di *Streptococcus pneumoniae*: un caso di variazione antigenica

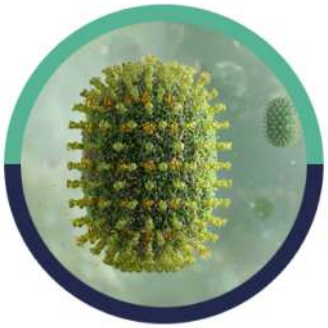


Più di 90 diversi sierotipi di *Streptococcus pneumoniae o pneumococco* agente della polmonite batterica, della bronchite, otite media, setticemia e meningite.



Ceppi influenzali *2024*-2025 del virus dell'influenza di tipo A

- **H1N1**
- **H3N2** **(portatore della influenza australiana)**

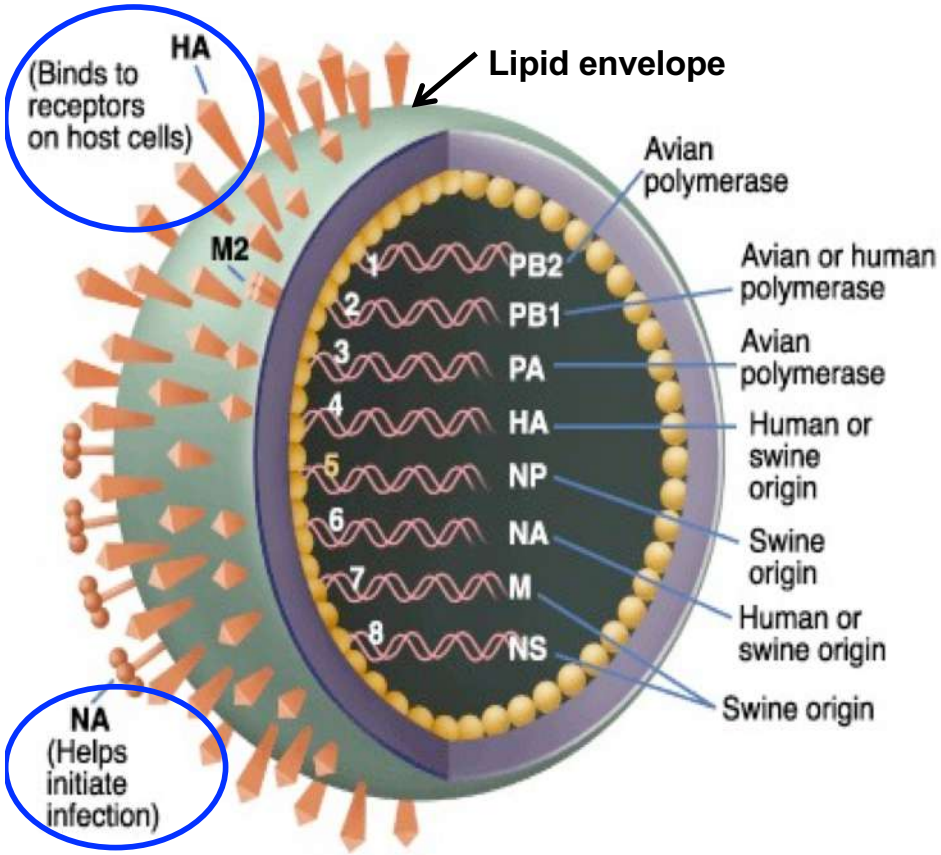


Variazioni antigeniche del virus dell'influenza di tipo A

Le glicoproteine coinvolte sono:

