

**CODICE OPIS
PATOLOGIA MOLECOLARE E
IMMUNOPATOLOGIA (1041600)**

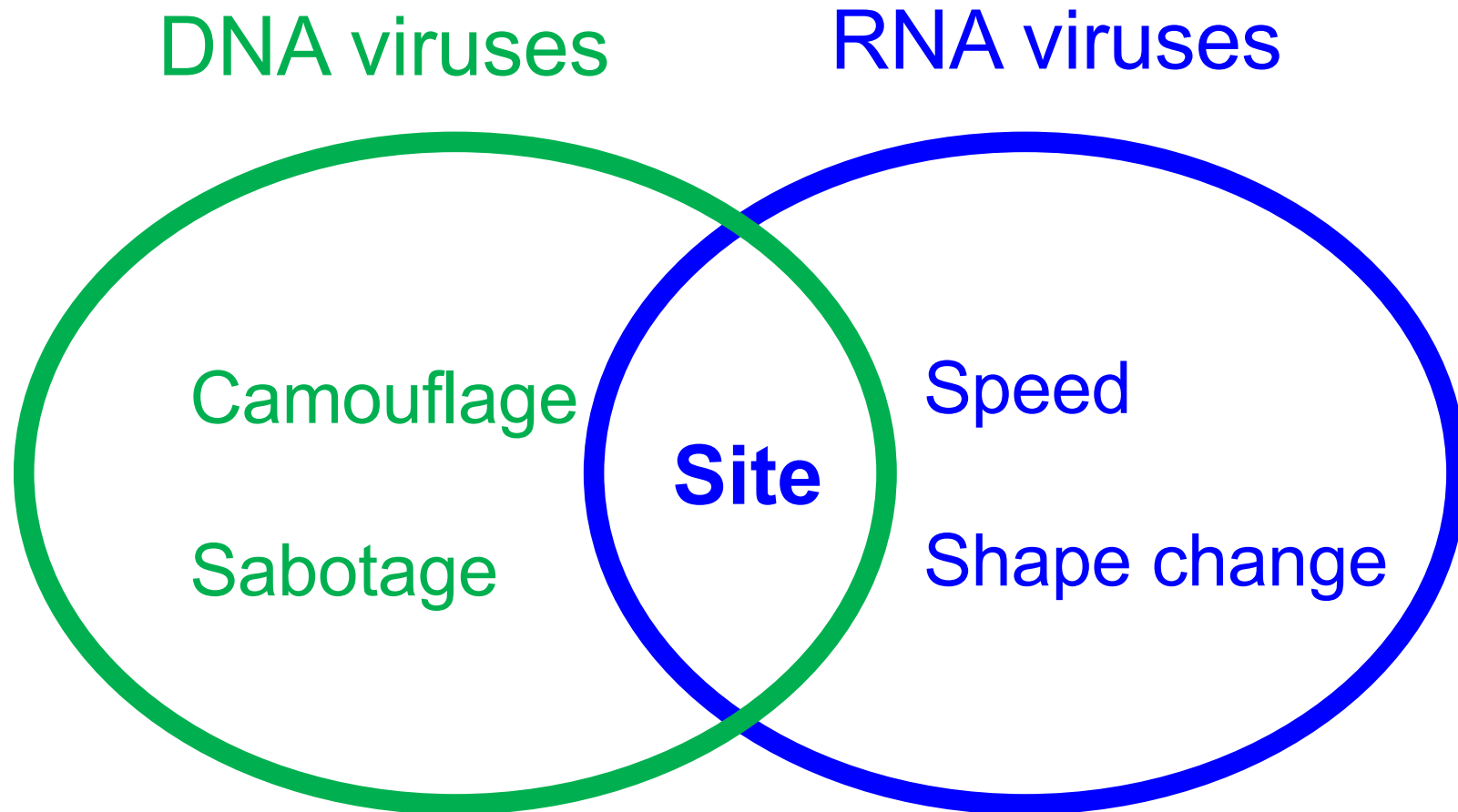
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**PATOLOGIA MOLECOLARE
(1041600_2)**

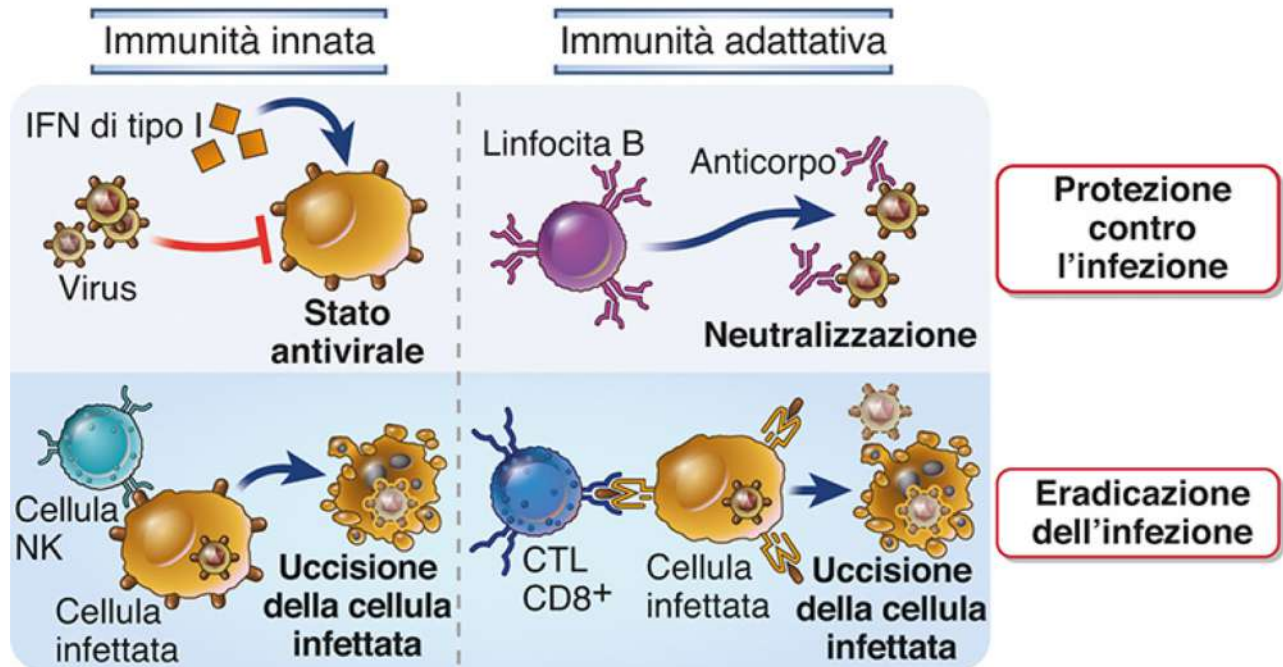
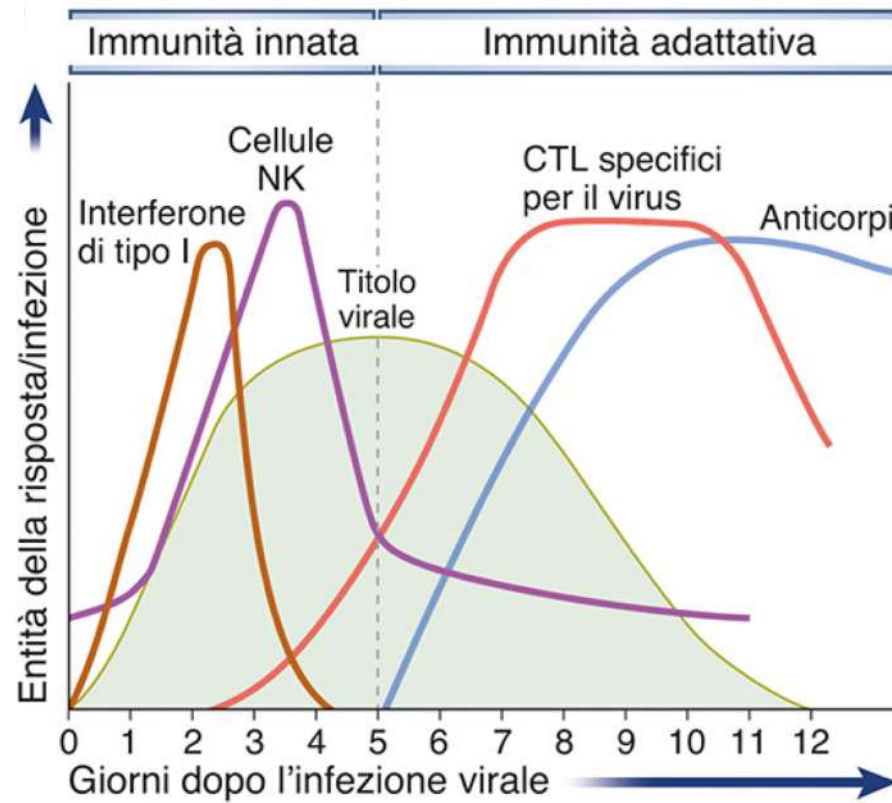


Risposte immunitarie antivirali e meccanismi virali di evasione

A simple model for understanding viral persistence



Risposte immunitarie innate ed adattative ai virus



Strategie virali per eludere l'immunità innata ed acquisita

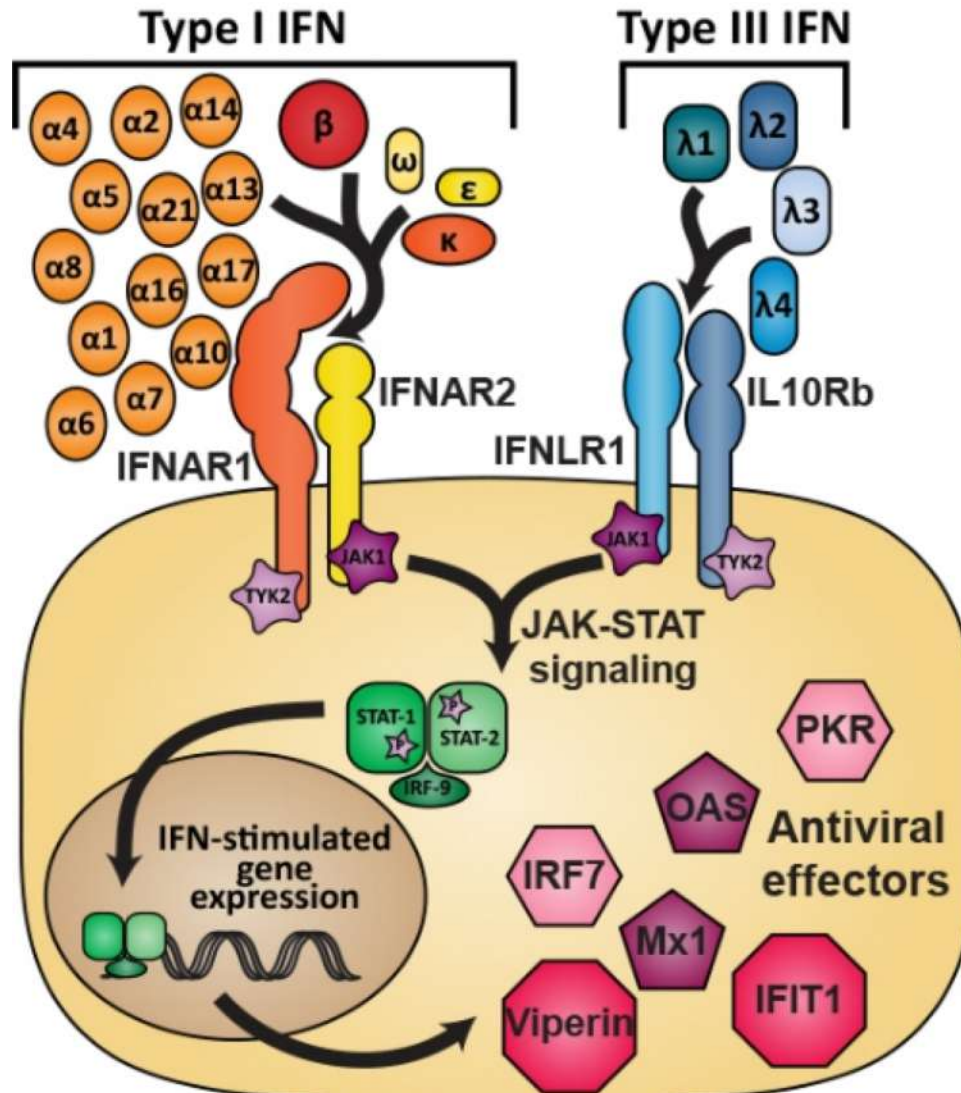
- ❖ **Interferenza con la produzione degli IFN di tipo I e le relative vie di trasduzione del segnale**
- ❖ **Interferenza con la presentazione dell'antigene mediata dalle molecole MHC di classe I e II; con l'espressione dei corecettori CD4 e CD8 e di altre proteine di superficie mediante:**
 - interferenza con la biosintesi delle molecole MHC
 - interferenza con la trascrizione
 - interferenza con la sintesi proteica
 - accelerazione della degradazione di molecole chiave per la presentazione
 - ritenzione intracellulare
 - aumento dell'endocitosi di molecole MHC e non-MHC dalla superficie cellulare
- ❖ **Inibizione dell'attivazione delle cellule NK**
- ❖ **Interferenza con i processi apoptotici e autofagici**
- ❖ **Interferenza con l'azione di citochine e chemochine e dei loro recettori**
- ❖ **Inibizione delle risposte umorali agendo sul sistema del complemento e sui recettori Fc**

Interferoni di tipo I

Mediatori precoci della risposta antivirale dell'immunità innata

IFN α : 13 geni (cromosoma 9) che codificano per le diverse isoforme, sono espressi principalmente da cellule dendritiche plasmacitoidi e fagociti mononucleati

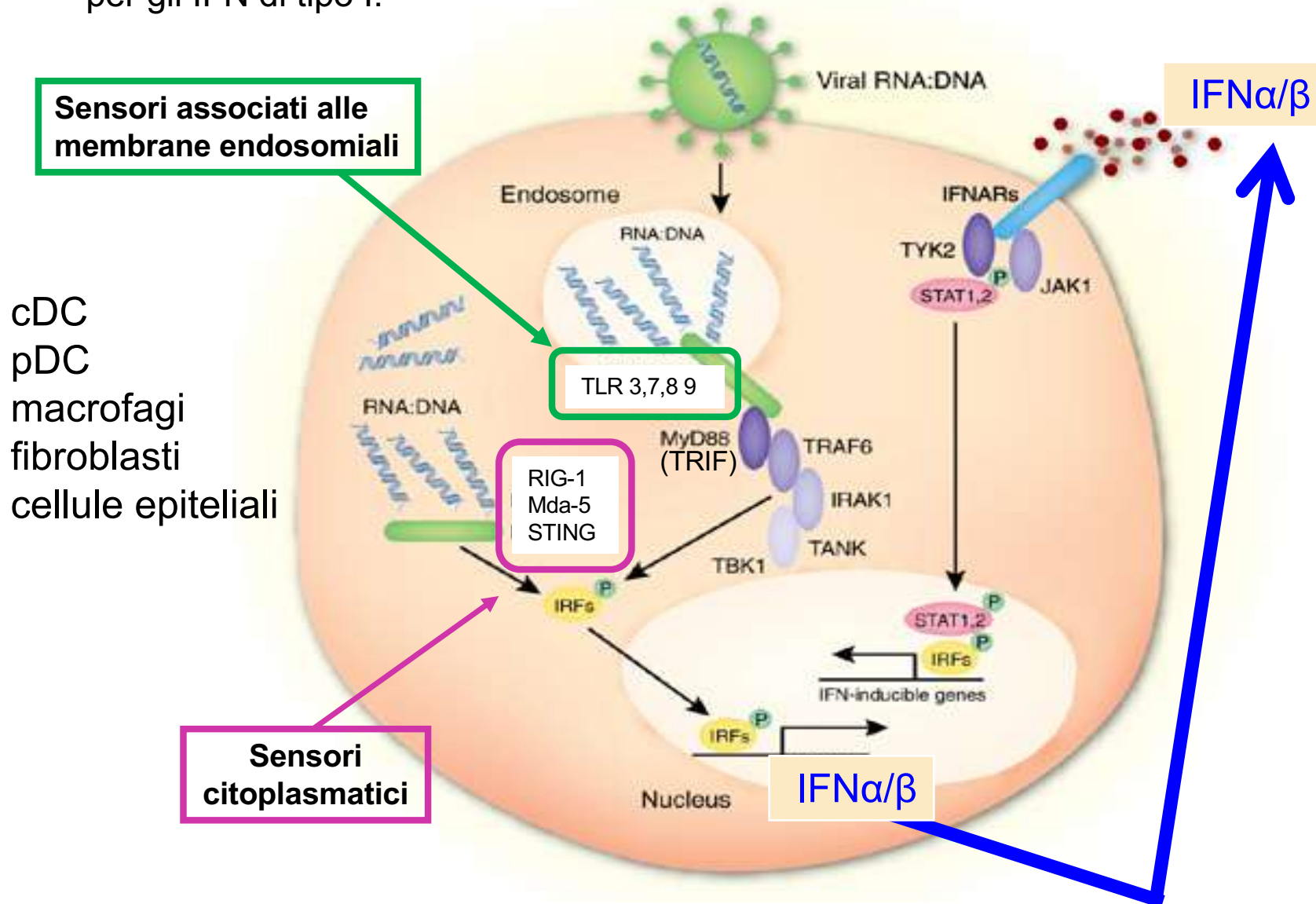
IFN β : una sola proteina prodotta da vari tipi cellulari (tra cui i fibroblasti)



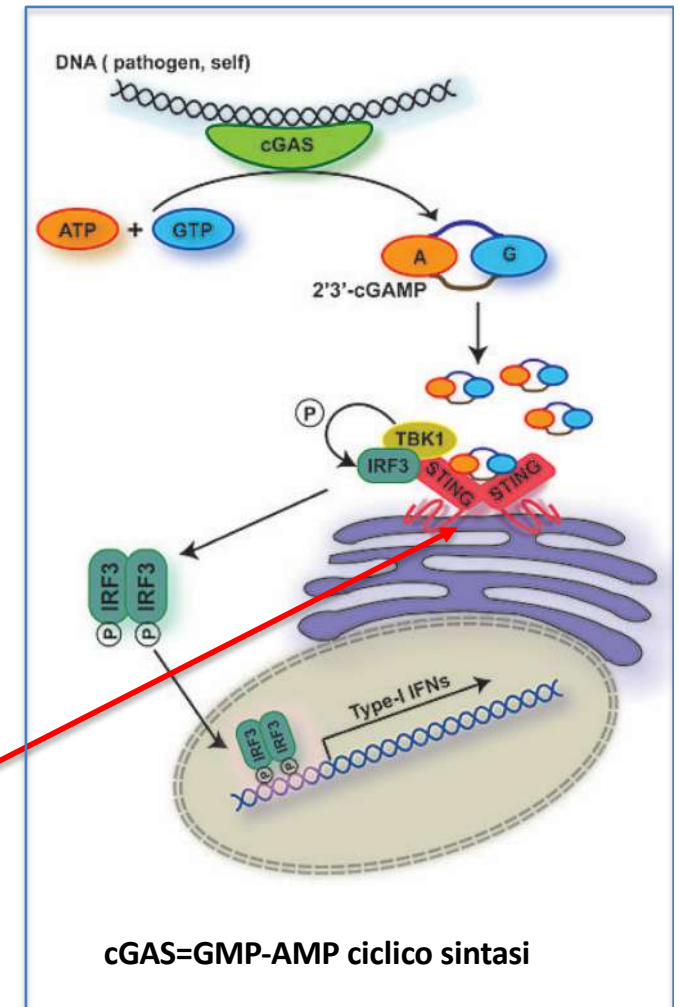
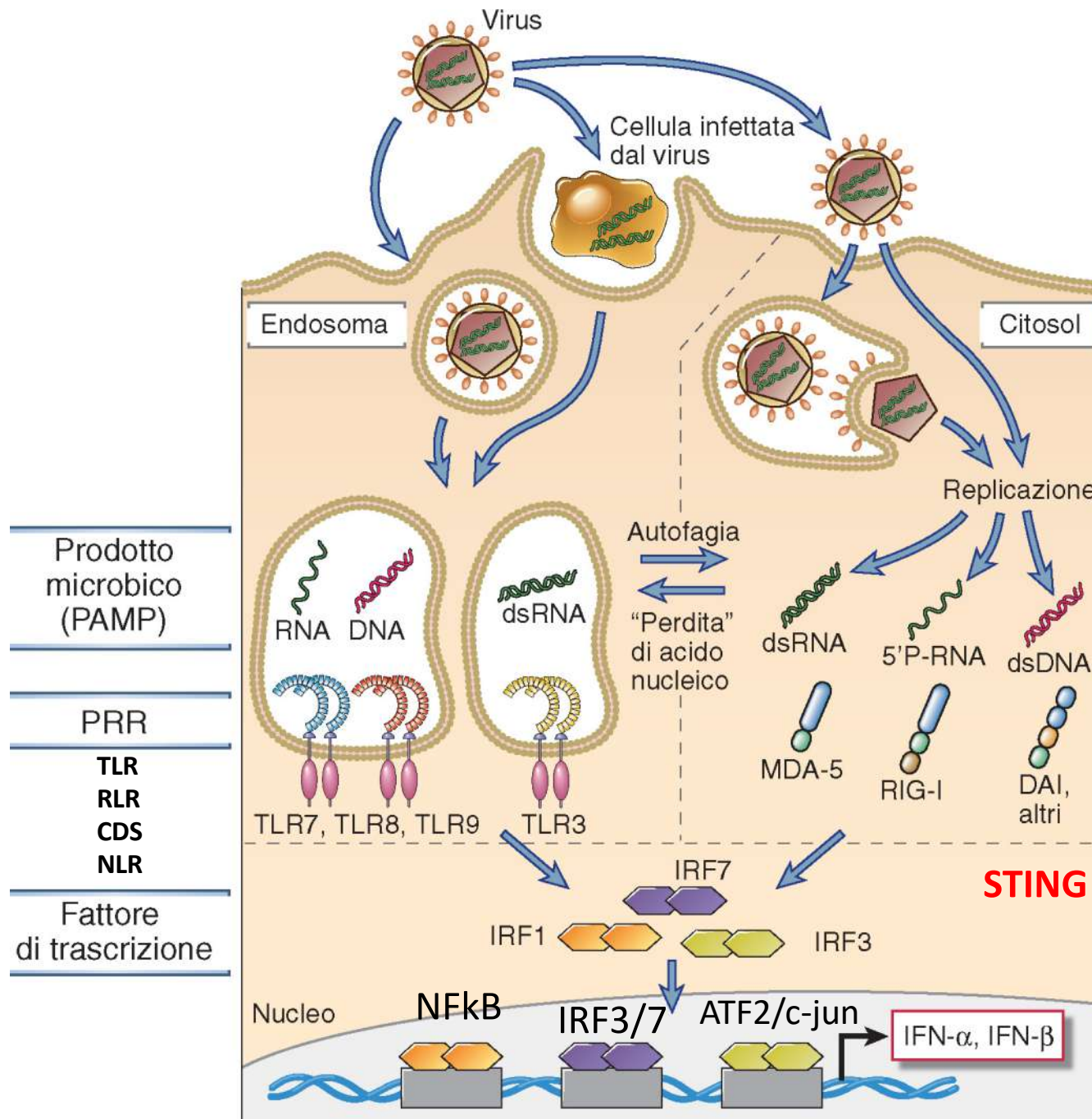
IFN α e IFN β
condividono gli
stessi recettori
IFNAR1 e IFNAR2

Produzione di IFN di tipo I

Gli acidi nucleici virali si legano ai PRR (TLR; RLR; NLR e CDS), recettori cellulari (“sensori”) associati a vie di trasduzione del segnale che attivano la famiglia di fattori di trascrizione IRF (Interferon Regulatory Factor) ed altri fattori di trascrizione inducendo la trascrizione dei geni per gli IFN di tipo I.



