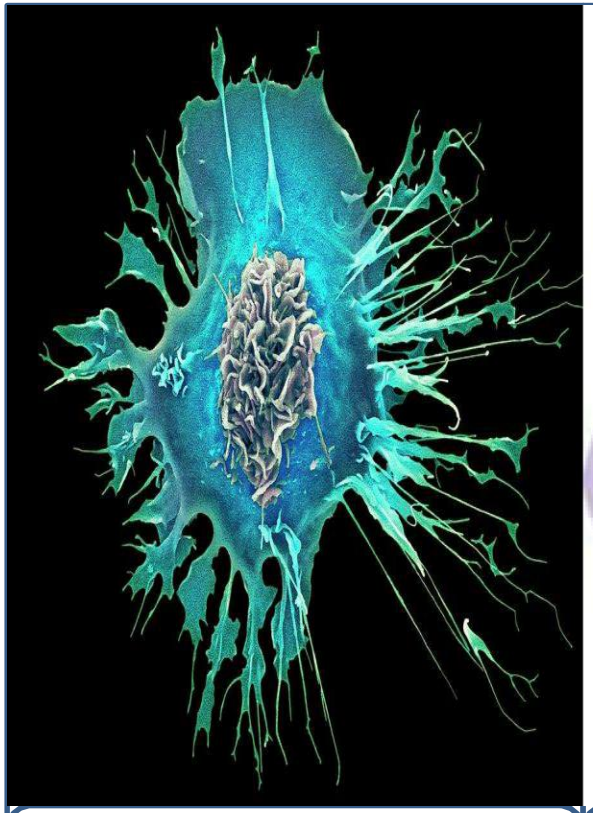
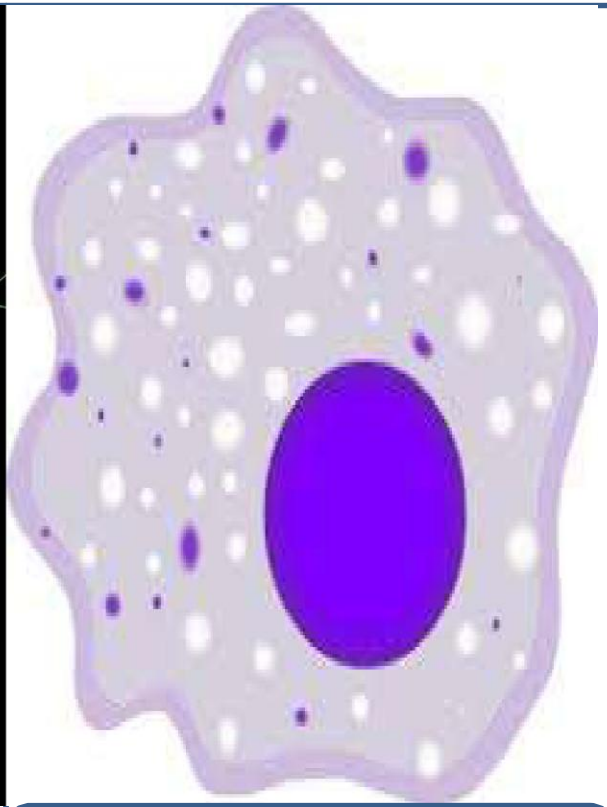
Antigen Presenting Cells