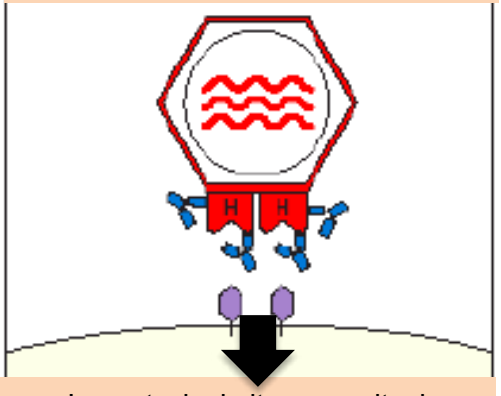
l'emoagglutinina

la neuroaminidasi (sialidasi)

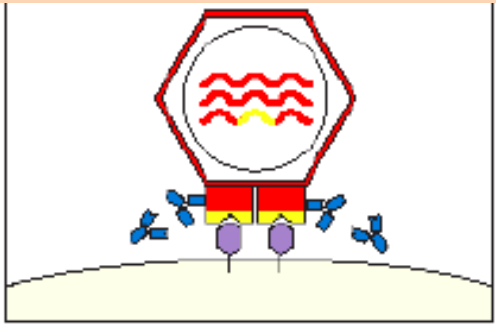


Antigenic drift
(mutazioni puntiformi)

Anticorpi neutralizzanti contro la emoagglutinina bloccano il legame con la cellula

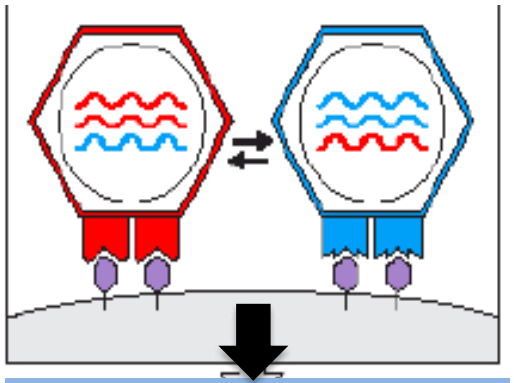


Le mutazioni alterano epitopi dell'emoagglutinina che non sono più riconosciuti dagli anticorpi neutralizzanti

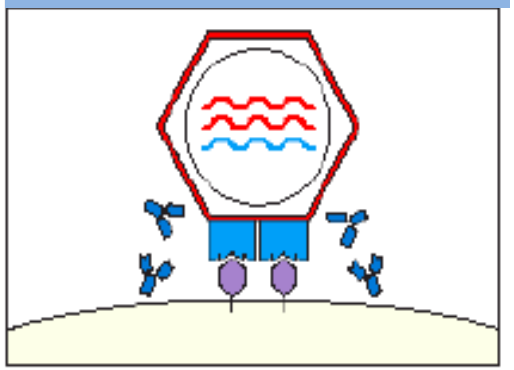


Antigenic shift
(riarrangiamento)

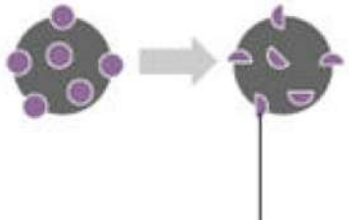
La variazione antigenica si ha quando sono scambiati segmenti di RNA tra ceppi virali in un ospite intermedio



Nessuna immunità crociata protettiva verso il virus che esprime una nuova emoagglutinina