Induzione degli interferoni di tipo I da parte dei virus

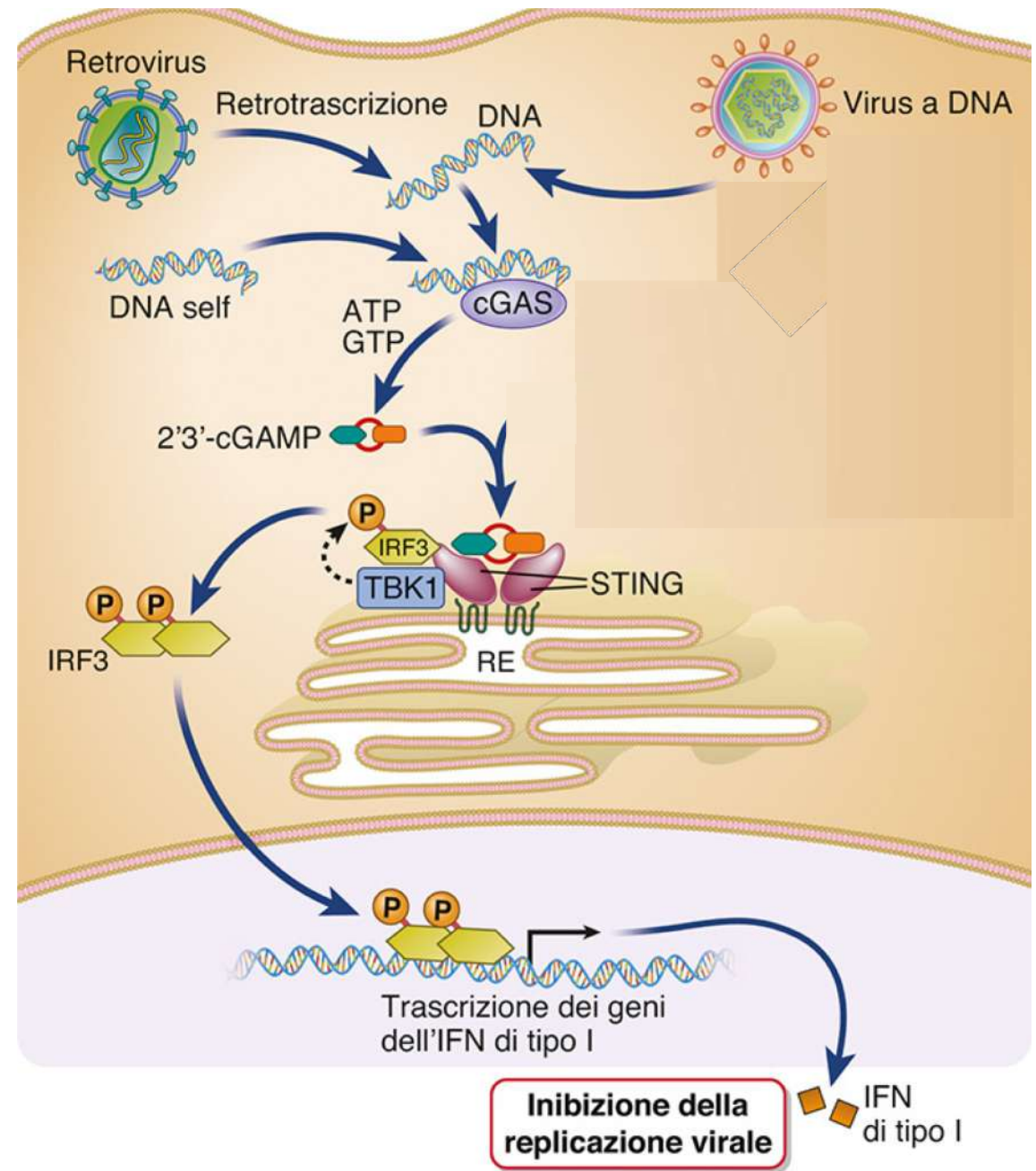


Via di trasduzione del segnale di STING per la produzione degli interferoni di tipo I

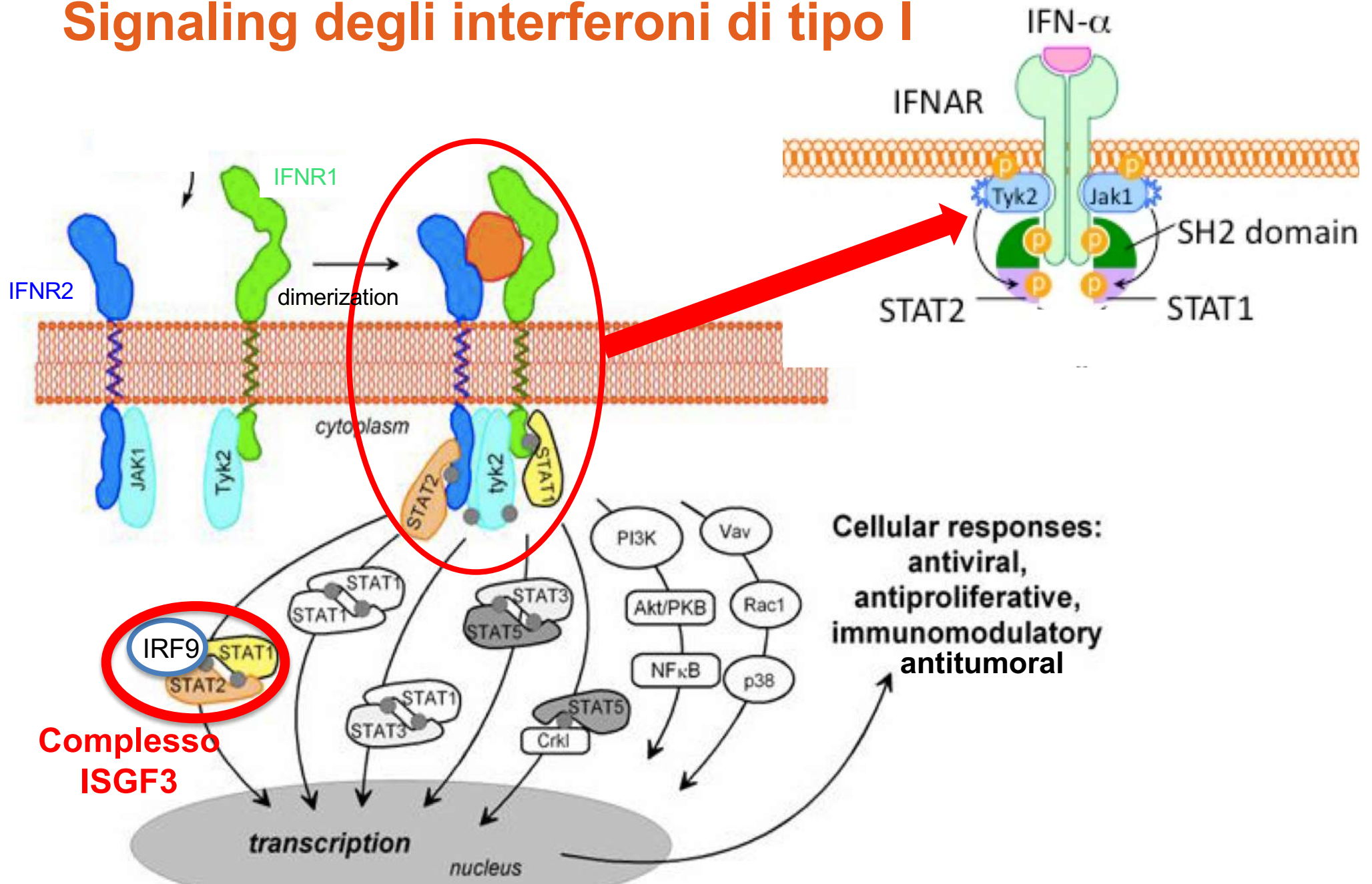
Il DNA virale nel citosol attiva l'enzima **cGAS** (cyclic GMP-AMP synthase) che catalizza la sintesi di GMP-AMP ciclico (**cGAMP**) a partire da ATP e GTP.

cGAMP si lega a **STING** (Stimulator of interferon genes), sulla membrana del RE, che a sua volta recluta e attiva la chinasi **TBK1** che fosforila **IRF3**.

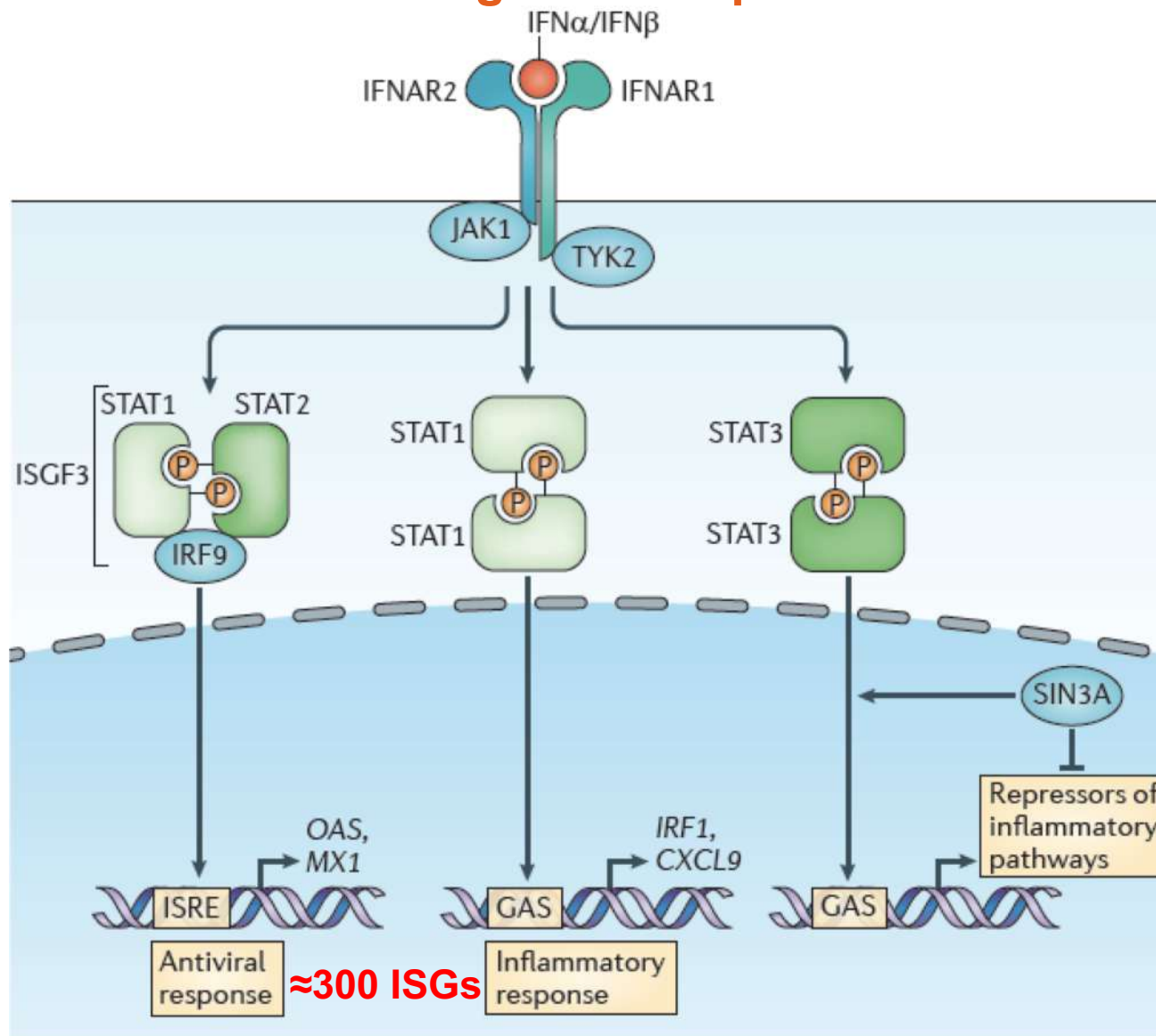
IRF3 fosforilato migra nel nucleo e induce l'espressione dei geni per gli IFN di tipo I.



Signaling degli interferoni di tipo I



Pathway classico di trasduzione del segnale degli IFN di tipo I



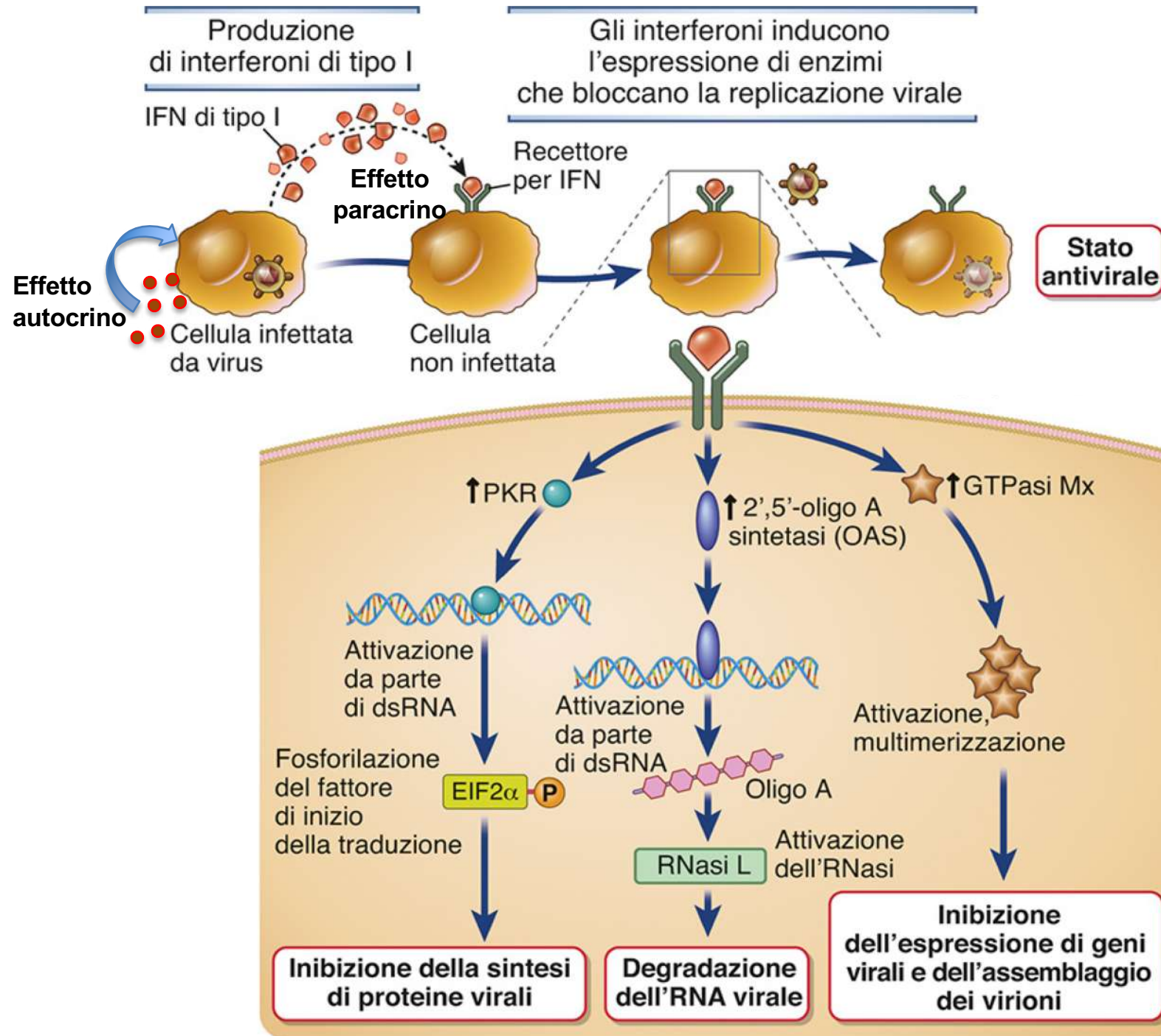
ISGs = IFN-stimulated genes

ISRE= IFN-stimulated response elements

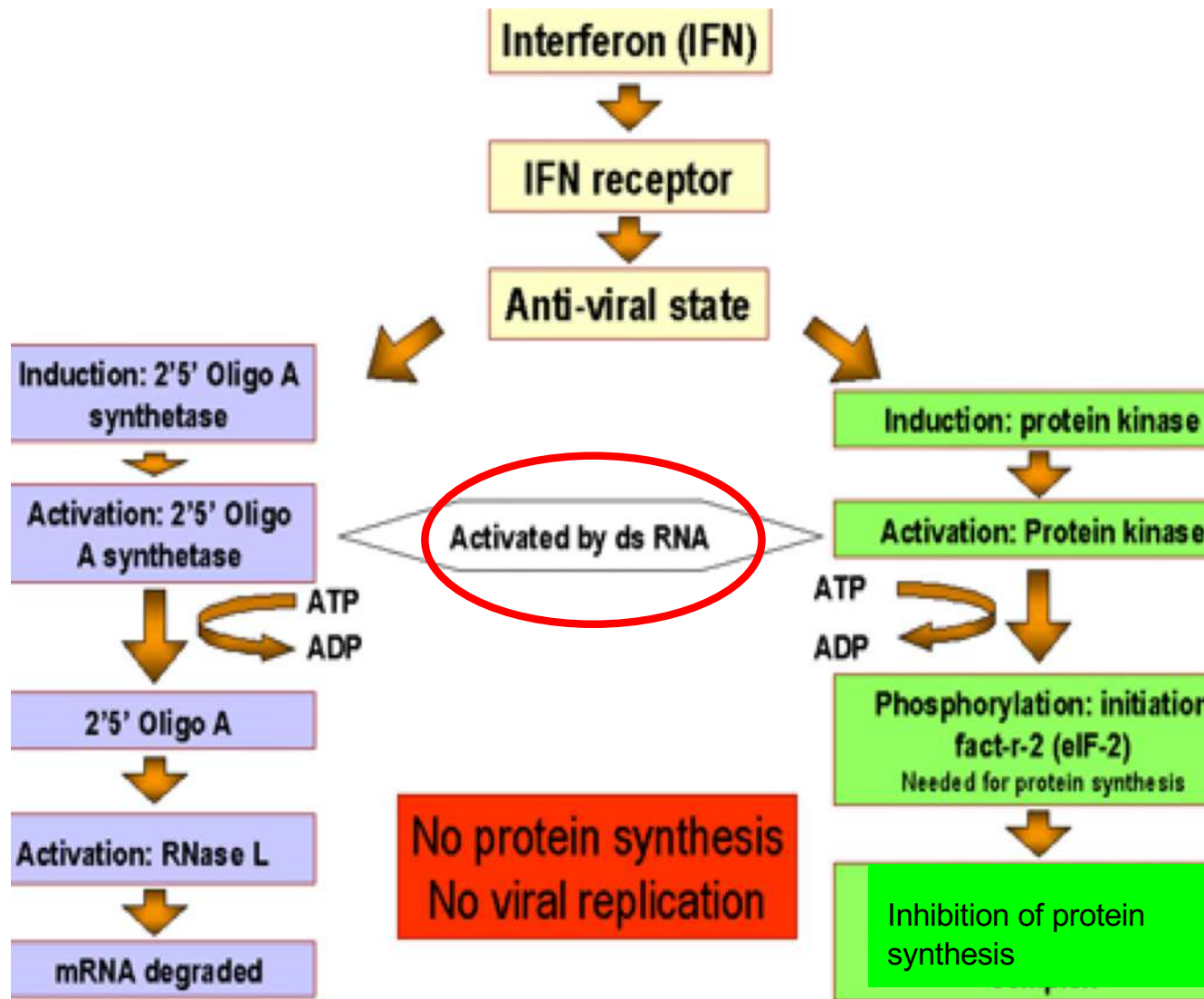
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On engagement, the interferon- α receptor (IFNAR, which is composed of the IFNAR1 and IFNAR2 subunits) activates Janus kinase 1 (JAK1) and tyrosine kinase 2 (TYK2). Phosphorylation of the receptor by these kinases results in the recruitment of signal transducer and activator of transcription (STAT) proteins, phosphorylation, dimerization and nuclear translocation. The three predominant STAT complexes that are formed in response to type I interferon (IFN) control distinct gene-expression programmes. The interferon-stimulated gene factor 3 (ISGF3) complex (which is composed of STAT1, STAT2 and IFN-regulatory factor 9 (IRF9)) binds to IFN-stimulated response element (ISRE) sequences to activate classical antiviral genes, whereas STAT1 homodimers bind to gamma-activated sequences (GASs) to induce pro-inflammatory genes. STAT3 homodimers indirectly suppress pro-inflammatory gene expression, probably by the induction of as-yet-unknown transcriptional repressors. Type I IFN-activated STAT3 is bound by the co-repressor complex SIN3A transcription regulator homologue A (SIN3A), which suppresses induction of direct STAT3 target genes by promoting de-acetylation of STAT3 and histones. CXCL9, CXC-chemokine ligand 9; MX1, IFN-induced GTP-binding protein Mx1; OAS, 2'-5'-oligoadenylate synthase; P, phosphate.