Dendritic cells



Macrophages

B cells

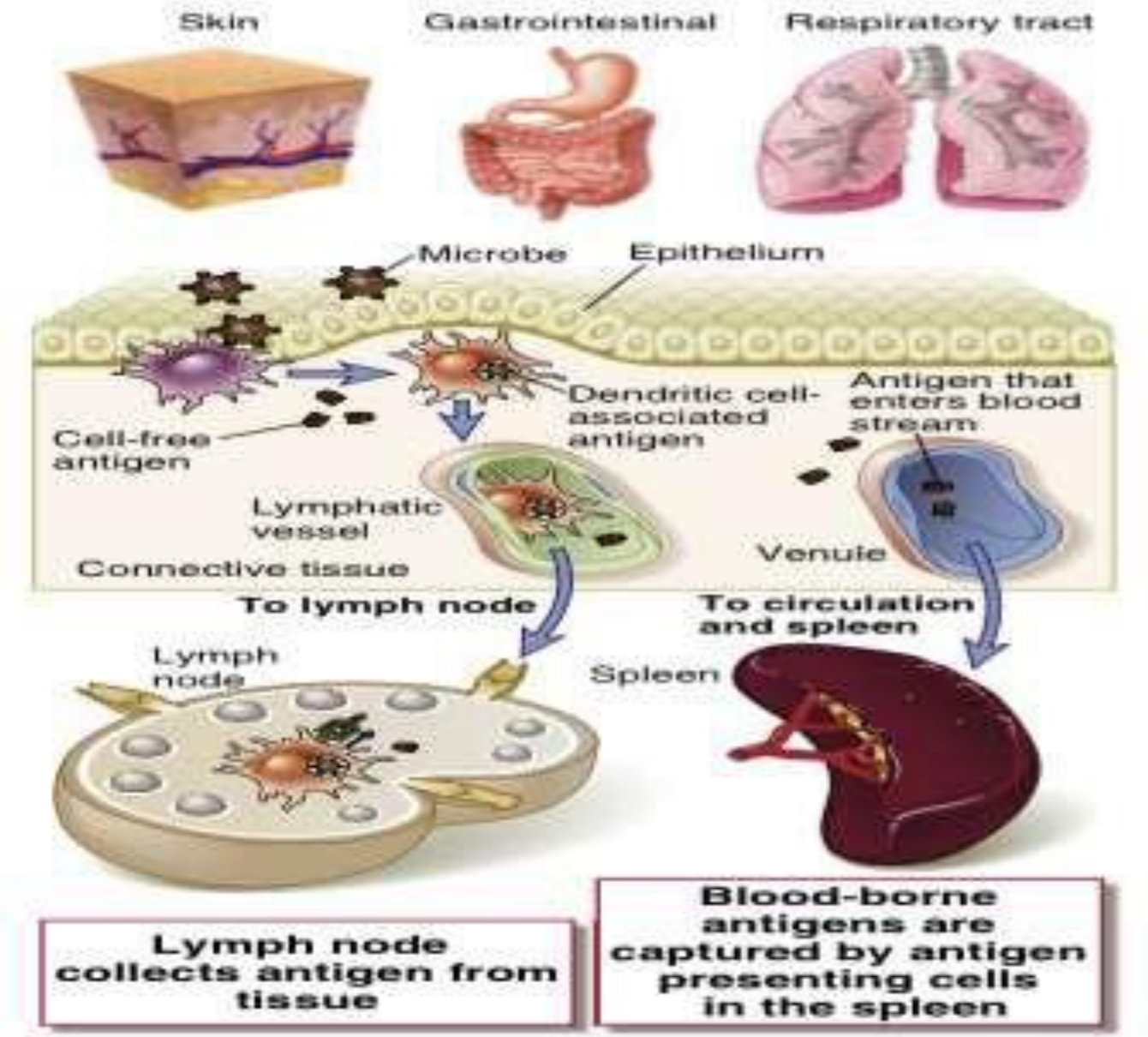
Antigen presenting cells(APCs)

- Can process a protein antigen, break it into peptides and present it in coconjunction with class **II MHC** molecules on the cell surface where it will interact with appropriate T cell receptors..
- Engulf a pathogen through phagocytosis and present it to the whole immune system.
- So that Cell Mediated and Humoral Immune response can build up.

TABLE 8-1 Antigen-presenting cells

Professional antigen-presenting cells	Nonprofessional antigen-presenting cells	
Dendritic cells (several types)	Fibroblasts (skin)	Thymic epithelial cells
Macrophages	Glial cells (brain)	Thyroid epithelial cells
B cells	Pancreatic beta cells	Vascular endothelial cells