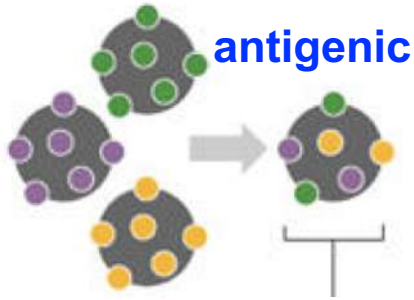


antigenic drift



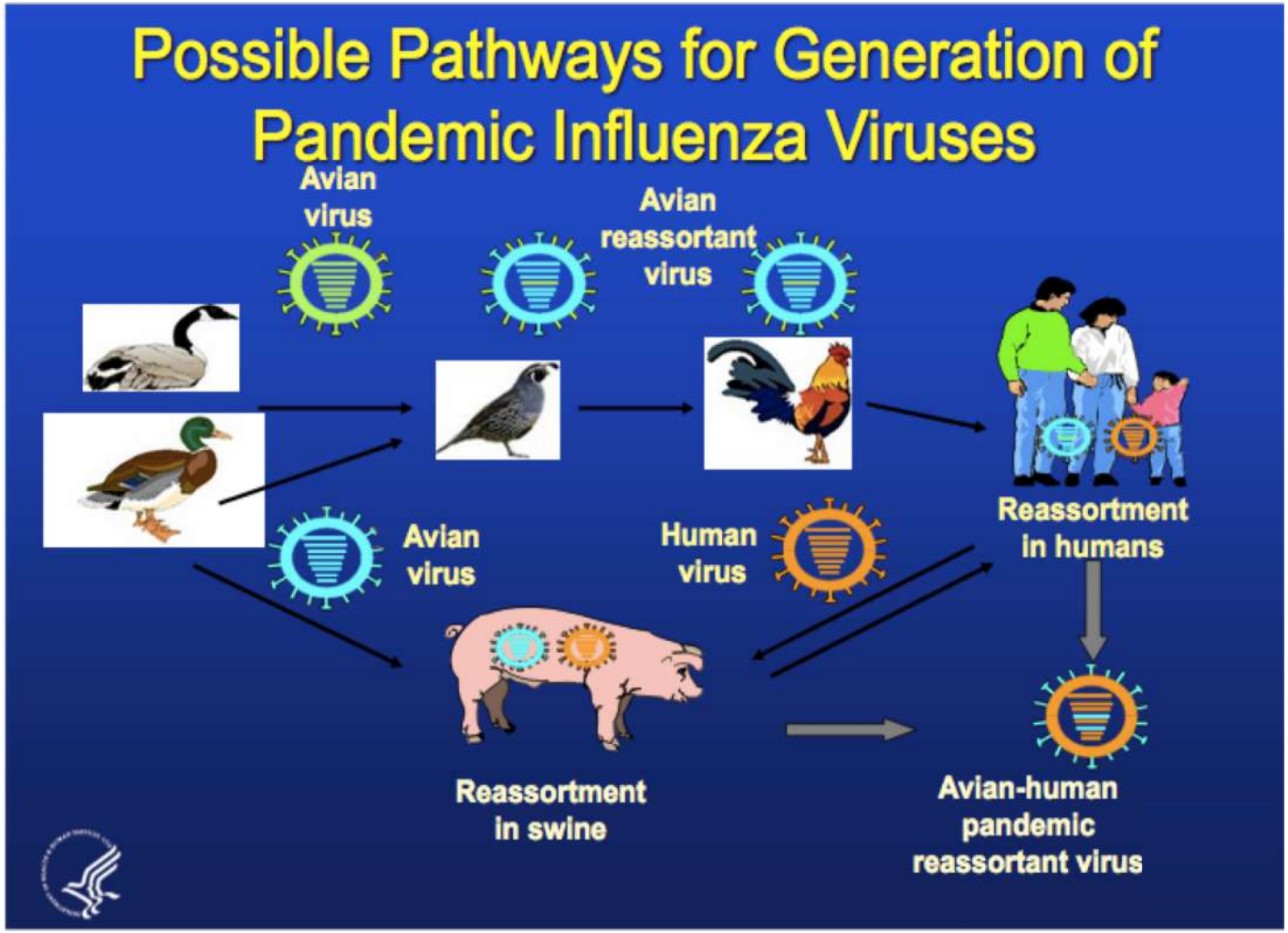
small mutations

antigenic shift



new strain

Generazione di nuovi ceppi di virus influenzali causata da ricombinazione genetica



Antigenic shift

Pandemie del:
1918 (spagnola)
1957 (asiatica)
1968 (Hong Kong influenza)
2009 (febbre suina ceppo H1N1)