Effetti biologici degli interferoni di tipo I



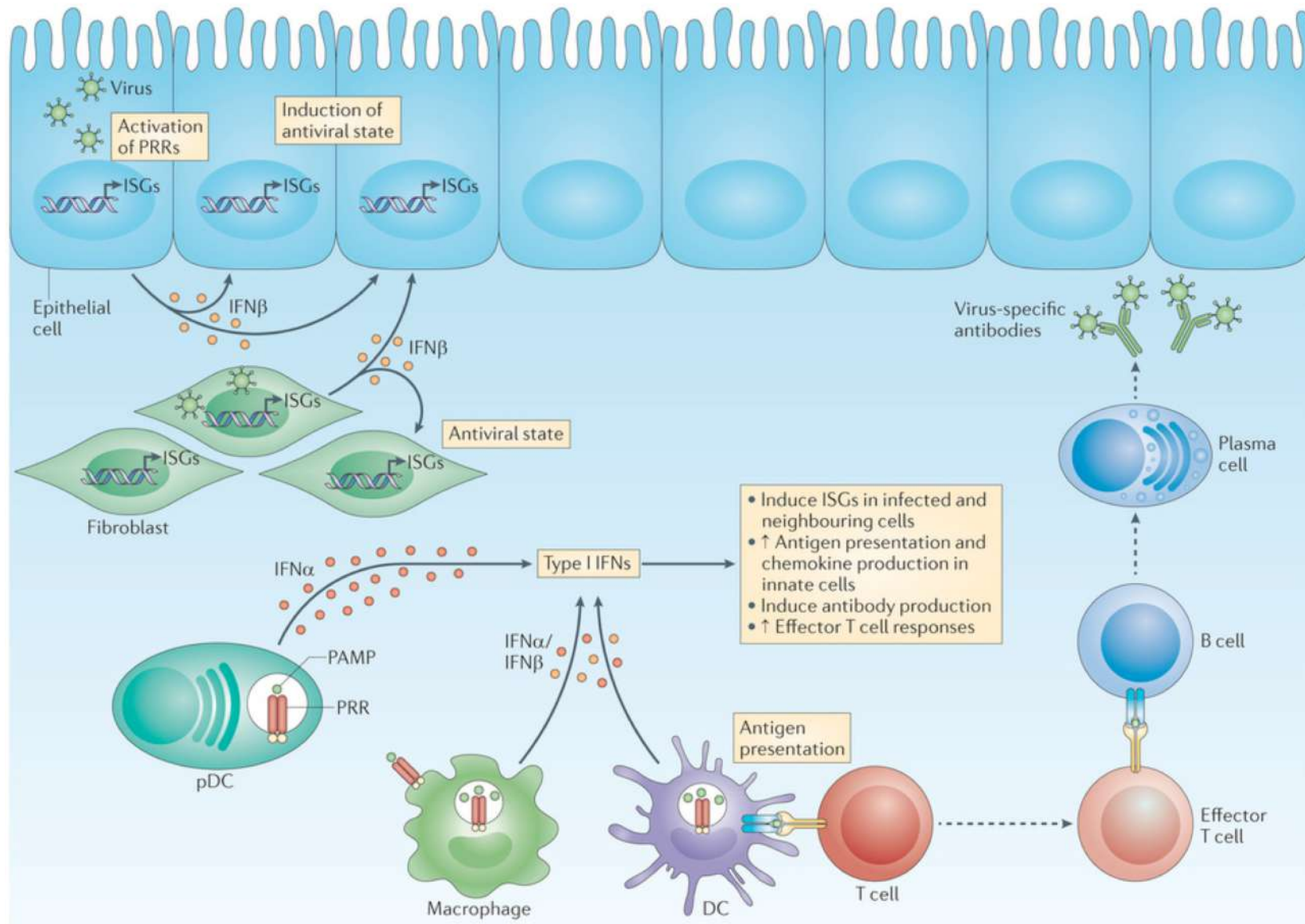
Induzione dello stato antivirale



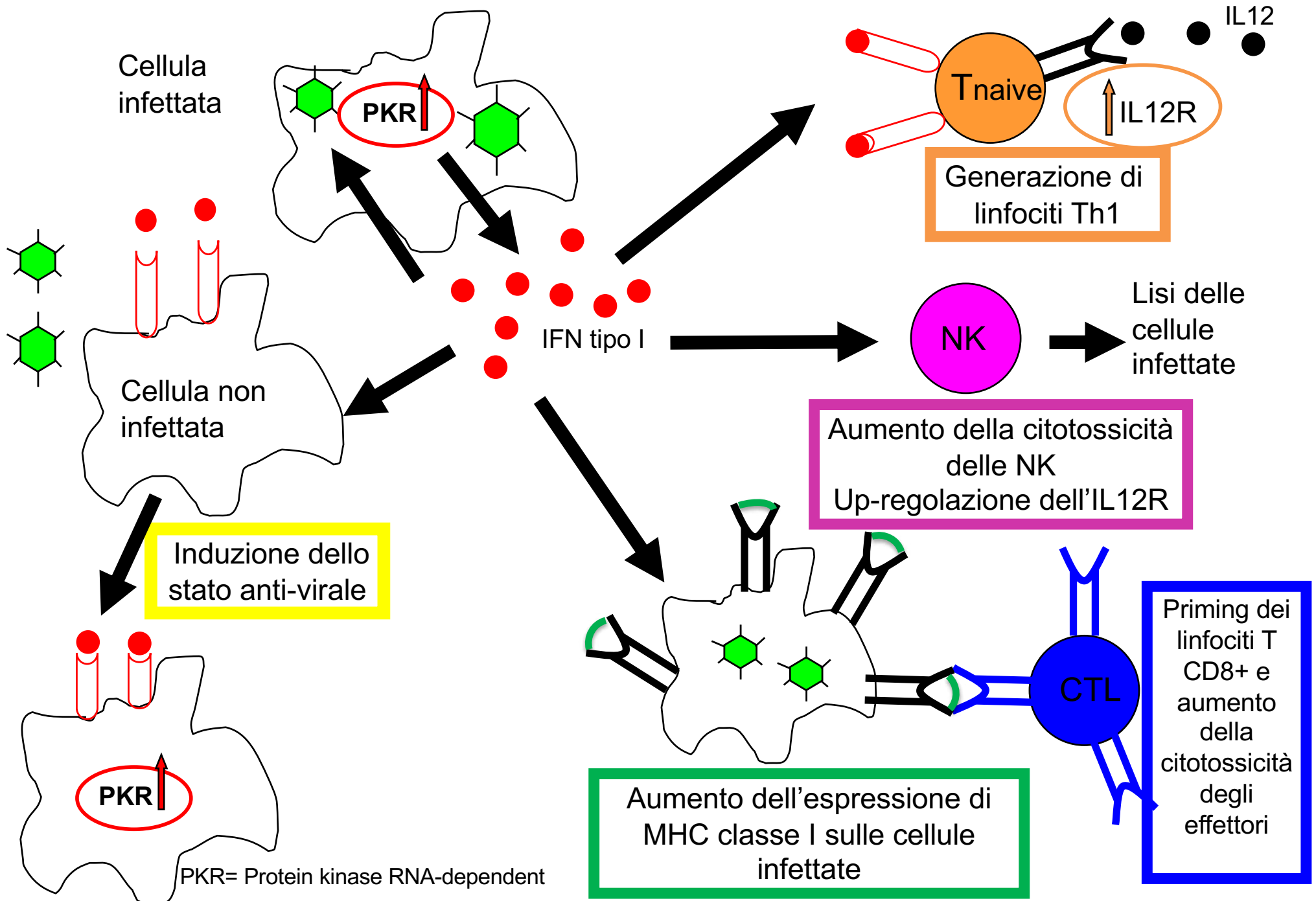
Induzione di enzimi che bloccano la replicazione virale:

- -2'-5'-oligoadenilato sintetasi che attiva una endo-ribonucleasi (RNasi L) che degrada mRNA virali e cellulari
- - protein kinase R (PKR) che fosforila e blocca il fattore eIF-2 α (fattore di inizio della sintesi proteica)

Type I interferon controls innate and adaptive immunity and intracellular antimicrobial programs



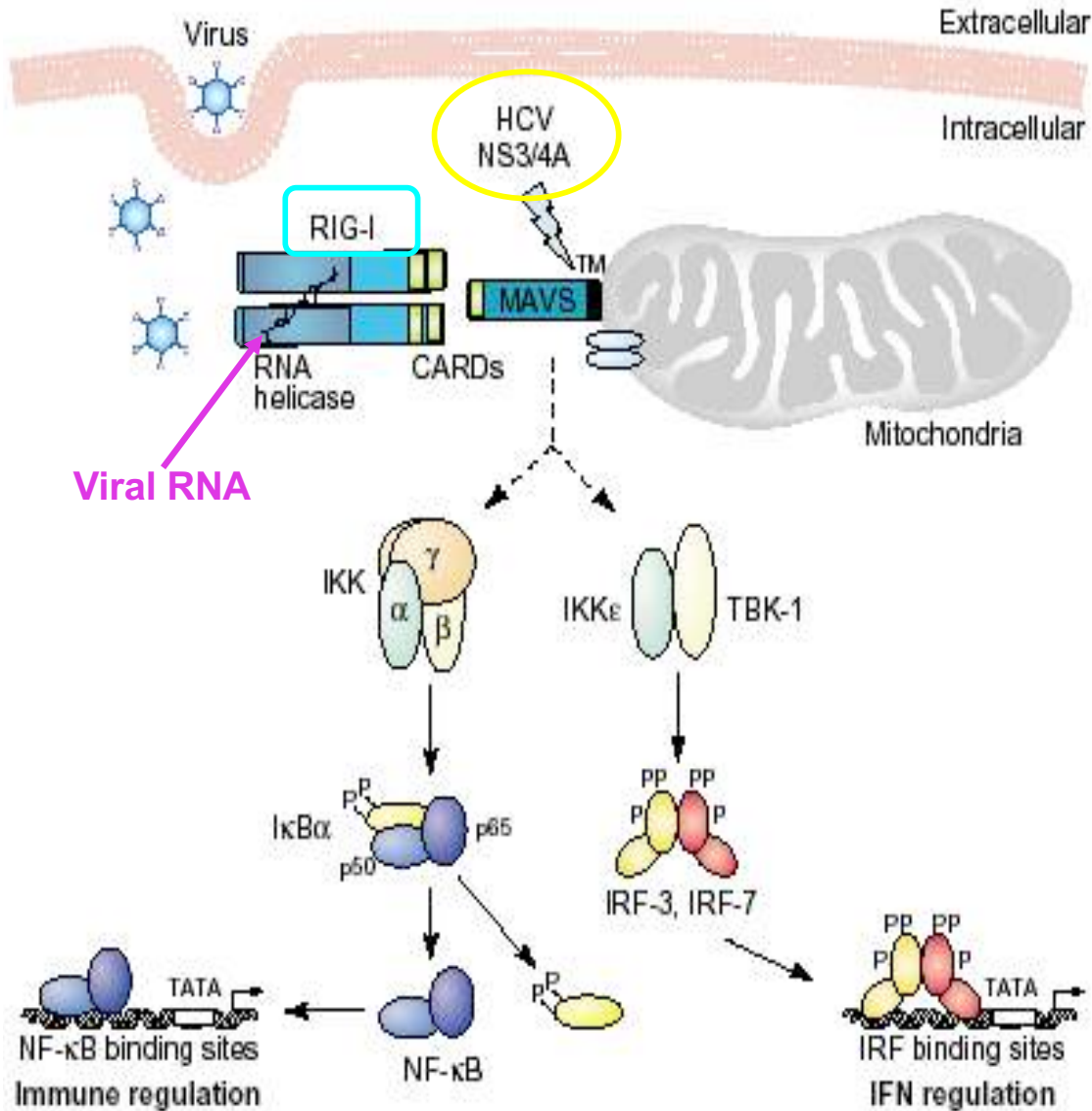
Effetti biologici degli IFN di tipo I



Gli IFN di tipo I svolgono tre funzioni principali:

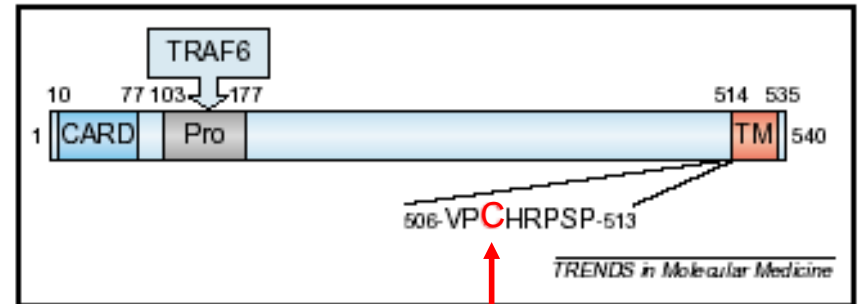
- ❖ Attivano lo stato antivirale nelle cellule infettate e in quelle limitrofe che limita la diffusione dell'infezione.
- ❖ Modulano le risposte immunitarie innate, compresa la presentazione dell'antigene e le funzioni delle cellule natural killer, limitando i pathways pro-infiammatori.
- ❖ Attivano il sistema immunitario adattativo per lo sviluppo di risposte linfocitarie T e B antigene-specifiche ad alta affinità

Interferenza con la produzione degli IFN di tipo I da parte di HCV



NS3/4A, proteasi di HCV, taglia **MAVS** (detta anche IPS-1 o VISA o Cardif) dopo la cisteina 508 ed impedisce l'attivazione del pathway di RIG-I che porta alla produzione degli Interferoni di tipo I.

Domini Strutturali di **MAVS**



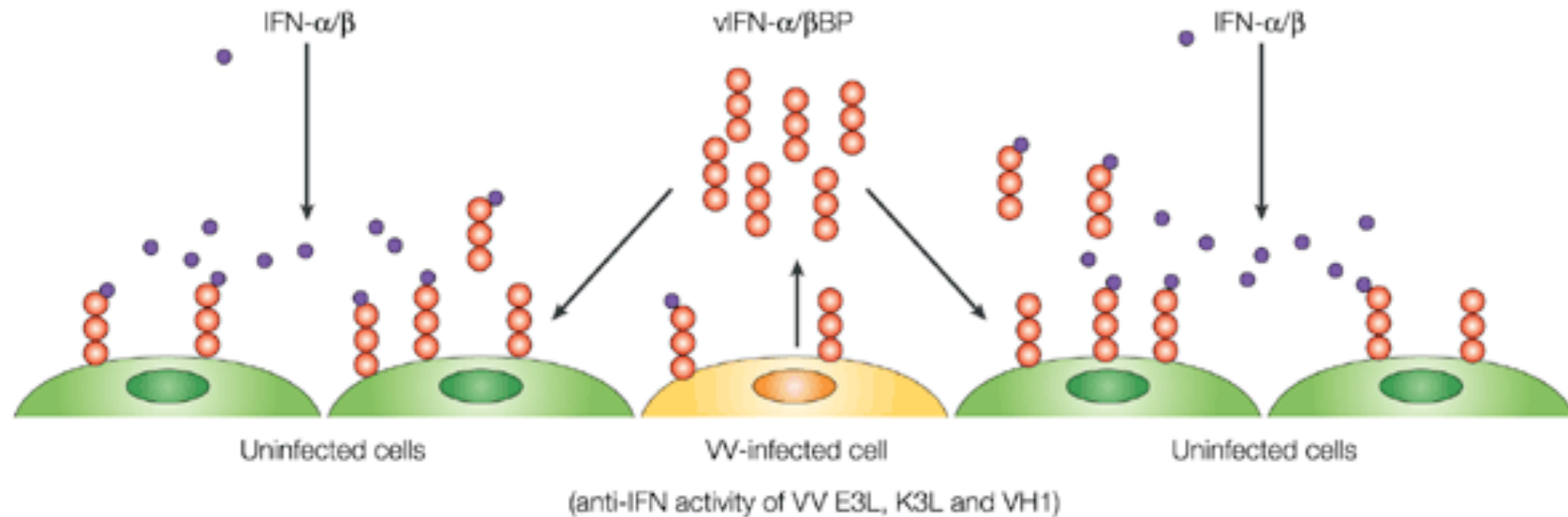
The MAVS/IPS-1/VISA/Cardif molecule (540 aa) contains a CARD domain, a proline-rich (Pro) region, which interacts with TRAF6, and a C-terminal mitochondrial transmembrane domain (TM). The region adjacent to the TM contains Cys508 (in red), which is the target residue for the HCV NS3/4 protease.

MAVS=mitochondrial antiviral signaling molecule
CARD=caspase activation and recruiting domain

The RIG-I/MAVS signaling pathway. RIG-I contains two N-terminal CARD domains and a C-terminal RNA helicase. Helicase interacts with incoming viral RNA. MAVS/IPS-1/VISA/Cardif is a link between RIG-I and downstream kinases. Upon sensing viral RNA the CARD domains of RIG-I and MAVS/IPS-1/VISA/Cardif interact leading to the activation of TBK-1 and IKKε kinases and the phosphorylation of IRF-3 and IRF-7 transcription factors. MAVS/IPS-1/VISA/Cardif can also lead to NF-κB activation via the IKKα/β/γ complex, which phosphorylates the inhibitory subunit IκBα, resulting in the release of NF-κB DNA-binding subunits. MAVS/IPS-1/VISA/Cardif contains a mitochondrial transmembrane domain (TM) that localizes it to mitochondria.

Come i poxvirus inibiscono l'attività degli interferoni di tipo I

Produzione di un recettore “decoy” denominato vIFN- α/β BP



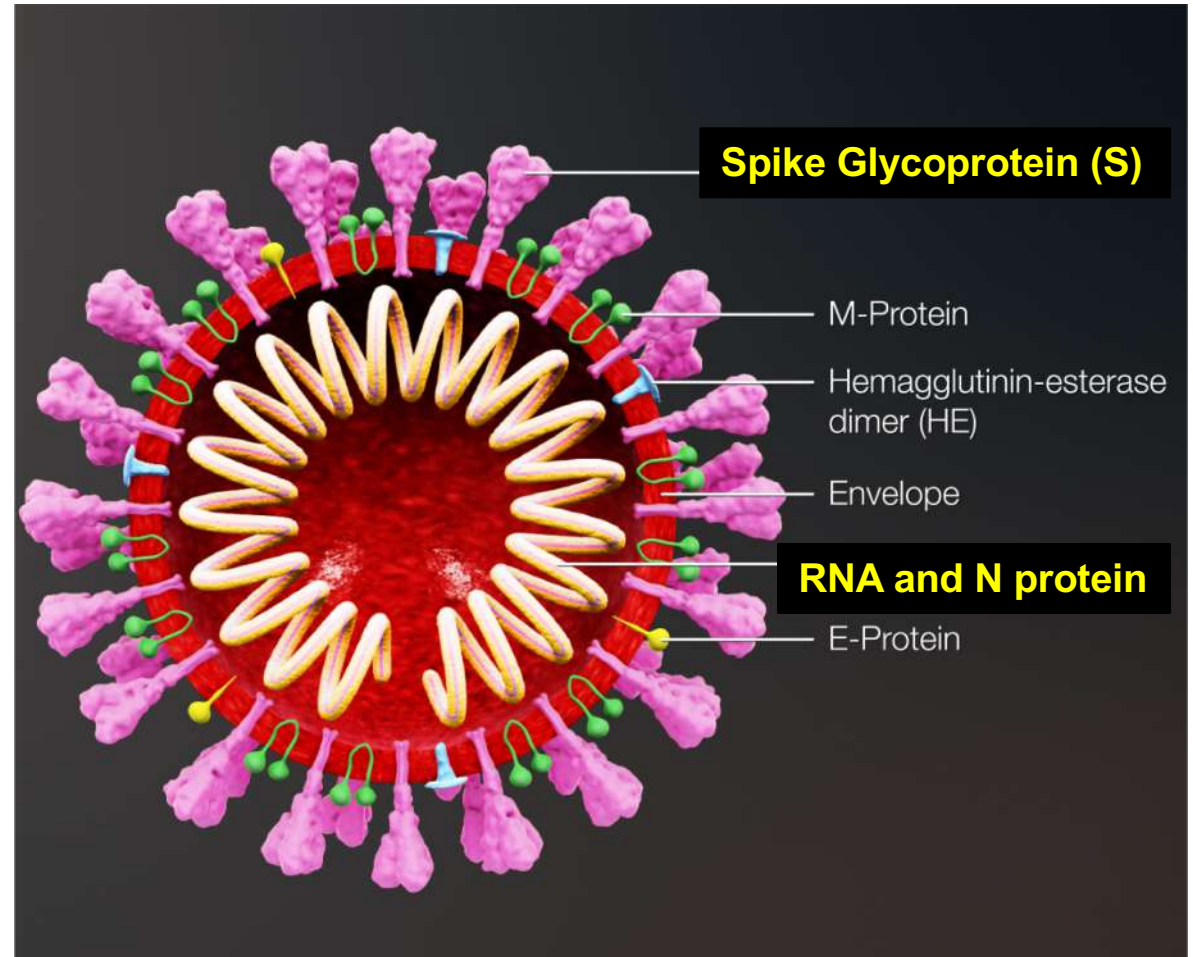
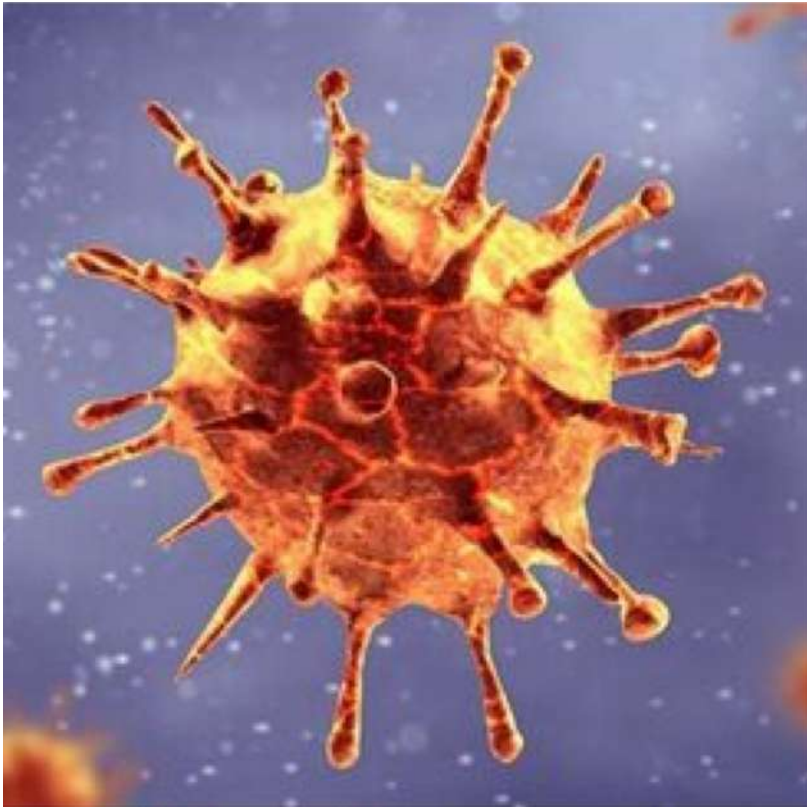
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Inoltre producono:

VV VH1= fosfatasi che blocca il pathway per la sintesi degli interferoni di tipo I

VV E3L e K3L bloccano i pathways indotti da IFN α/β che inducono lo stato antivirale delle cellule

Strategy used by poxvirus vIFN- α/β /BP to block the biological activity of IFNs. Vaccinia virus and other poxviruses encode intracellular proteins that protect the infected cell from the effects of IFNs. The VV VH1 protein is a phosphatase present in the virus particle that prevents the phosphorylation and activation of proteins in the IFN signalling pathway. The VV E3L and K3L proteins block the intracellular pathways that are induced by the actions of IFNs, which lead to the establishment of an anti-viral state that can restrict virus replication. However, the expression of IFNs is induced rapidly in the host after viral infection and these will induce an anti-viral state in uninfected neighbouring cells, which are fully susceptible to the effects of IFNs. As a result of the IFN response, by the time that progeny virions are released from the few cells that are infected initially, virus replication will be highly restricted in neighbouring cells. To counteract this IFN response, the vIFN-/BPs are secreted early during infection and efficiently bind to neighbouring cells. The vIFN-/BP that is present at the cell surface will act as a decoy receptor, preventing the binding of IFNs to cellular receptors and the induction of an efficient anti-viral response. By covering the cells with decoy receptors, vaccinia virus creates an environment in which IFNs cannot induce a protective anti-viral response and prevent virus replication.