1- Dendritic Cells



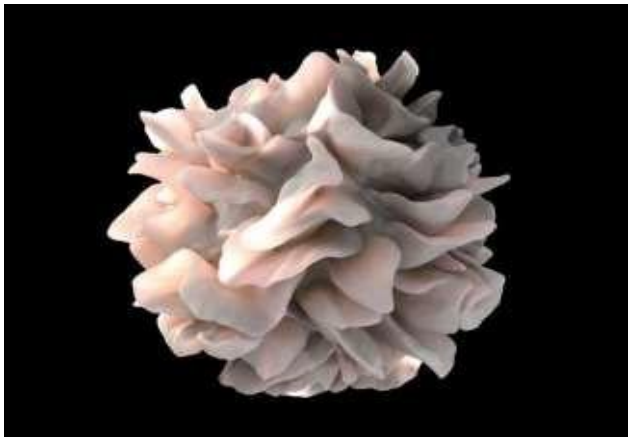
Dendritic cells in antigen presentation

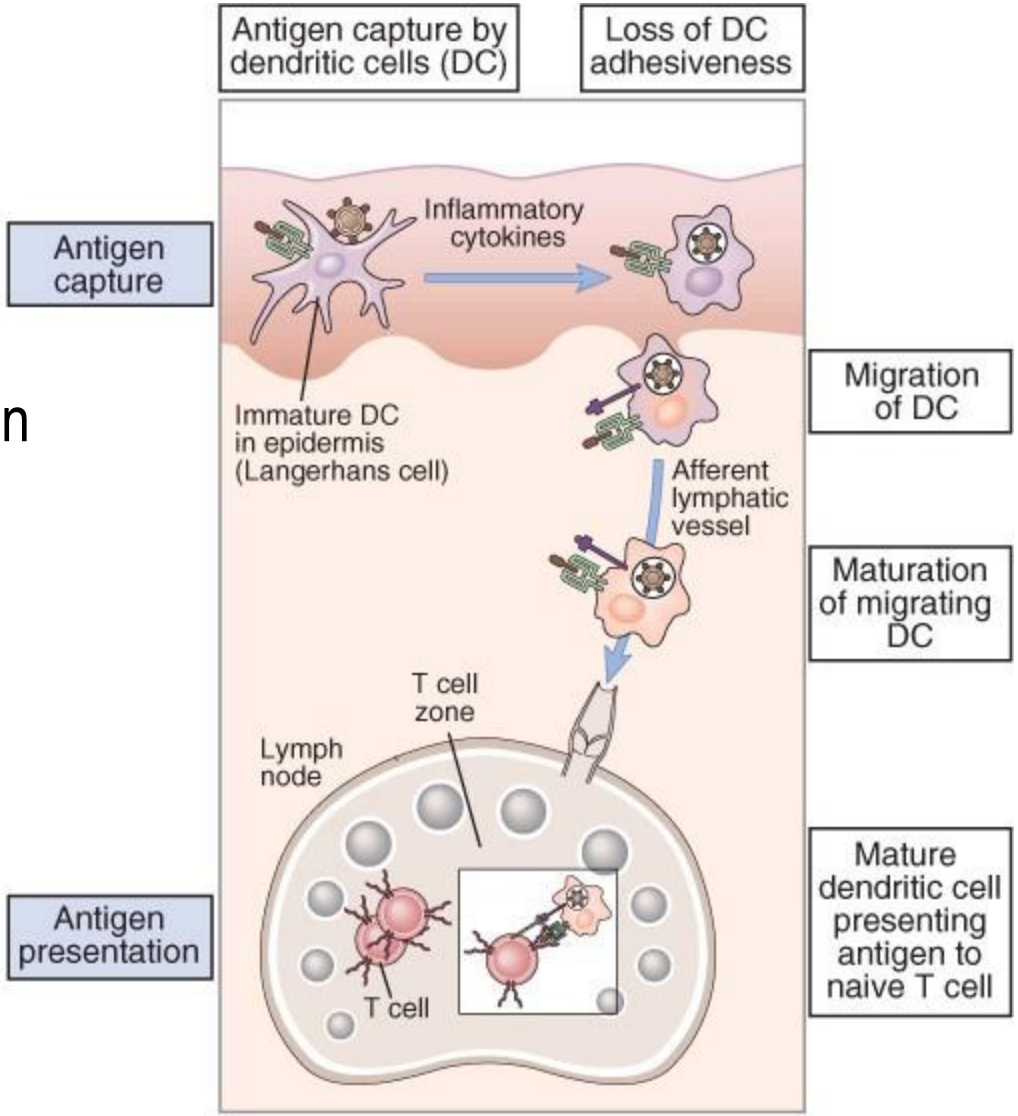
Dendritic cells

- most effective population in T cell activation
- used as immunotherapeutic tools in cancer vaccines