SARS-CoV-2

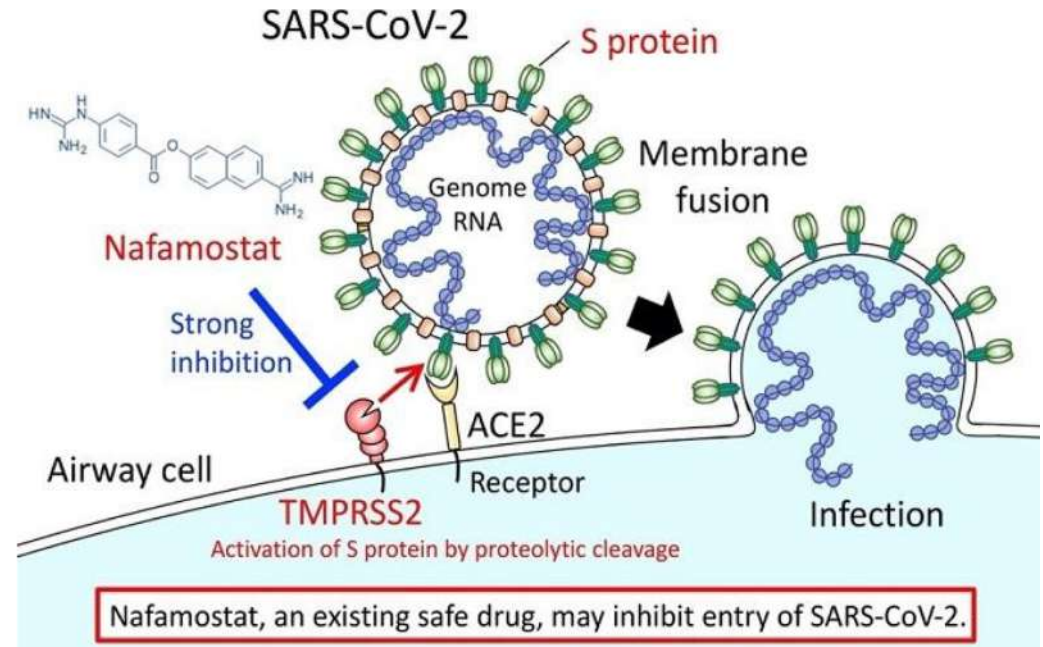
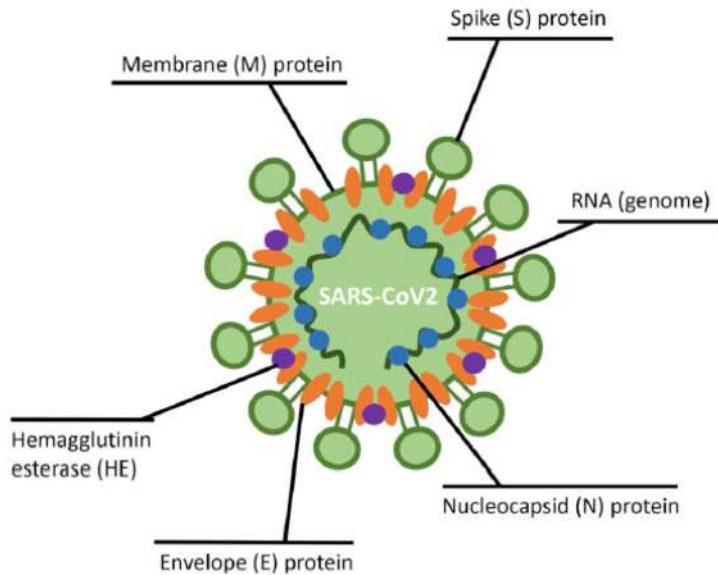
e

strategie di immunoevasione

Severe respiratory disease outbreaks caused by coronaviruses (CoVs)

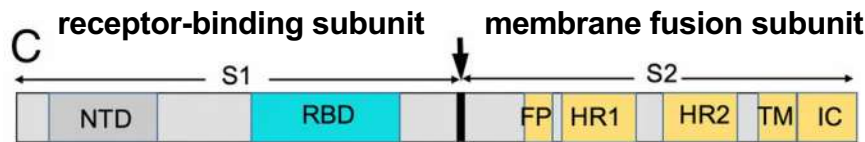
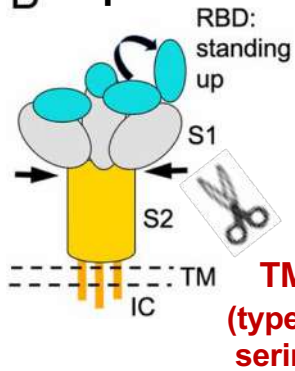
- ❖ The COVID-19 pandemic started in December **2019** in Wuhan, Hubei Province, China caused by Severe acute respiratory syndrome coronavirus 2 (**SARS-CoV-2**)
- ❖ **2012** MERS outbreak in Saudi Arabia caused by Middle East Respiratory Syndrome CoV (**MERS-CoV**) sharing 50% homology to SARS-CoV2
- ❖ **2003-2004** SARS outbreak in China caused by **SARS-CoV-1** which shares 83% homology to the SARS-CoV2

Sars-CoV2 structure and viral cell entry



The spike protein (S) facilitates binding to the transmembrane ACE2 host receptor; the envelope (E) protein together with the membrane (M) protein form the viral envelope and determine its shape; the hemagglutinin esterase (HE) protein may resemble another cell entry mechanism of novel CoVs; the nucleocapsid (N) protein is bound to the RNA genome of the virus to form the nucleocapsid.

B Spike

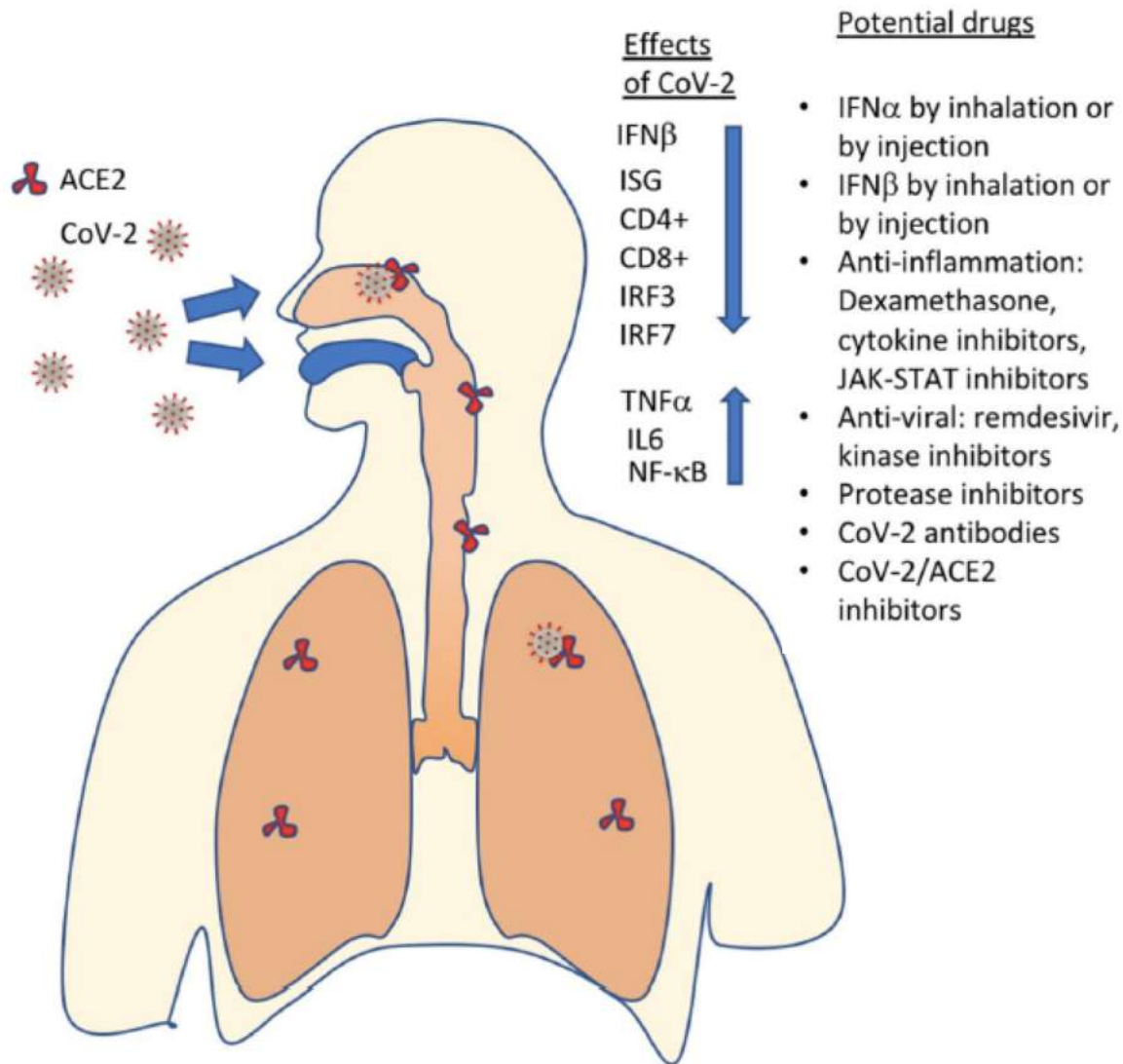


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SARS-CoV-2:	QTQ TNSP RRAR SVA	685	(PPC site)
SARS-CoV:	HTV SLL ---- RSTS	667	
Rs3367-bat:	HTV SLL ---- RSTS	668	
RaTG13-bat:	QTQ TNS ---- RSVA	681	

(B) Schematic drawing of the three-dimensional structure of coronavirus spike. S1, receptor-binding subunit; S2, membrane fusion subunit; TM, transmembrane anchor; IC, intracellular tail. (C) Schematic drawing of the 1D structure of coronavirus spike. NTD, N-terminal domain. **FP (fusion peptide)**, HR1 (heptad repeat 1), and HR2 (heptad repeat 2) are structural units in coronavirus S2 that function in membrane fusion. (D) Sequence comparison of the spike proteins from SARS-CoV-2, SARS-CoV, and two bat SARS-like coronaviruses in a region at the S1/S2 boundary.

Effetti di SARS-CoV-2 sul sistema immunitario e potenziali trattamenti




L'inibizione della produzione di IFN β è mediata da Orf6, nsp1, nsp12, nsp14, nsp15 e proteina M di CoV-2.

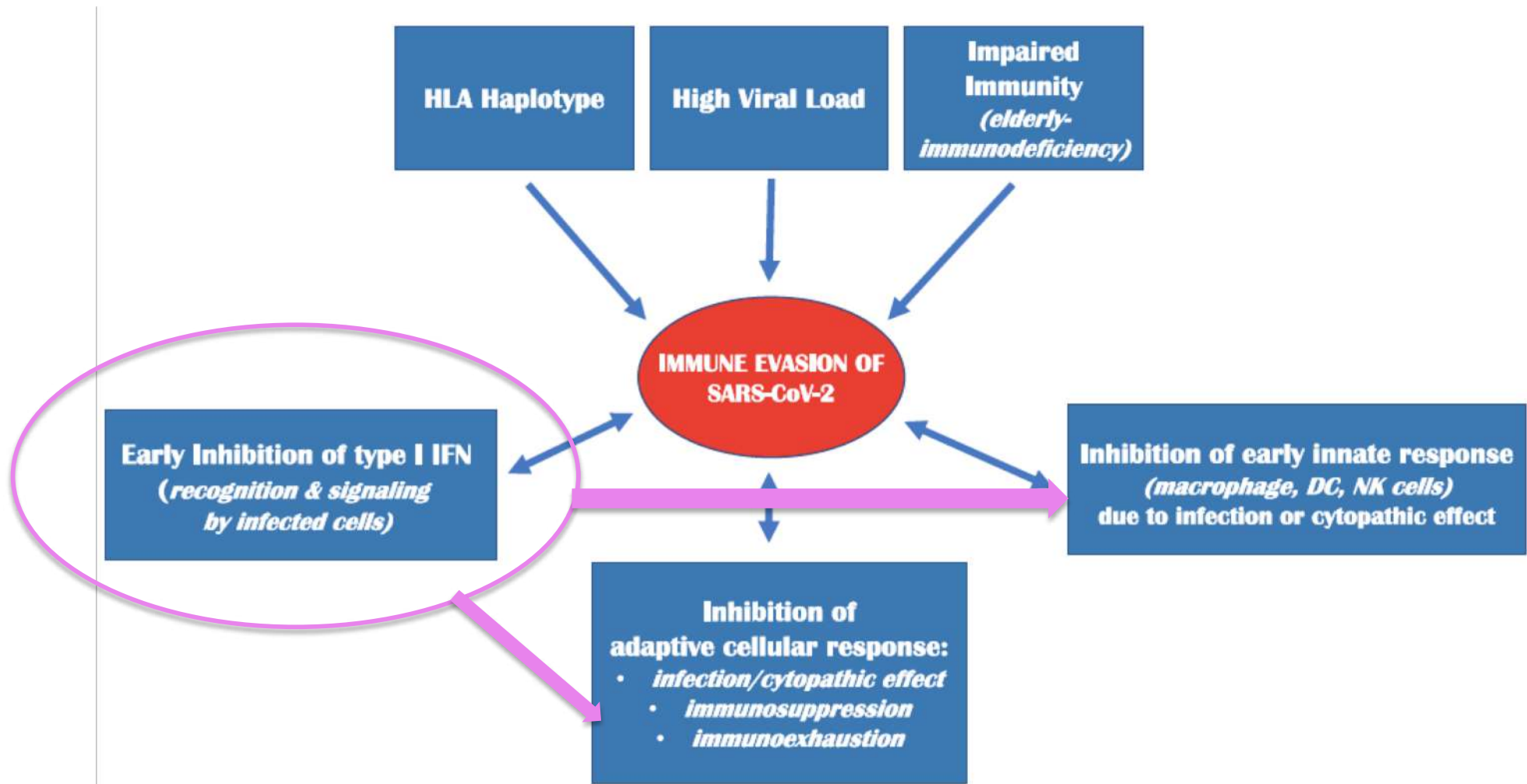
L'inibizione della produzione di IFN β da parte del CoV-2 è confermata dalla misurazione dei livelli di diverse citochine nei pazienti COVID-19. Un'analisi immunitaria integrata di trascrittomico da sangue intero e quantificazione delle citochine su pazienti COVID-19 da 8 a 12 giorni dopo l'insorgenza della malattia ha mostrato una risposta IFN-I ridotta. Questo si traduce in una bassa produzione di geni stimolati dall'interferone (ISGs). Al contrario, sono riscontrati alti livelli di IL6 e TNF α .

SARS-CoV-2 has multiple effects on the immune system, including **inhibition of IFN β production**, which results in ISGs not to be produced, CD4+ and CD8+ exhaustion and increased levels of pro-inflammatory proteins (TNF α , IL6, NF- κ B). Currently, the most promising drugs against COVID-19 include IFN-Is, antiinflammatory and antiviral drugs, protease inhibitors, antibodies, SARS-CoV2 – ACE2 (receptor) binding inhibitors and more.

The mechanisms used by Coronaviruses to manipulate the IFN type I response can be divided into three categories:

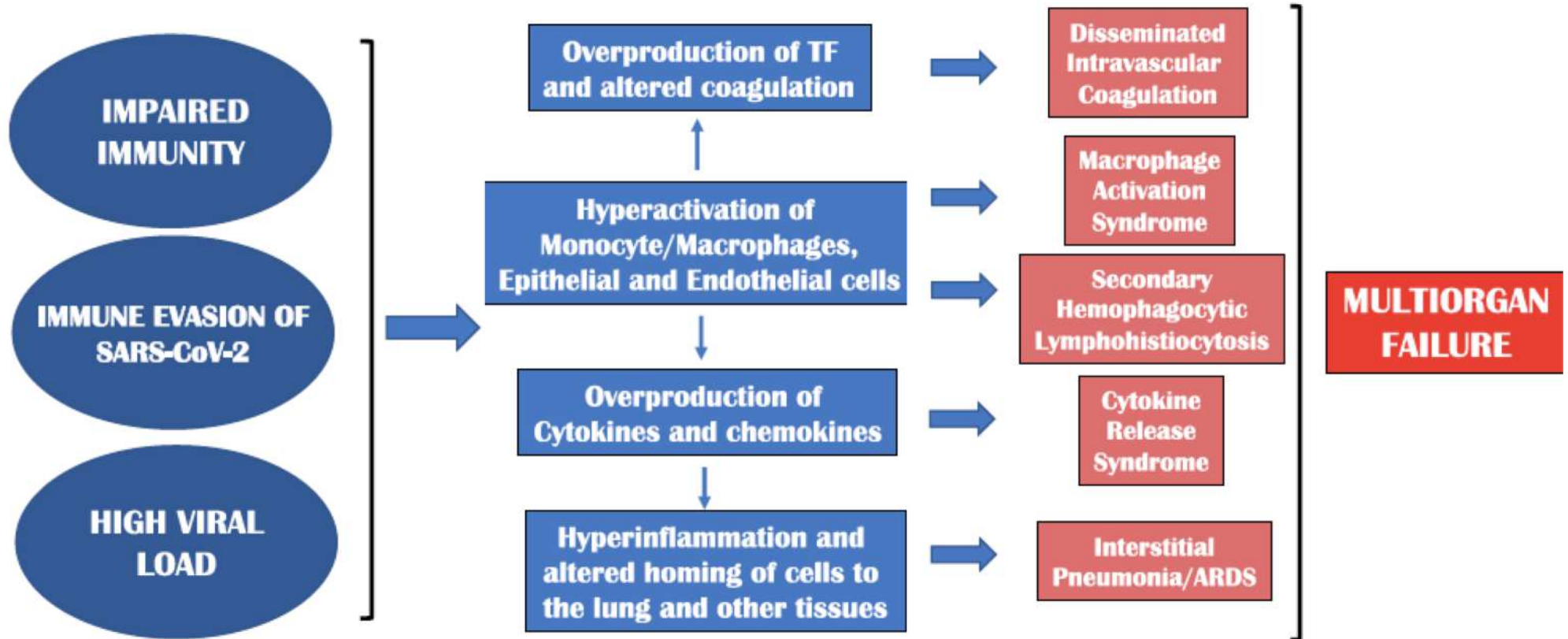
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1. Avoidance, where the virus protects itself from **recognition by PRRs**.
 2. IFN induction suppression, where the virus inhibits the **transcription of interferons**.
 3. IFN **signaling** suppression, where viral proteins inhibit IFNAR signalling.

Possible mechanisms of immune evasion of SARS-CoV-2



Possible mechanisms of immune evasion of SARS-CoV-2. Immune evasion of SARS-CoV-2 may be favored in individuals with compromised ability to mount efficient immune responses such as old people and patients with immunodeficiency or individuals carrying HLA alleles unable to properly present SARS-CoV-2 peptides to T lymphocytes. In addition, a high viral load may overcome the barriers of the immune responses. Notably, viruses escaping control may inhibit IFN-I and infect cells of both innate and adaptive immunity by exerting a cytopathic effect. In turn, the compromised function of immune cells and the impaired antiviral effect of IFN-I would further favor immune evasion, resulting in highly detrimental pathological effects. DC, Dendritic cell.

Tentative pathogenetic mechanisms of severe COVID-19



The mechanisms of immune evasion adopted by SARS-CoV-2 with other factors such as the viral load or the impaired immune response can contribute to the immuno-pathogenesis of COVID-19. In the presence of a defective clearance of the virus, a persistent **hyperactivation of monocyte/macrophage** compartment predominates as a compensatory mechanism. This in turn leads to overproduction of cytokines and chemokines, altered homing of cells into the lung and other tissues, and epithelial and endothelial damage with overexpression of tissue factor and persistent thrombophilia. Each of these biological changes may be responsible for some pathological conditions (partially overlapping) observed in COVID-19 as the cytokine release syndrome, the macrophage activation syndrome, the interstitial pneumonia with acute respiratory distress syndrome, the secondary hemophagocytic lymphohistiocytosis, or the disseminated intravascular coagulation. Each disorder constitutes the final pathological pictures of COVID-19, each being able to set off the multiorgan failure and death. Importantly, comorbidities and concomitant therapies can influence/enhance the above cascade of events. ARDS, Acute respiratory distress syndrome; TF, Tissue Factors.

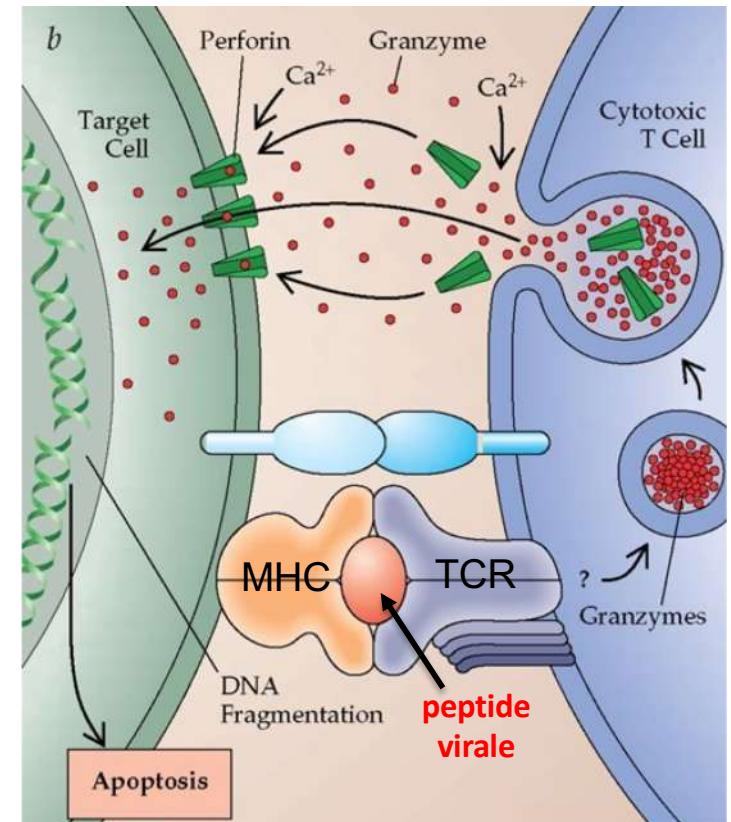
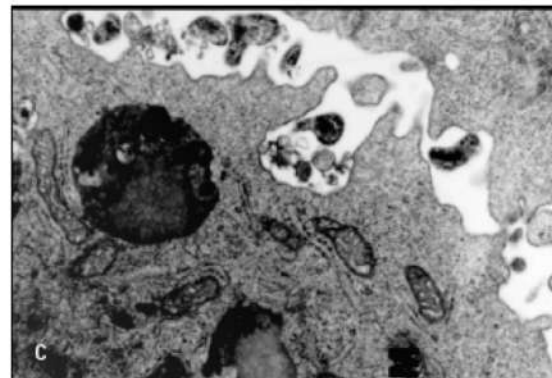
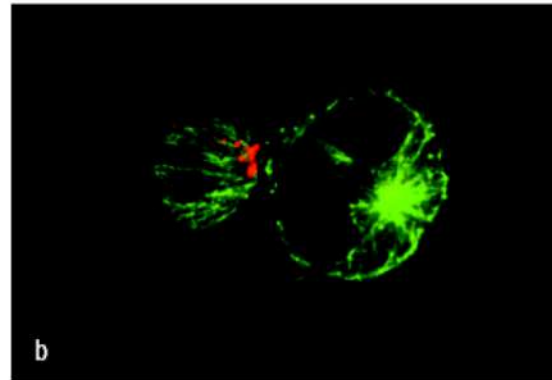
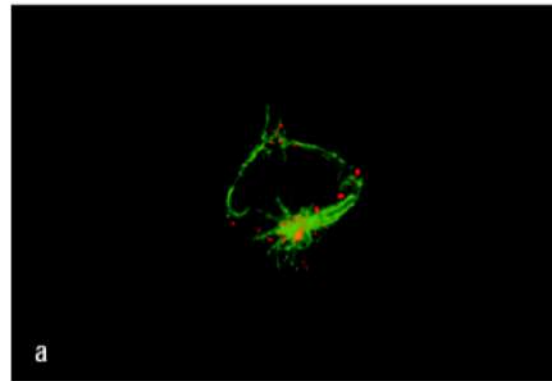
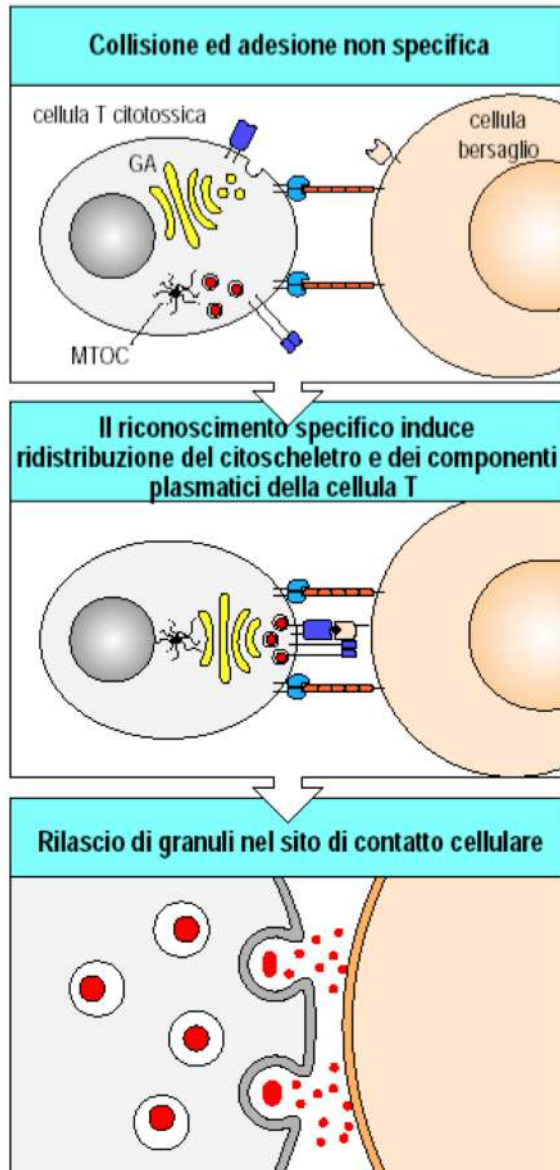
Immunità umorale e risposte T citotossiche sono essenziali per risolvere le infezioni virali



Virus

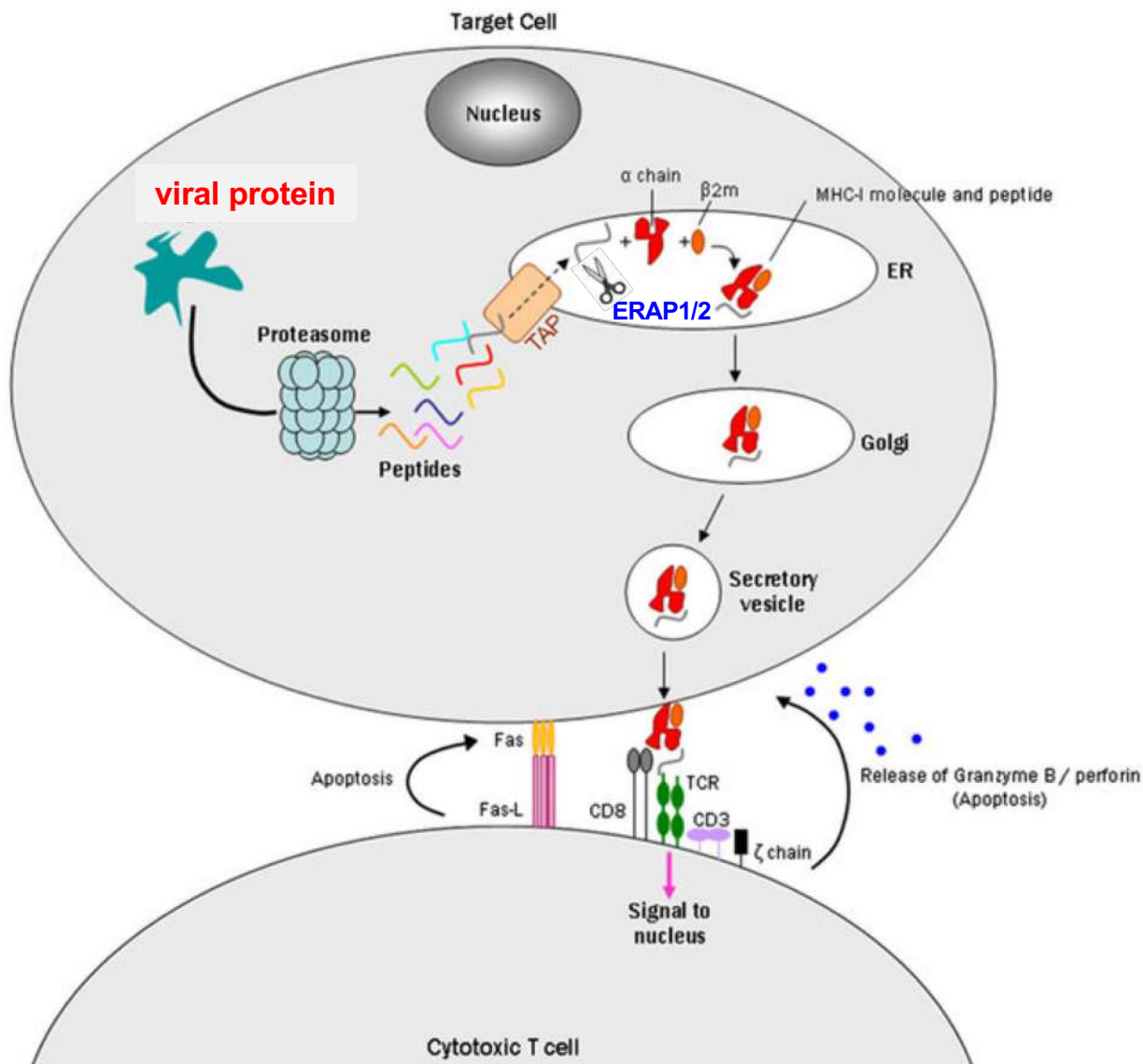
Agente infettivo	Malattia	Immunità umorale				Immunità cellulo-mediata	
		IgM	IgG	IgE	IgA	Cellule T CD4 (macrofagi)	Cellule T killer CD8
Virus del vaiolo	Vaiolo					Yellow triangle	Red triangle
Varicella zoster	Varicella	Yellow triangle	Yellow triangle				Yellow triangle
Virus di Epstein-Barr	Mononucleosi		Yellow triangle				Red triangle
Virus dell'influenza	Influenza		Red triangle		Red triangle		Yellow triangle
Virus della parotite	Parotite		Red triangle				Yellow triangle
Virus del morbillo	Morbillo		Red triangle				Red triangle
Polio virus	Poliomielite		Yellow triangle		Yellow triangle		Yellow triangle
Virus dell'immunodeficienza umana	AIDS		Yellow triangle				Yellow triangle

I linfociti T CD8+ citotossici eliminano la cellula bersaglio infettata dal virus dopo riconoscimento dell'antigene



Molecole citotossiche effettrici	Altre
Perforina Granzimi Ligando Fas	IFN- γ TNF- β TNF- α

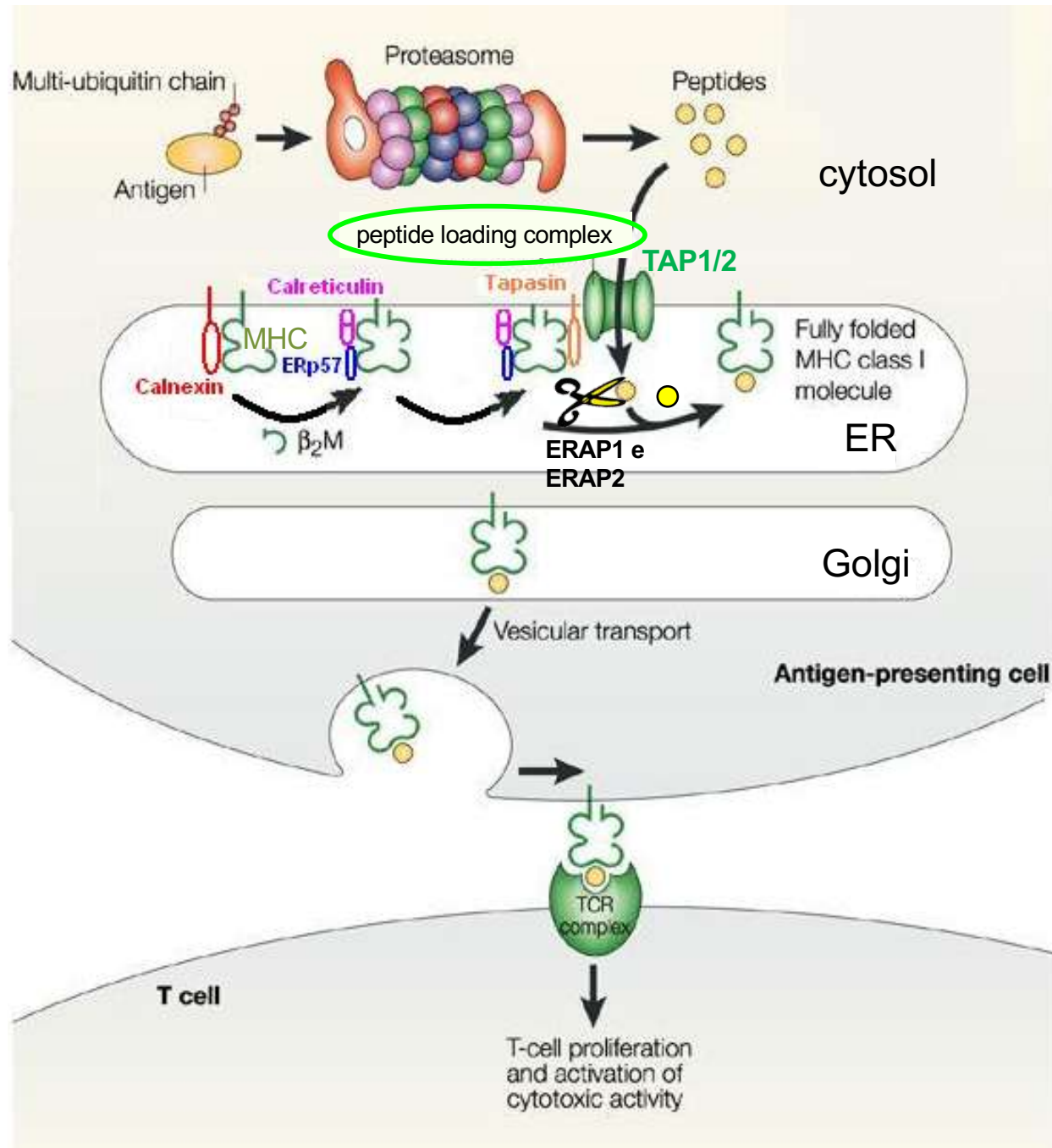
Model of a CD8+ cytotoxic T cell targeting an infected cell



Intracellular antigens are digested by the proteasome into small peptides. A specialized carrier, the transporter associated with antigen processing (TAP) complex, translocates the peptide into the endoplasmic reticulum (ER), allowing the antigen to bind MHC class I, which consists of an alpha and beta 2 microglobulin ($\beta 2m$) chain. Endoplasmic reticulum-derived vesicles that contain the complex fuse with the Golgi. The complex is then packaged in secretory vesicles that fuse with the plasma membrane to insert the complex on the cell surface. Major histocompatibility complex I molecules present antigens to CD8+ cytotoxic T cells. The T cell CD8+ molecule recognizes MHC-I, while TCRs recognize specific antigenic peptides. The TCR complex contains CD3 and ζ (zeta) chains. Once the CD8+ T cell recognizes the target cell, apoptosis can occur in two ways: one uses secretion of granzyme B and perforin from T cell granules into the intercellular space between the cells.

The other uses the Fas ligand protein, which is expressed on the surface of the CD8+ T cells and binds to the Fas receptor on the target cell, which triggers apoptosis through the classical caspase cascade.

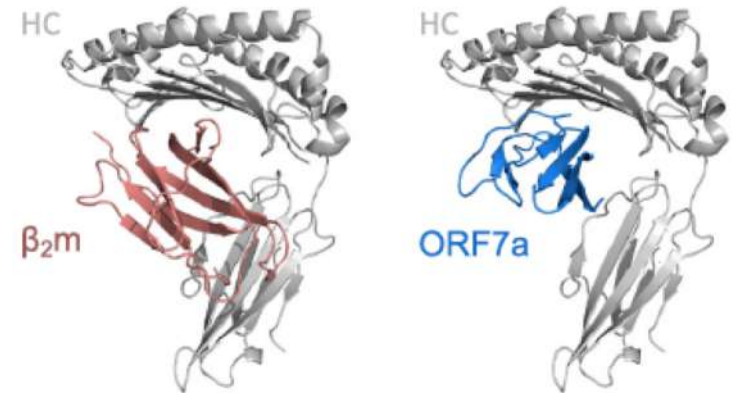
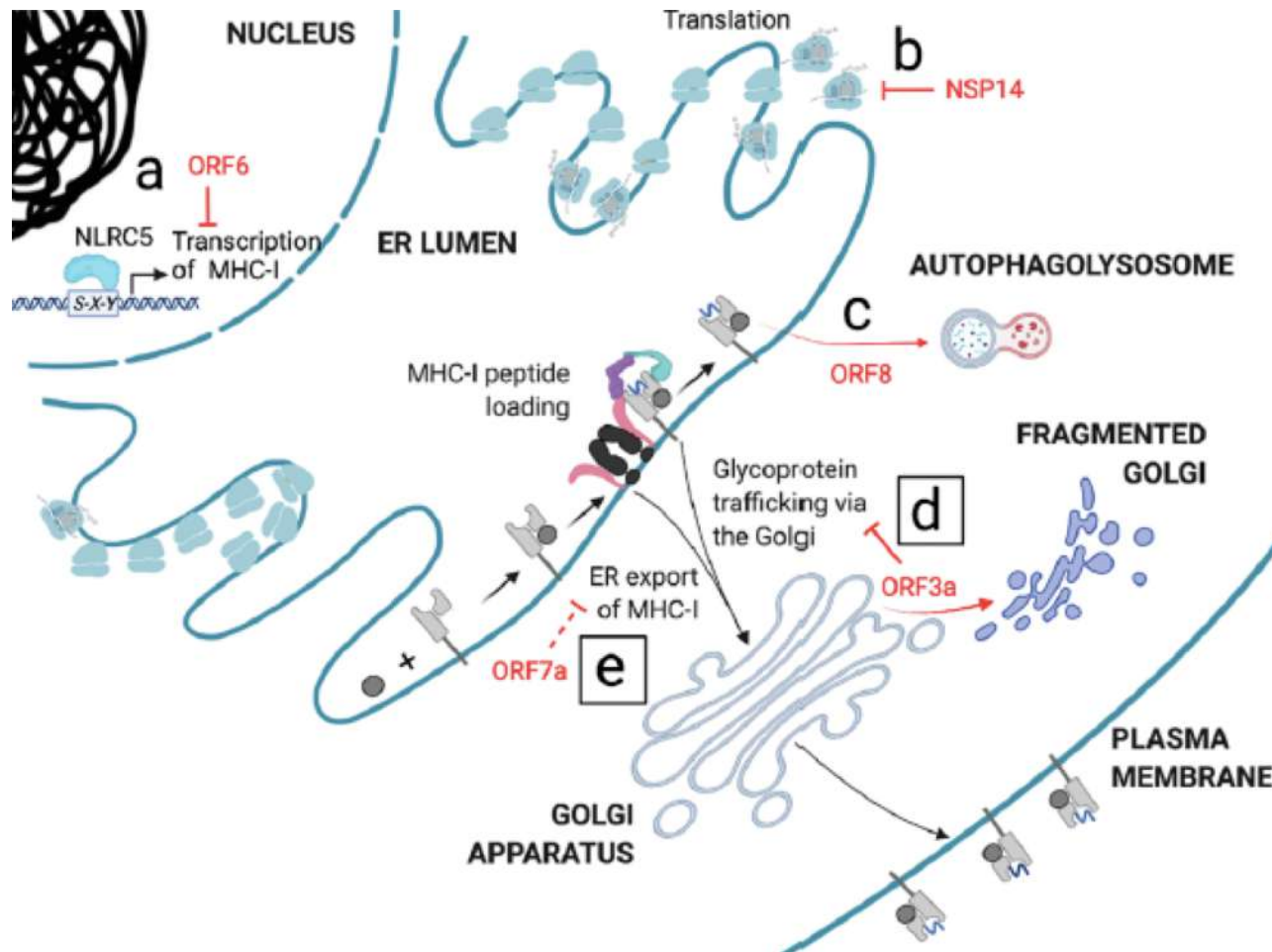
Pathway di processazione degli antigeni e presentazione da parte delle molecole di classe I dell'MHC



The MHC class I antigen presentation pathway.

- (1) Proteins are proteolytically processed in the cytosol by the proteasome.
- (2) Peptides generated by the proteasome are translocated into the ER lumen by TAP.
- (3) MHC class I molecules (heavy chain and associated β_2M) fold and assemble in the ER lumen with the aid of the ER chaperones calnexin, calreticulum and ERp57.
- (4) The MHC class I molecule in a complex with calreticulum and ERp57 associates with TAP and tapasin (peptide loading complex) facilitates peptide binding.
- (5) Peptide loaded MHC class I molecules dissociate from TAP and are transported through the secretory pathway to the plasma membrane.

Meccanismi di SARS-CoV-2 per evadere la risposta adattativa mediata dalle molecole MHC-I

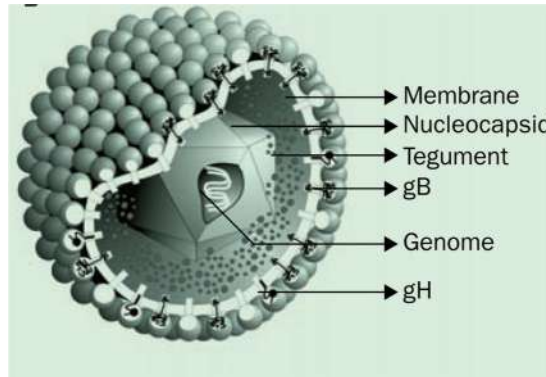
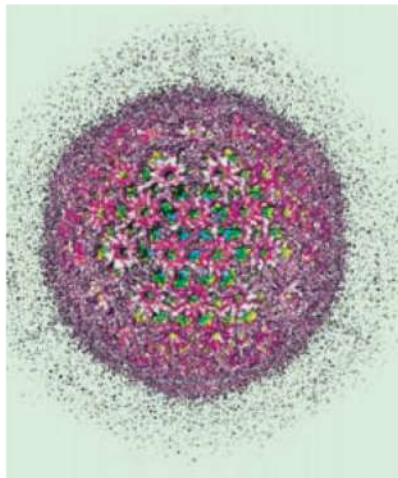


Proposed mechanism by which ORF7a slows the export of MHC-I from the ER. In uninfected cells, the free heavy chain associates with β_2m to form the MHC-I heterodimer. This associates with the PLC and MHC-I molecules loaded with peptide (pMHC-I) are released for export from the ER en route to the cell surface (Upper). ORF7a interacts with free heavy chain, interfering with its association with β_2m and thus the peptide-loading complex, delaying exit from the ER (Lower).

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<https://doi.org/10.1073/pnas.2208525120>

Various gene products of SARS-CoV-2 down-regulate MHC-I-mediated immune responses in an infected cell. ORF6 prevents the transcriptional upregulation of MHC-I following infection (a), while NSP14 globally down-regulates translation (b). ORF8 interacts with HLA-A2 and targets it to the autophagosome where it is destined to be degraded on fusion with lysosomes to form the autophagolysosome (c). We report that the expression of ORF3a causes Golgi fragmentation, inhibiting the trafficking of MHC-I and other surface proteins (d) and that ER-localized ORF7a impedes the export of MHC-I from the ER, down-regulating surface MHC-I (e).

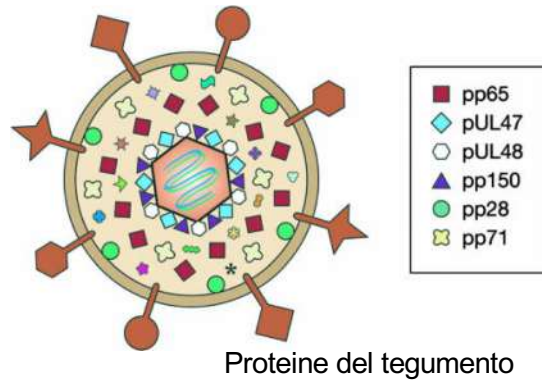
Herpesvirus e interferenza con la presentazione antigenica da parte delle molecole MHC di classe I



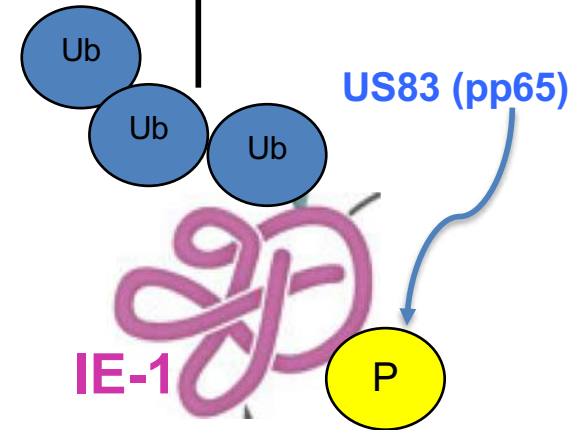
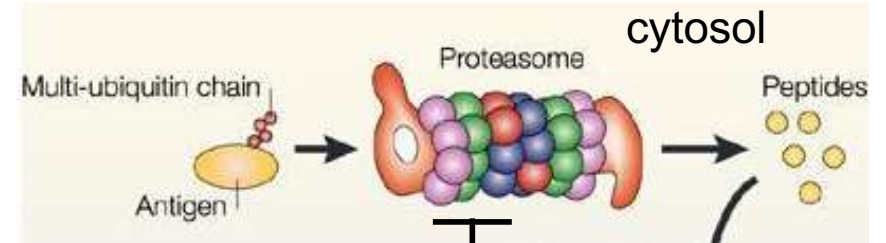
Meccanismi di immunoevasione di Citomegalovirus (CMV)

Interferenza con il processamento degli antigeni

-Inibizione dell'attività del proteasoma

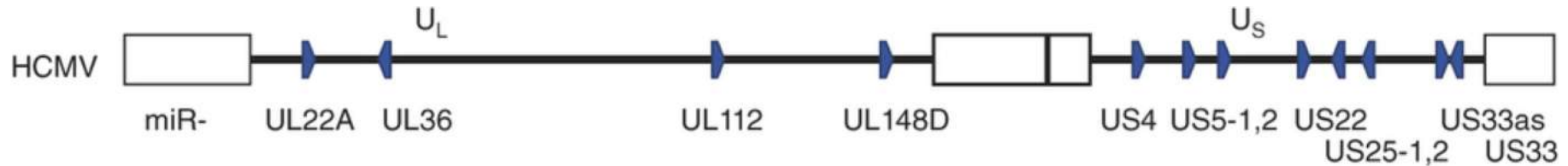


ESEMPIO: **US83 (pp65)**
 di **HCMV** media la fosforilazione della proteina **IE-1** (immediate early-1) inibendone l'interazione con il proteasoma e quindi la degradazione e la presentazione antigenica



-Inibizione dell'attività di ERAP1 (ER aminopeptidase 1)

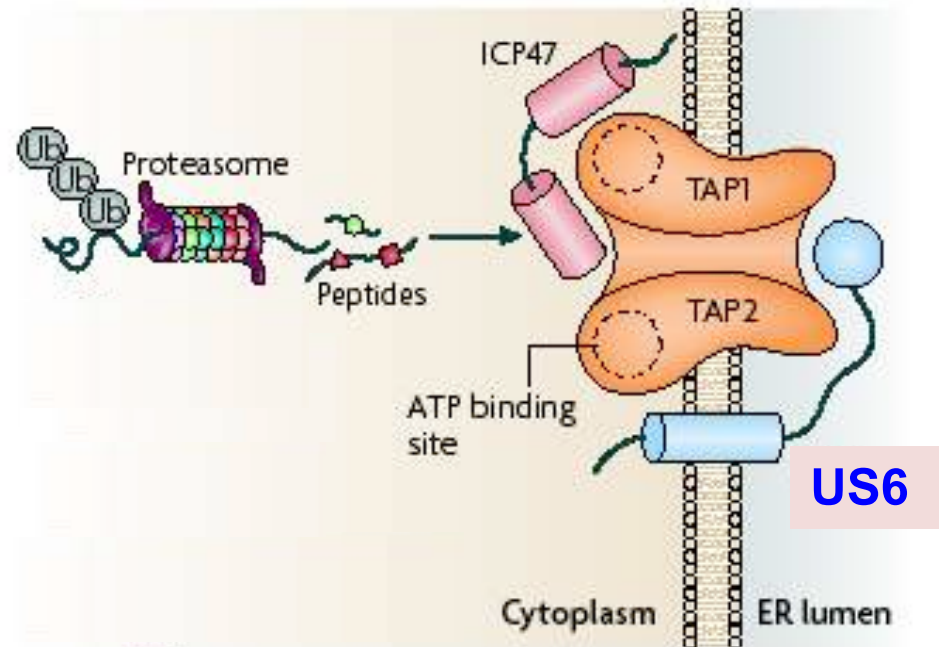
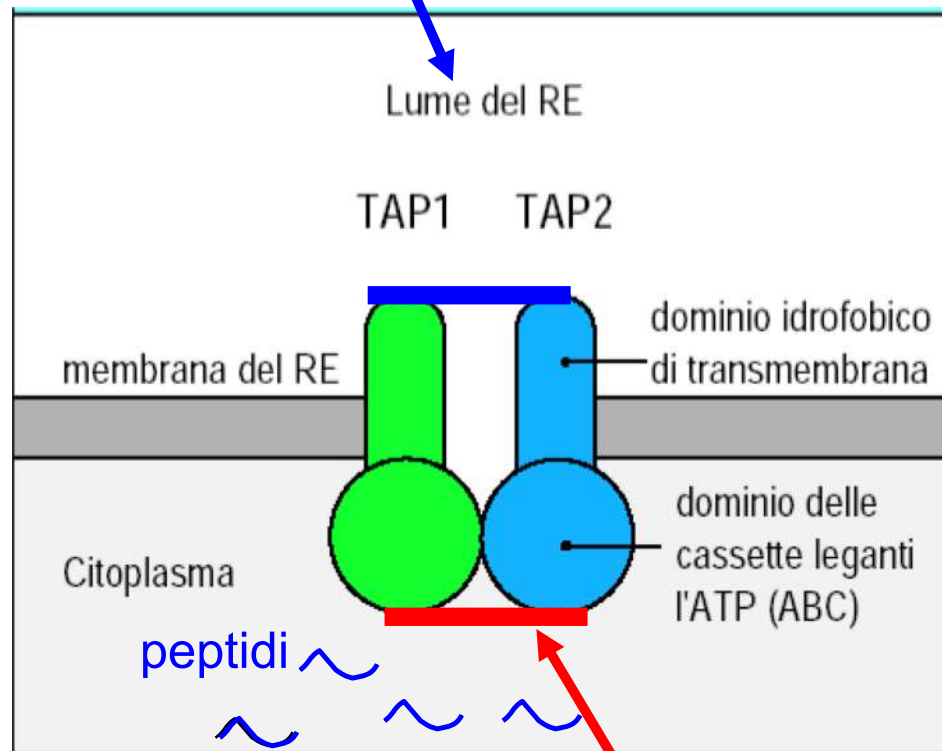
HCMV produce miR-US4-1 che specificamente down-modula ERAP1



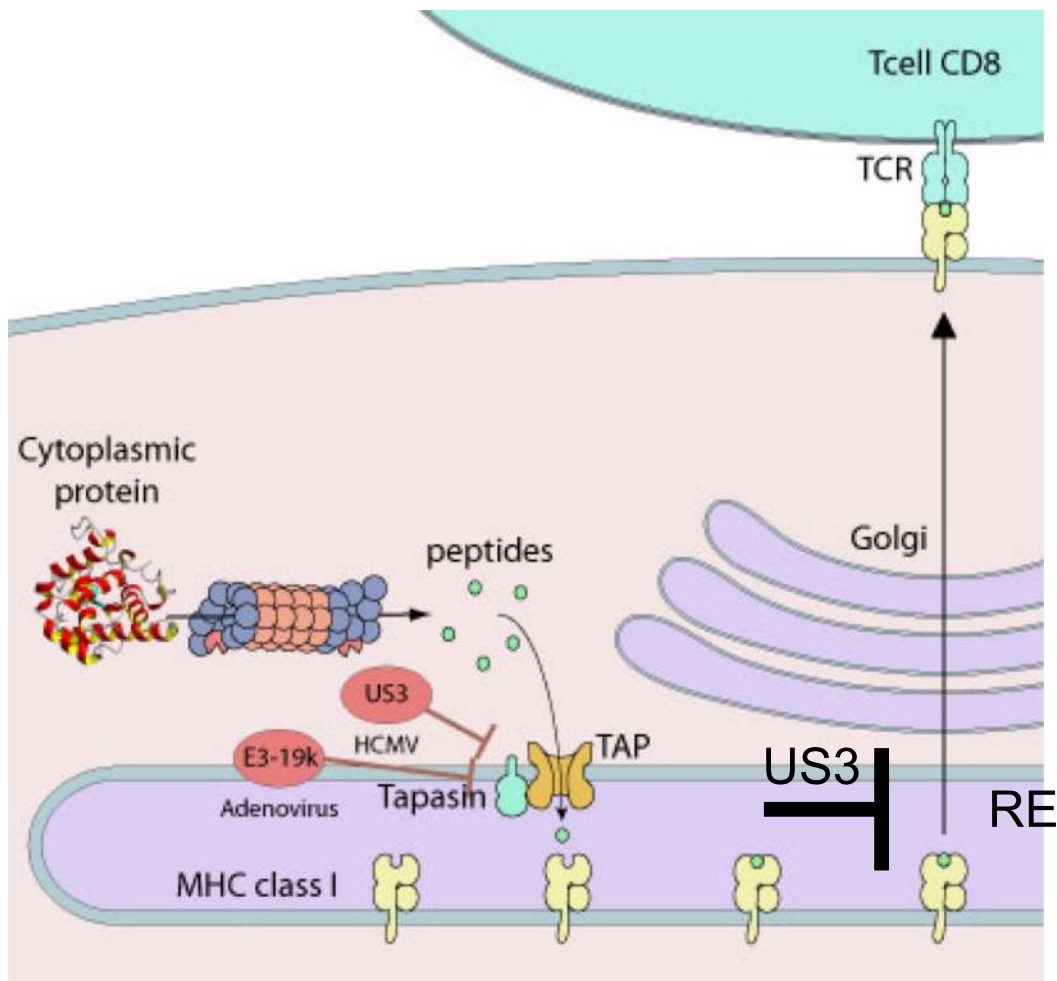
This represents an overview of the repeat elements and viral miRNAs present in HCMV

Interferenza con la traslocazione dei peptidi dal citosol al reticolo endoplasmatico attraverso le molecole TAP

Il prodotto di **US6** (unique short region) di HCMV inibisce il riarrangiamento conformazionale delle molecole TAP richiesto per il trasferimento dei peptidi all'interno del lume del RE e lo fa interferendo con il binding dell'ATP alle molecole TAP e la successiva idrolisi che fornirebbe energia per il cambio conformazionale.



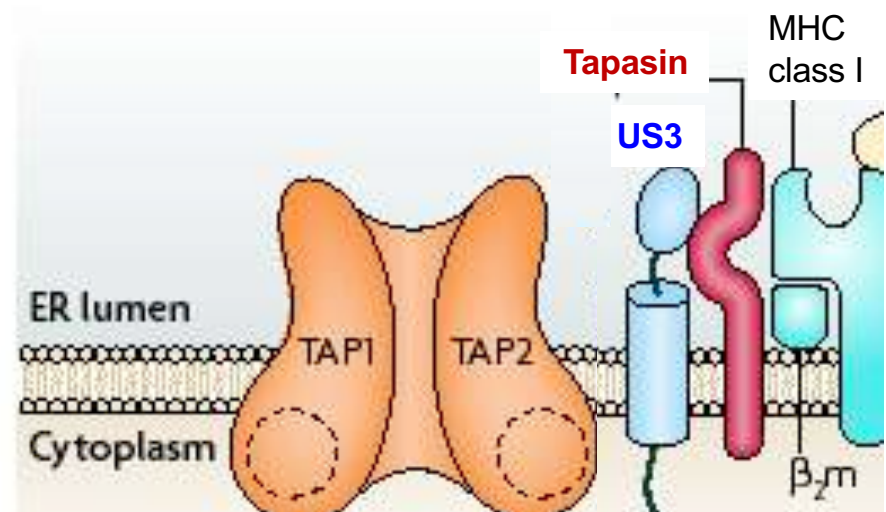
HSV1 e 2 (herpes simplex virus) producono **ICP47** (infected-cell protein 47) che impedisce il binding dei peptidi ai TAP fungendo da inibitore competitivo (la minima regione richiesta per l'inibizione va dall'aa 3 al 34)



Interferenza di US3 (HCMV) con il processo di “peptide editing” delle molecole MHC di classe I nel RE

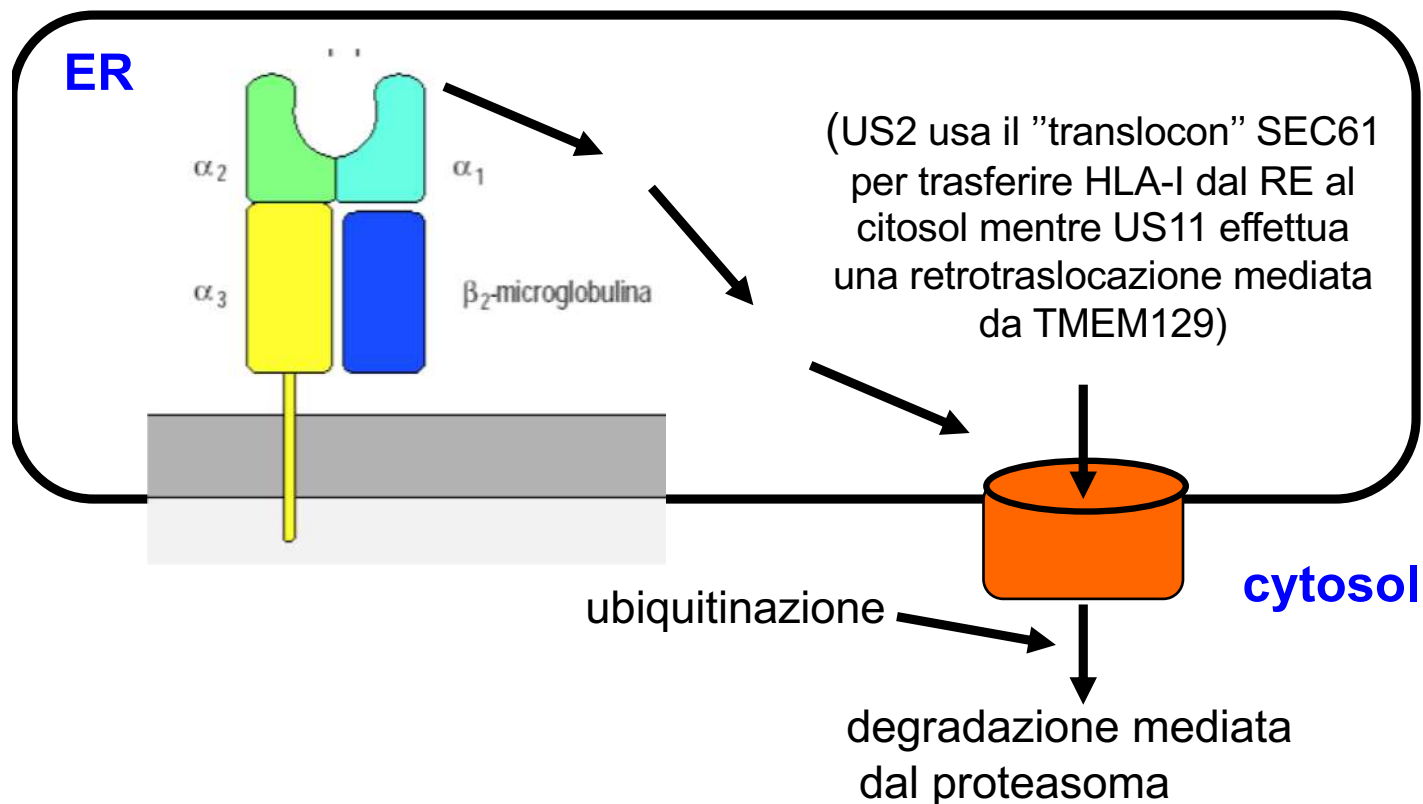
US3 di HCMV è una glicoproteina transmembrana di tipo I che agisce come inibitore della tapasina impedendo che questa faccia da ponte tra TAP e MHC di classe I nel PLC (peptide loading complex), alterando il processo definito di “peptide editing” e causando ritenzione delle molecole di classe I nel RE ed inibizione del processo di maturazione.

Figure 3 | **Modulation of tapasin function and retention of MHC class I molecules by viral immune evasion proteins.** Tapasin is a crucial component of the peptide-loading complex (PLC) and has a key role in influencing the generation of the repertoire of peptides presented by MHC class I molecules on the cell surface. The US3 protein from human cytomegalovirus binds to tapasin and inhibits its ability to facilitate the binding of kinetically stable peptides to MHC class I molecules (known as peptide optimization),

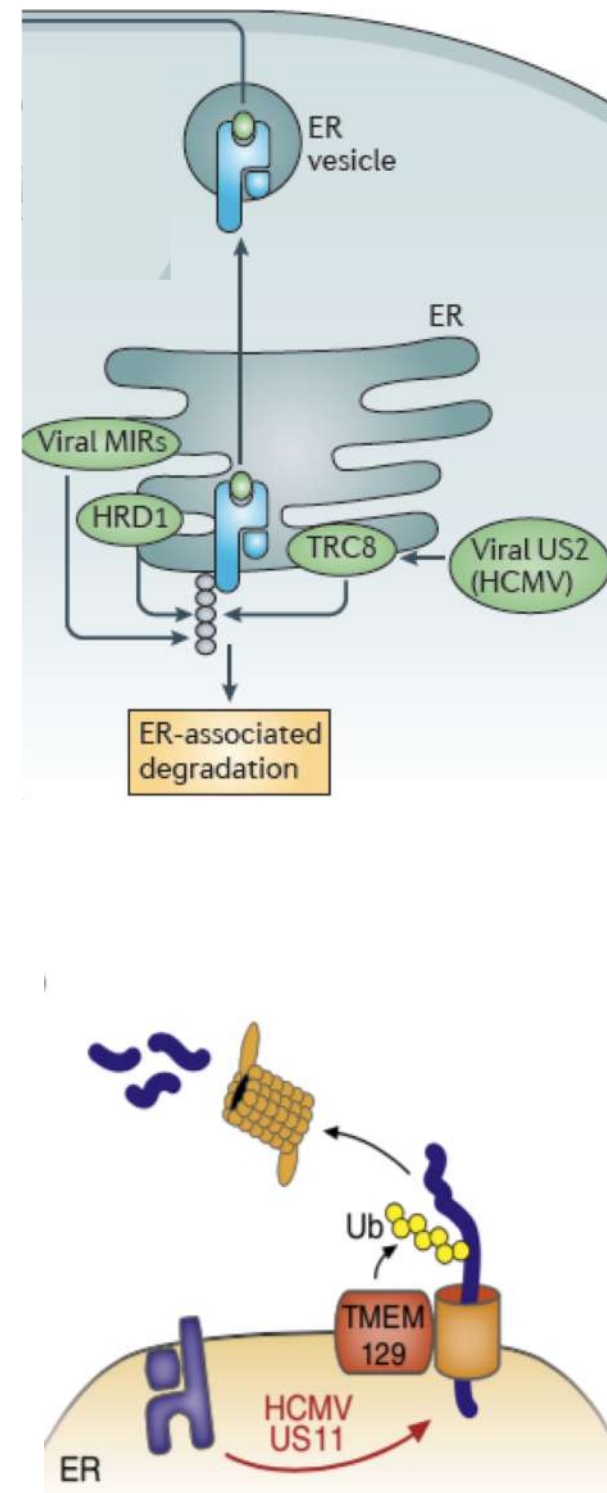


Trasporto retrogrado delle molecole HLA di classe I dal RE al citosol, ubiquitinazione e degradazione via proteasoma

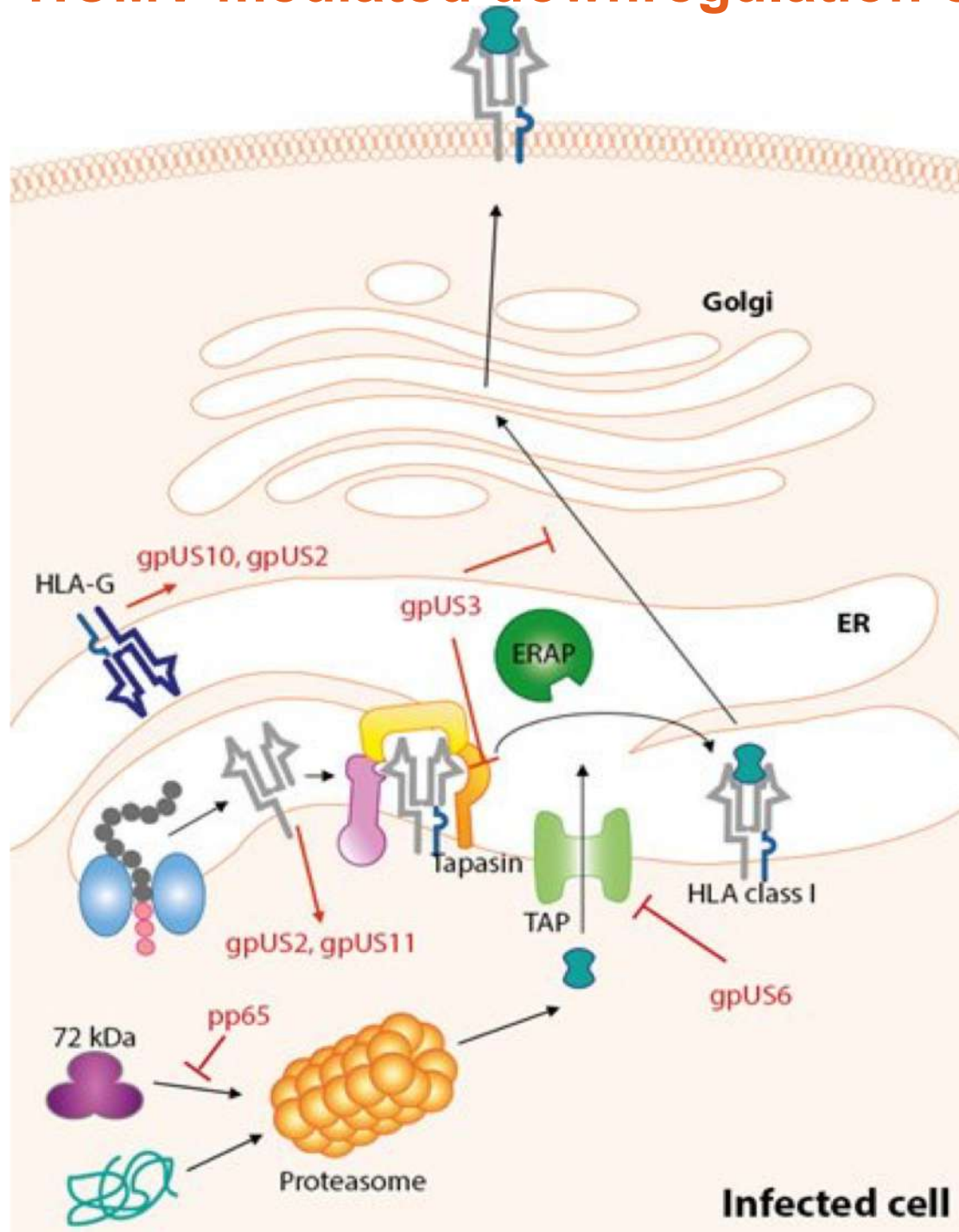
US2 e US11 di HCMV sono glicoproteine transmembrana di tipo I che inducono la degradazione selettiva delle molecole HLA di classe I dei loci A e B (ma non E e C) fungendo da **“ERAD inducers”**. Agiscono con meccanismi diversi infatti US2 utilizza TRC8 come E3-ubiquitina ligasi mentre US11 usa TMEM129. Non agiscono su UL18 di HCMV. La reazione è ATP dipendente (ruolo cruciale della AAA p97 ATPasi) ed influenzata dal potenziale redox.



ERAD= ER-associated protein degradation



HCMV-mediated downregulation of HLA-class I pathway

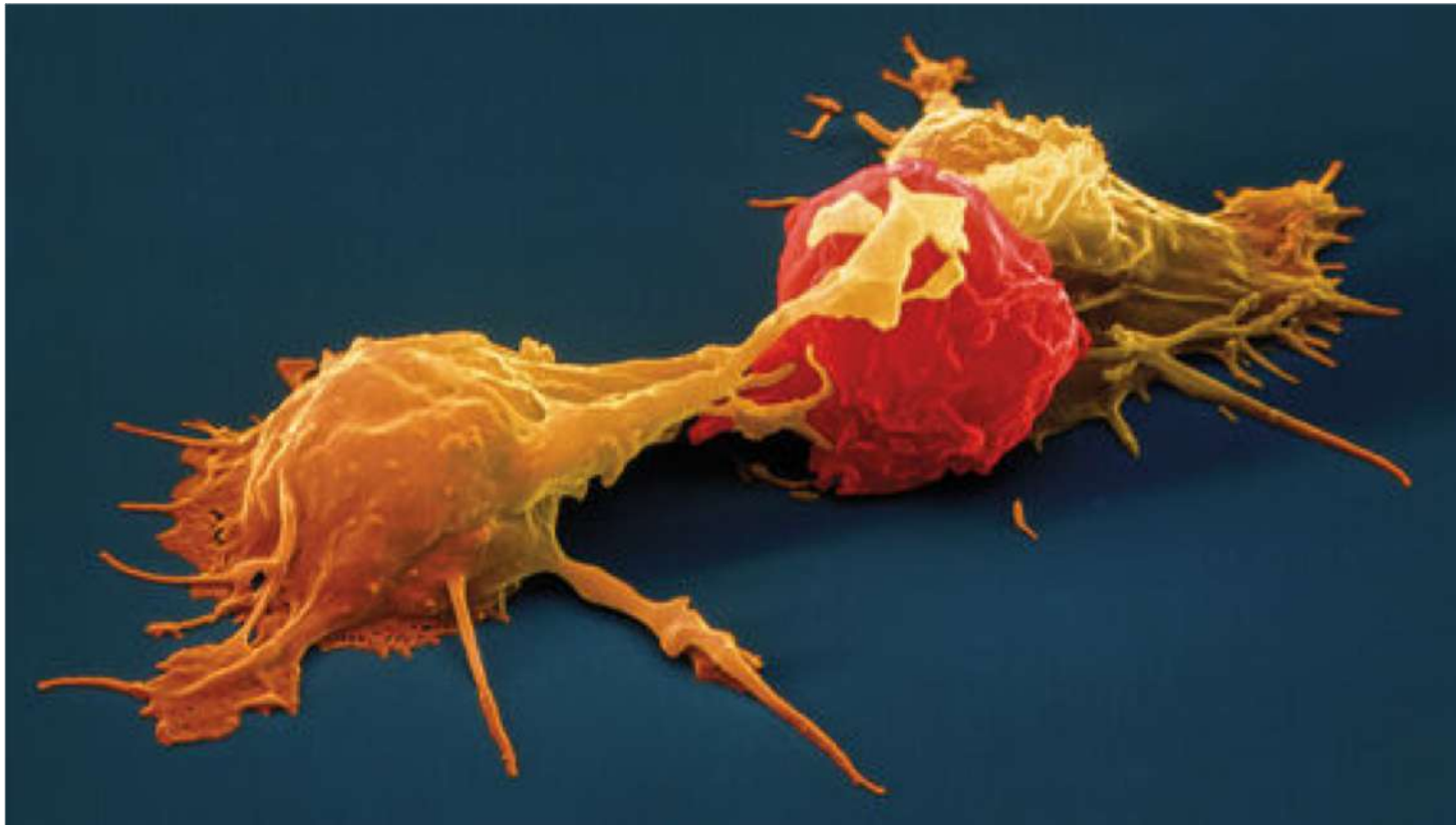


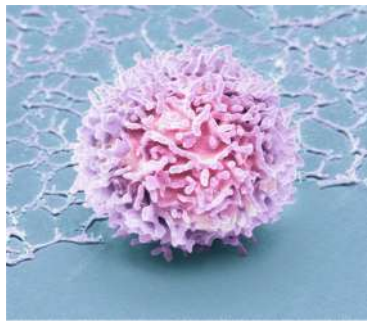
The **protein pp65** prevents the presentation of IE-derived peptides. Glycoprotein unique short 2 (**US2**) and **US11** (gpUS11) redirect HLA class I heavy chain (hc) from the endoplasmic reticulum (ER) to the cytosol and induce the proteasomal degradation of the molecule. Glycoprotein **US6** (gpUS6) inhibits the function of transporter associated with antigen processing (TAP). Glycoprotein **US3** (**US3**) inhibits the tapasin-dependent peptide loading and retains HLA class I molecules in the endoplasmic reticulum (ER). Glycoprotein **US10** (gpUS10) retains HLA class I hc in the ER. gpUS10 and gpUS2 induce degradation of HLA-G.

Immunità delle cellule natural Killer (NK)

e

meccanismi virali di evasione



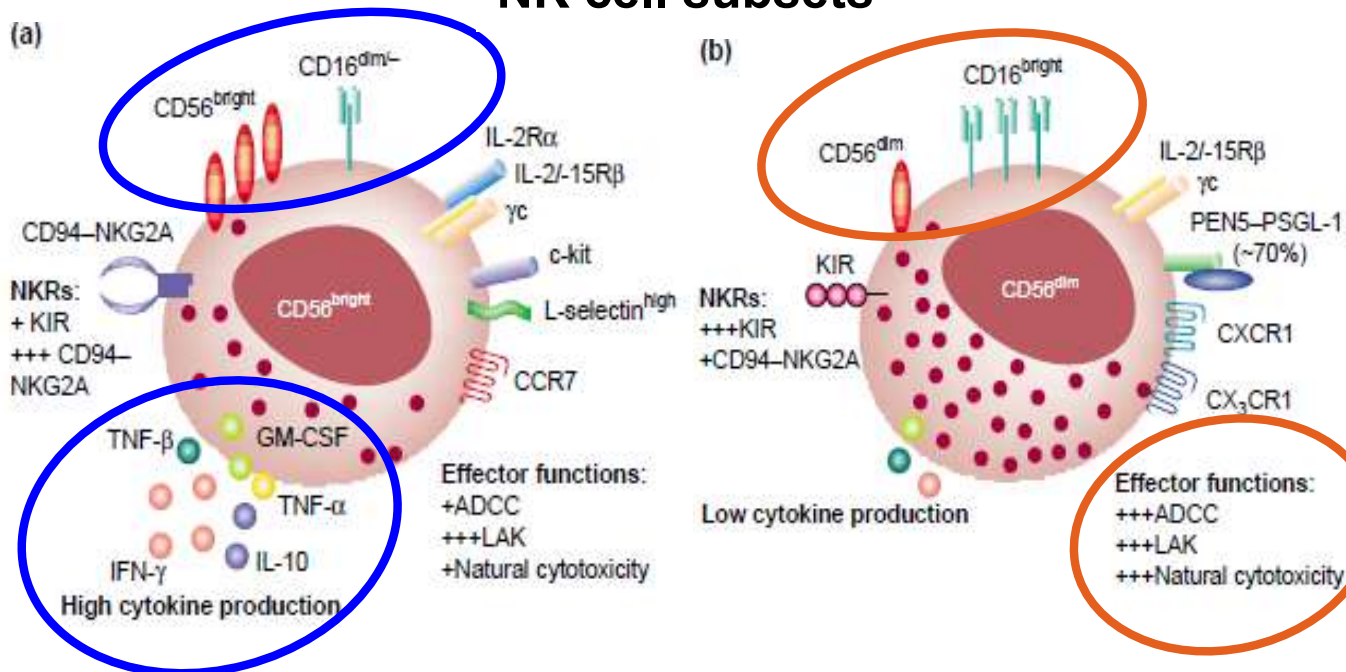


Cellule NK (cellule linfoidi innate ILC1)

- Originano da precursori midollari linfoidi
- Rappresentano il 5-15% delle cellule mononucleate del sangue periferico e della milza

- A differenza dei linfociti T e B non esprimono recettori come il TCR o il BCR derivati da riarrangiamento somatico ma recettori in configurazione “germinativa”
- L’attivazione è finemente regolata da segnali trasmessi da recettori attivatori ed inibitori e potenziata dalle citochine
- L’attivazione tramite recettori attivatori induce le funzioni effettrici (lisi citotossica, produzione di citochine)
- Marcatori caratteristici CD56 e CD16

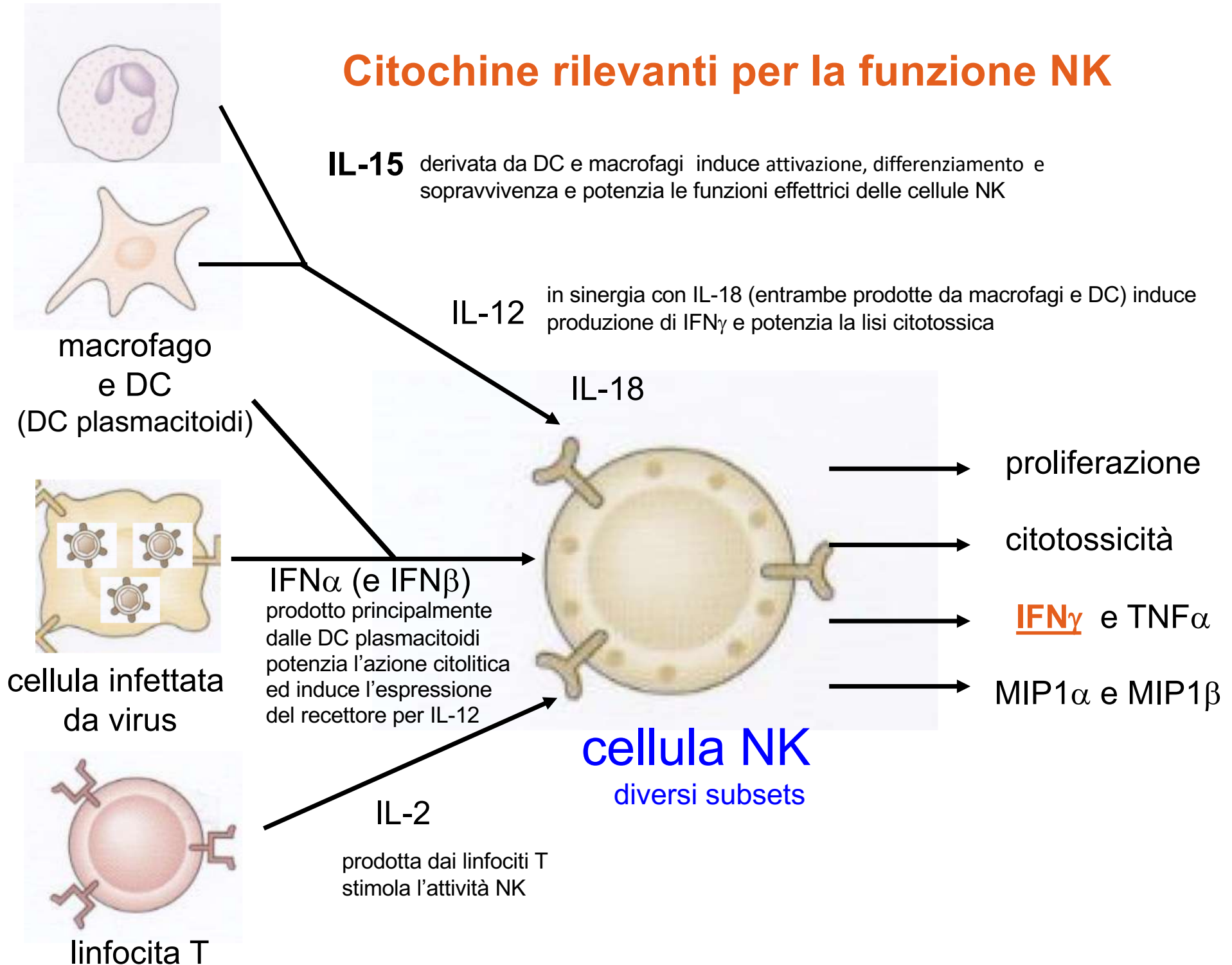
NK cell subsets



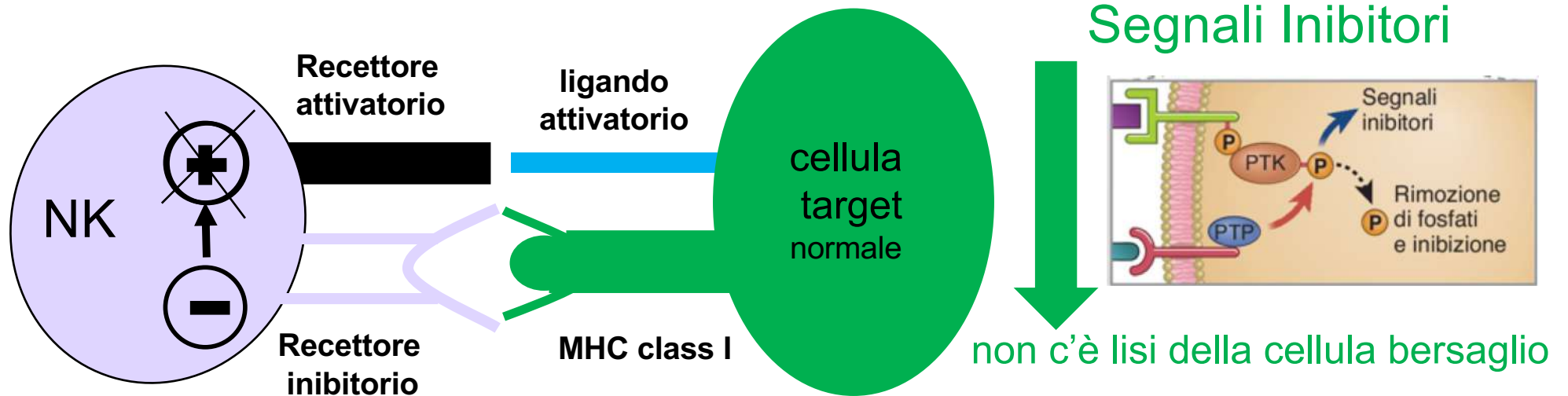
(a) CD56^{bright} NK cells produce high levels of cytokines following stimulation with monokines. This subset has low-density expression of CD16 and exhibits low natural cytotoxicity and antibody-dependent cellular cytotoxicity (ADCC), but potent lymphokine activated killer (LAK) activity. CD56^{bright} NK cells have high-level expression of the inhibitory CD94–NKG2A C-type lectin NK receptor (NKR) but have low level expression of killer Ig-like receptors (KIRs). The adhesion molecule L-selectin, which, in combination with CCR7, is involved in trafficking to secondary lymph nodes, is also found on CD56^{bright} NK cells.

(b) By contrast, CD56^{dim} NK cells produce low levels of NK-derived cytokines but are potent mediators of ADCC, LAK activity and natural cytotoxicity, and have a more granular morphology than CD56^{bright} NK cells. The CD56^{dim} NK-cell subset has high-level expression of KIRs. These cells have distinct expression of cytokine (e.g. IL-2R β) and chemokine (e.g. CXCR1 and CX3CR1) receptors. CD56^{dim} NK cells lack L-selectin.

Citochine rilevanti per la funzione NK

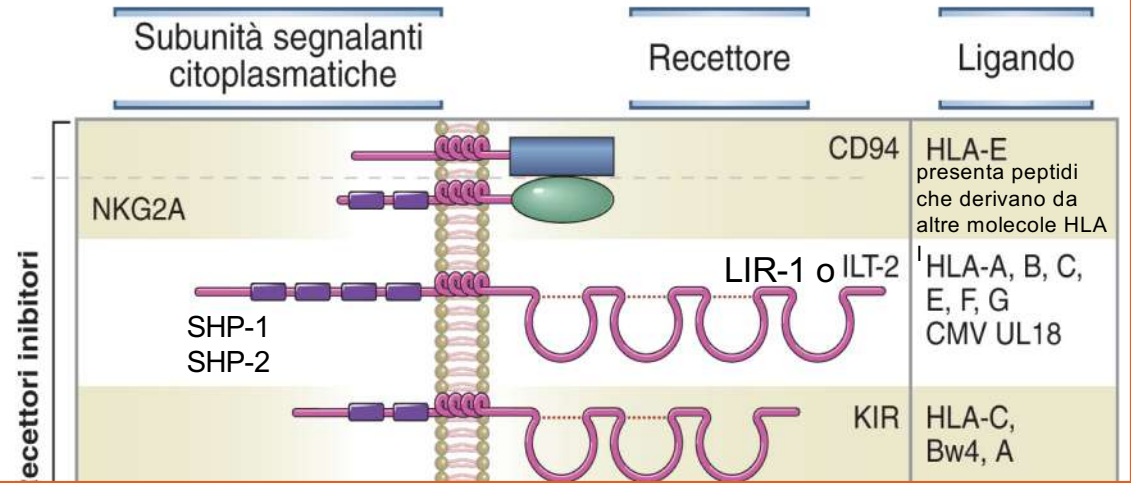


Come agiscono i recettori inibitori e attivatori delle cellule NK



Recettori NK inibitori

ITIM=immunoreceptor tyrosine-based inhibitory motif
 trasmettono segnali inibitori perché reclutano le PTP (protein tirosin fosfatasi) es: SHP-1; SHP-2; in seguito a fosforilazione delle tirosine

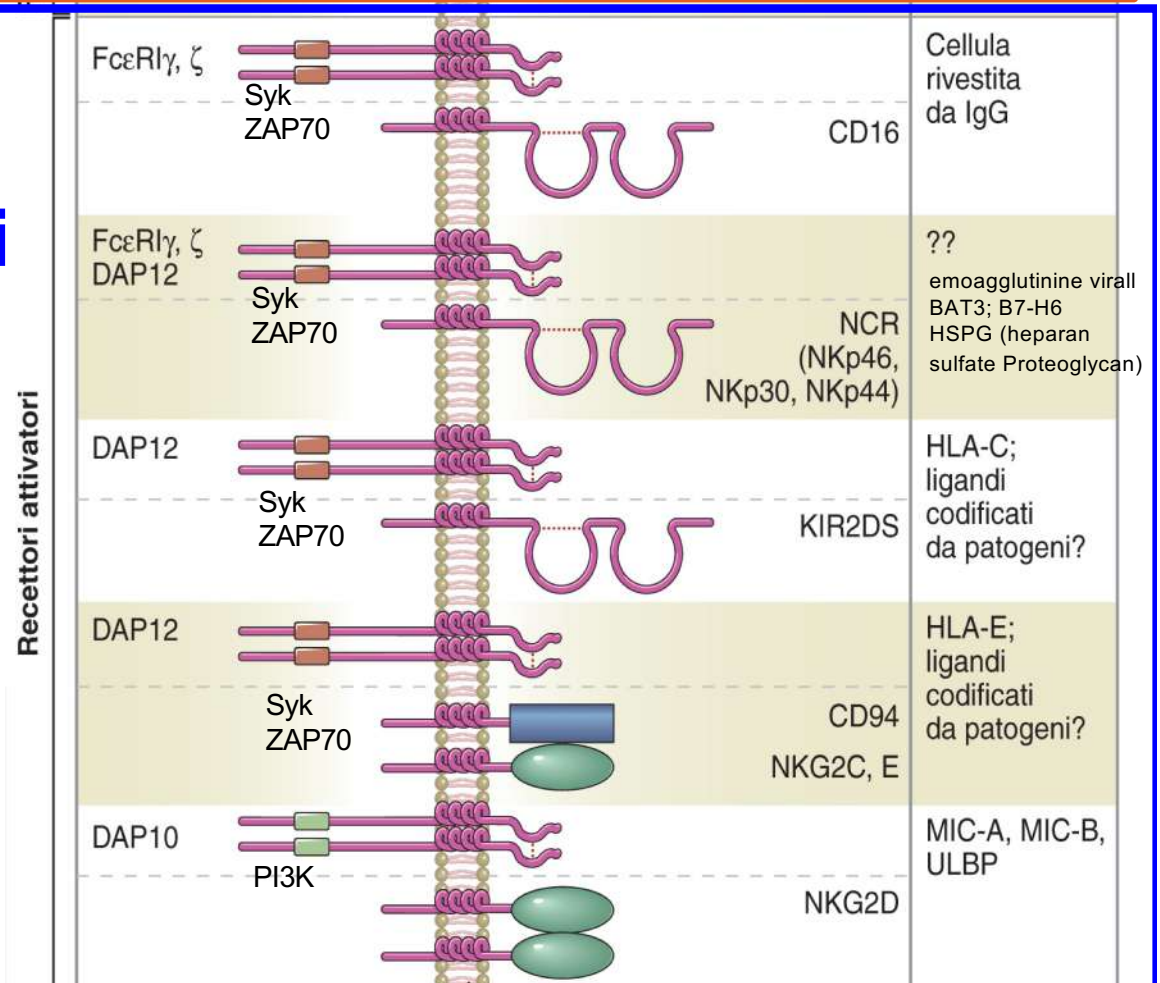


Recettori NK attivatori

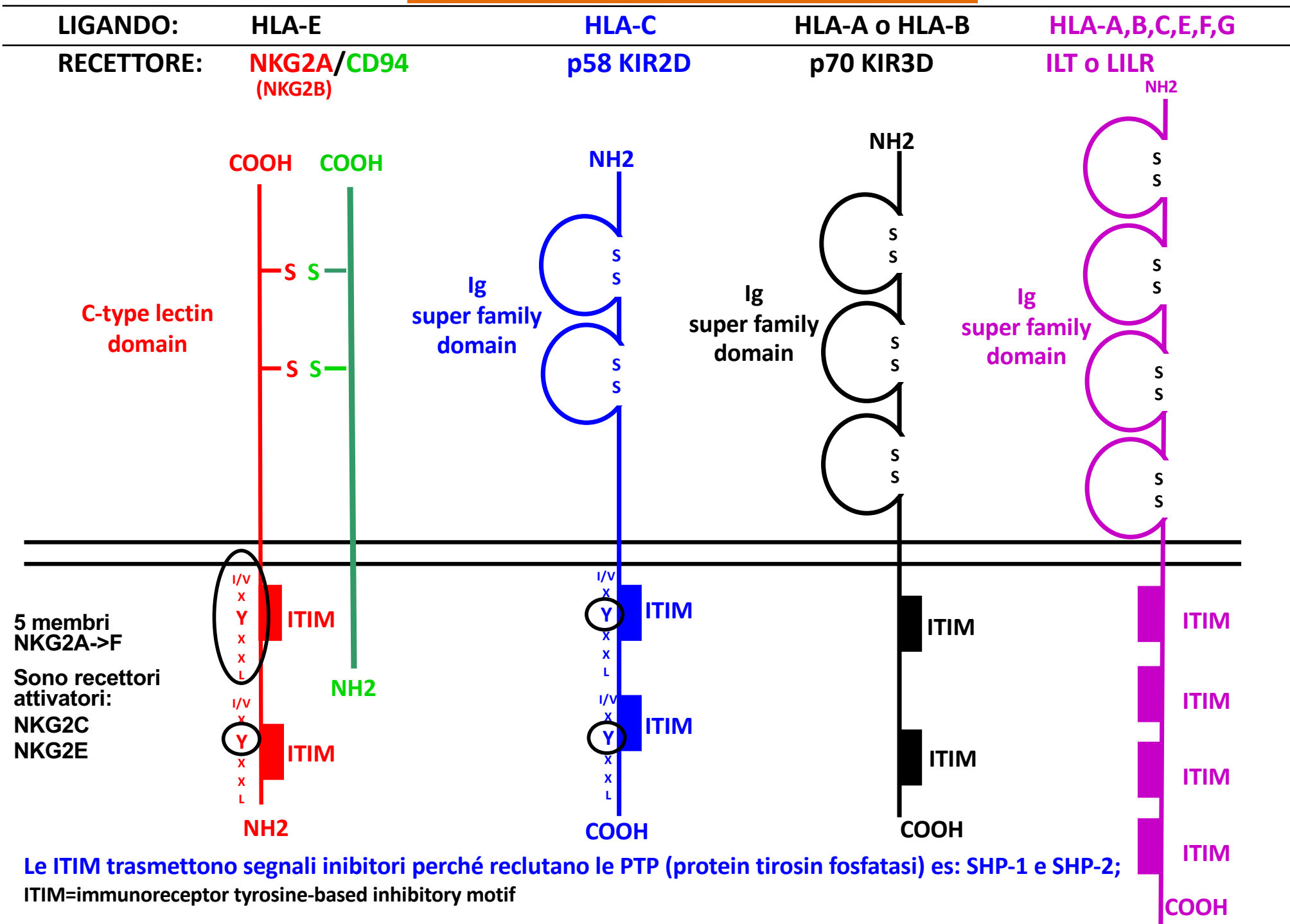
YXXL



ITAM=immunoreceptor tyrosine-based activating motif



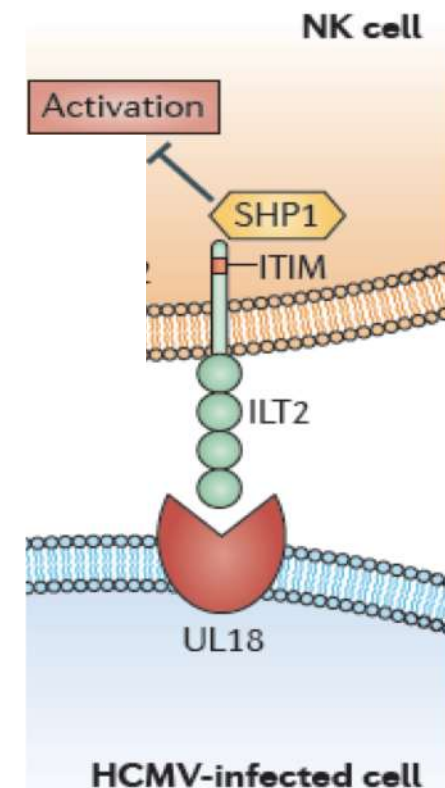
Recettori inibitori delle cellule NK



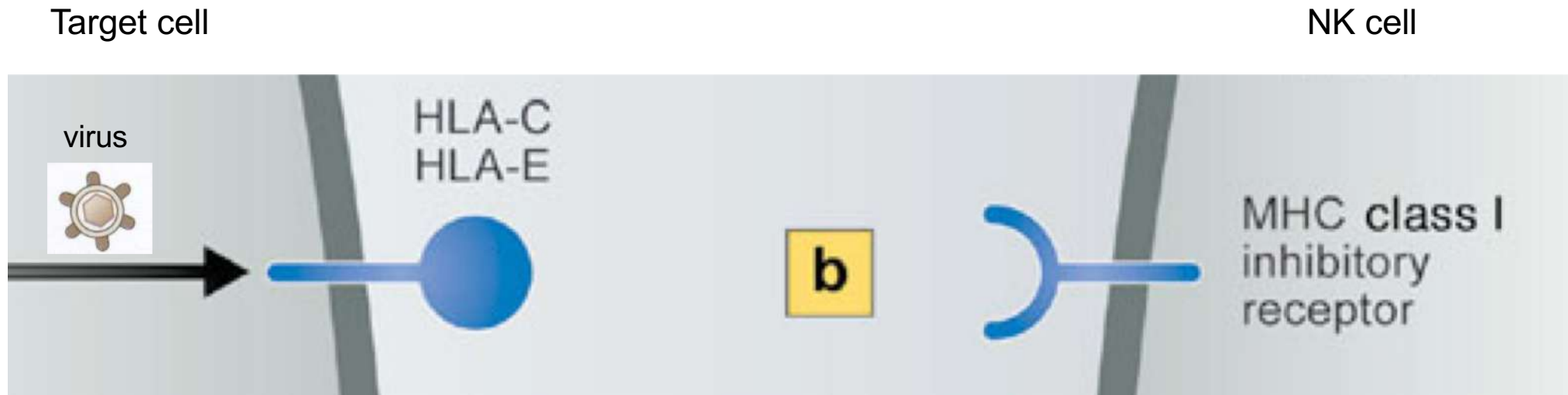
Inibizione delle cellule NK per espressione di “viral MHC I decoys” che agiscono come ligandi dei recettori inibitori



UL18 di HCMV condivide un'omologia del 25% con le molecole HLA di classe I e lega peptidi (anche la signal sequence di UL40) e $\beta 2m$. La sua azione inibitoria dipende dal legame ad alta affinità con il recettore inibitorio ILT2 (o LIR-1) espresso su alcuni subset NK ma anche su cellule B, macrofagi e cellule dendritiche. US2 e US11 non agiscono su IL18.



Modulazione selettiva delle molecole MHC di classe I

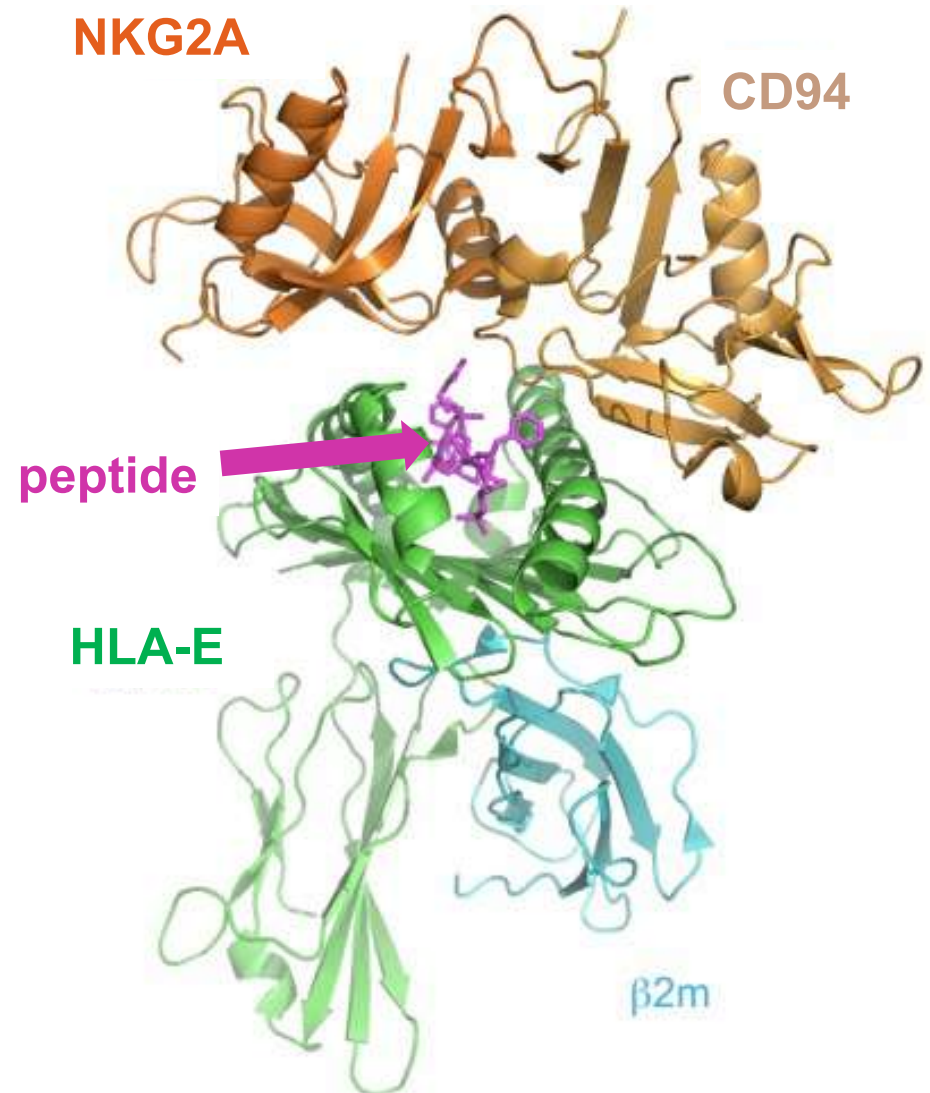


I virus tendono generalmente a down-modulare l'espressione delle molecole HLA-A e B importanti per il riconoscimento da parte delle cellule T CD8+ citotossiche ma non quella delle molecole HLA-E e -C, ligandi per i recettori inibitori delle NK

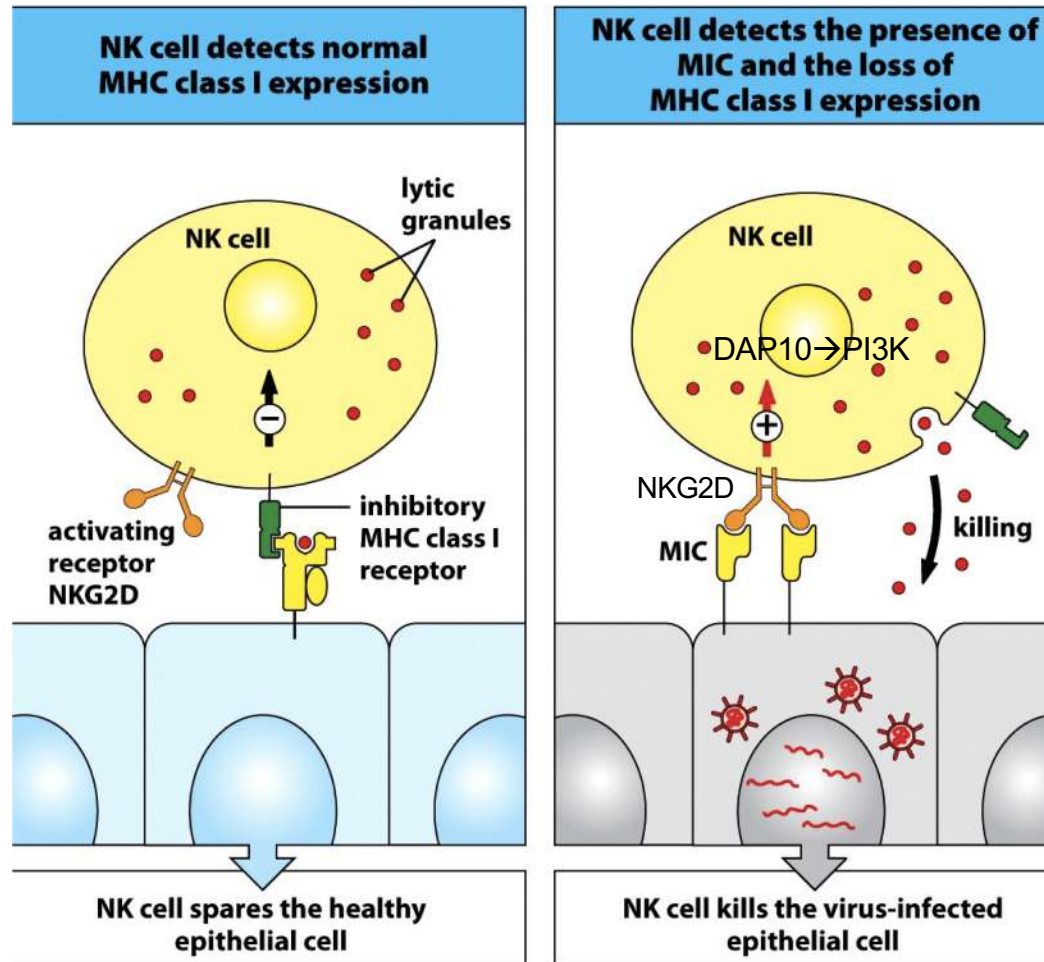
La proteina UL40 prodotta da HCMV fornisce un peptide “mimetico” che stabilizza l’espressione della molecola HLA-E (ligando del recettore inibitorio NKG2A/CD94)

HCMV produce la molecola **UL40** che stabilizza e mantiene elevata l’espressione dell’HLA-E che è il ligando del recettore inibitorio NKG2A-CD94

Meccanismo: HLA-E lega la “sequenza segnale” (o peptide leader) derivante dalle molecole HLA di classe I che però sono down-modulate dal virus. Quindi, la strategia di **HCMV** consiste nella produzione della **proteina UL40** dalla quale deriva una sequenza segnale ottimale per il legame e la stabilizzazione delle molecole HLA-E.

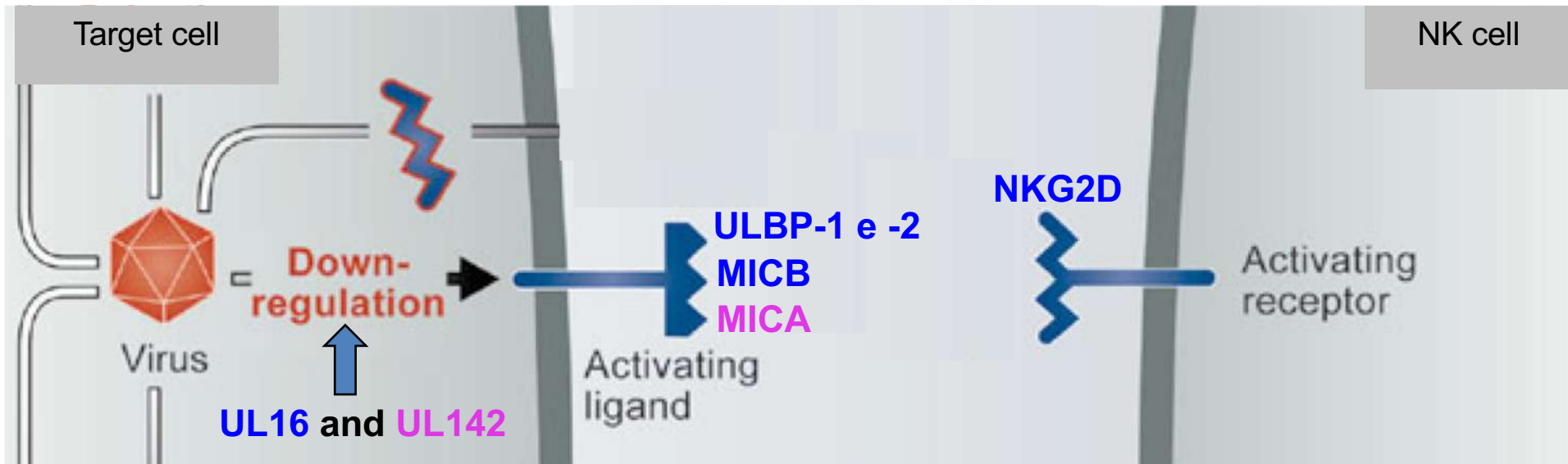


Il legame di MICA, MICB e ULBP 1-6 al recettore attivatorio NKG2D induce attivazione delle cellule NK



I ligandi per NKG2D sono espressi dalle cellule come risposta alle **infezioni virali**, stress cellulare, metabolico, danno al DNA o trasformazione neoplastica

Inibizione dei segnali di attivazione delle NK

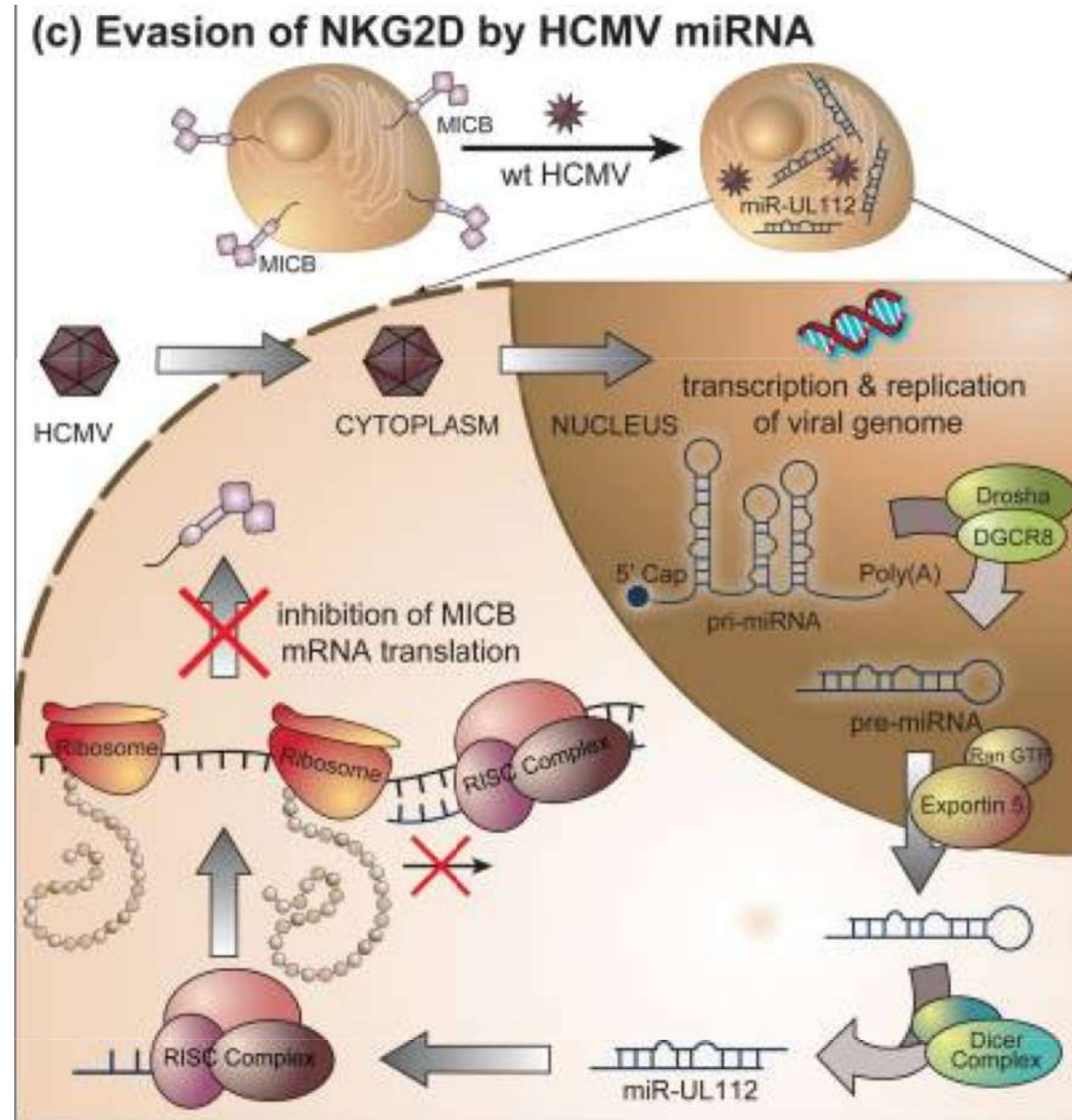


Interferenza con l'espressione dei ligandi attivatori

- HCMV produce **UL16** che modula negativamente l'espressione di MICB, ULBP-1 e -2 e poi **UL142** che modula negativamente l'espressione di MICA, tutti ligandi del recettore attivatorio NKG2D
- HCMV produce miR-UL112 che blocca la trascrizione del mRNA per MICB

MICB è un ligando del recettore attivatorio NKG2D

HCMV produce il miR UL112 che ha come target il trascritto di MICB inibendone la traduzione



After virus enters the cell, its genome is transcribed into mRNA but also noncoding RNA molecules. By using the cellular machinery, HCMV expresses its own miRNAs, **including miR-UL112**, which forms the RISC complex and binds the 3- UTR region of MICB. In this way, miR-UL112 inhibits the translation of MICB mRNA, thus preventing it in reaching the cell surface and signaling.

HCMV inibisce l'espressione di ULBP-1, -2, -6, MICA e MICB (ligandi del recettore attivatorio NKG2D)

UL16 (HCMV) induce ritenzione intracellulare di **ULBP-1, -2, -6 e MICB**

UL142 (HCMV) invece down-modula **MICA** (nella sua versione full-length) ma non la variante allelica tronca (ovvero **MICA-t** o MICA*008)