Immature DC: capture antigens in periphery

Mature DC: activation of T lymphocytes in lymphatic nodes





Sets of Dendritic Cells

1-Classical DC	• Most numerous subset of dendritic cells in the lymphoid organs • Mostly derived from myeloid precursors

	• Constantly sample the environment
	• May also present self antigens for regulation/self-tolerance.
	• Upon encountering microbes/cytokines:
	  • Upregulate costimulatory molecules
	  • Produce inflammatory cytokines
	  • Migrate from peripheral tissue to draining lymph node

Plasmacytoid DC	• Resemble plasma cells • Develop in Bone Marrow from same precursor as Classical DC. • Found in blood and in small numbers in lymphoid organs
	• Poorly phagocytic and do NOT sample environmental antigens • **Major function**: Secretion of **Type I IFN** in response to viral infections • May also differentiate into cells similar to Classical DC and **present antigen to Virus-specific T-cells**

Membrane Receptors (C-type lectins)

Capture and Endocytose microbes or microbial products

Process ingested proteins into peptides capable of binding to MHC

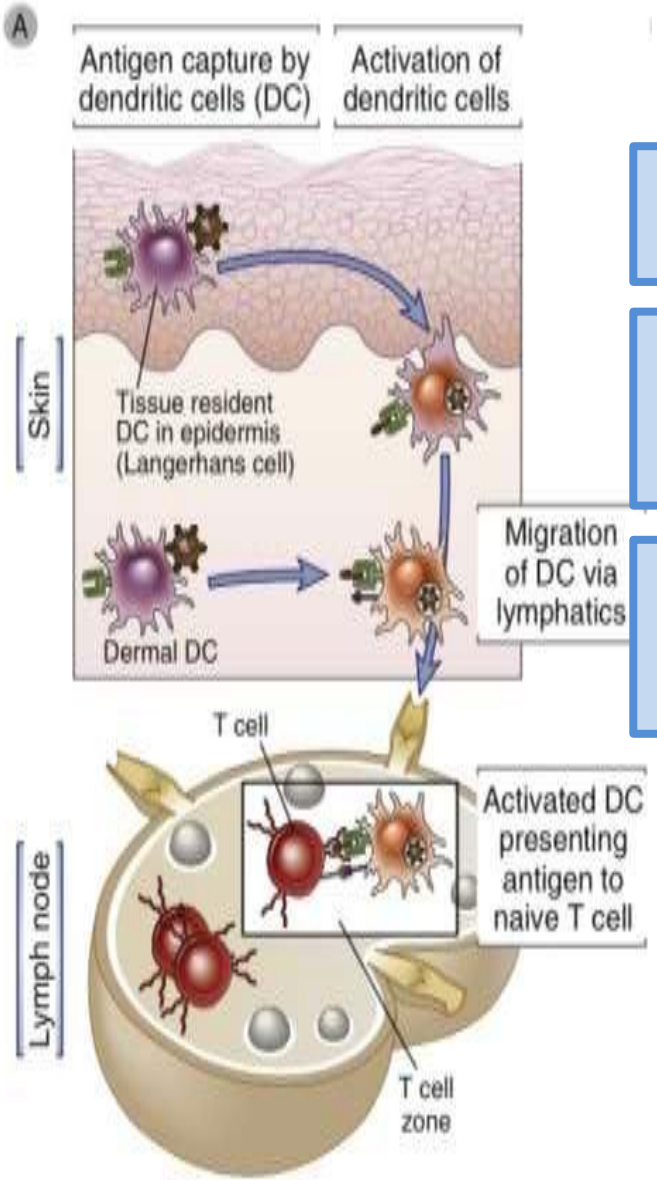
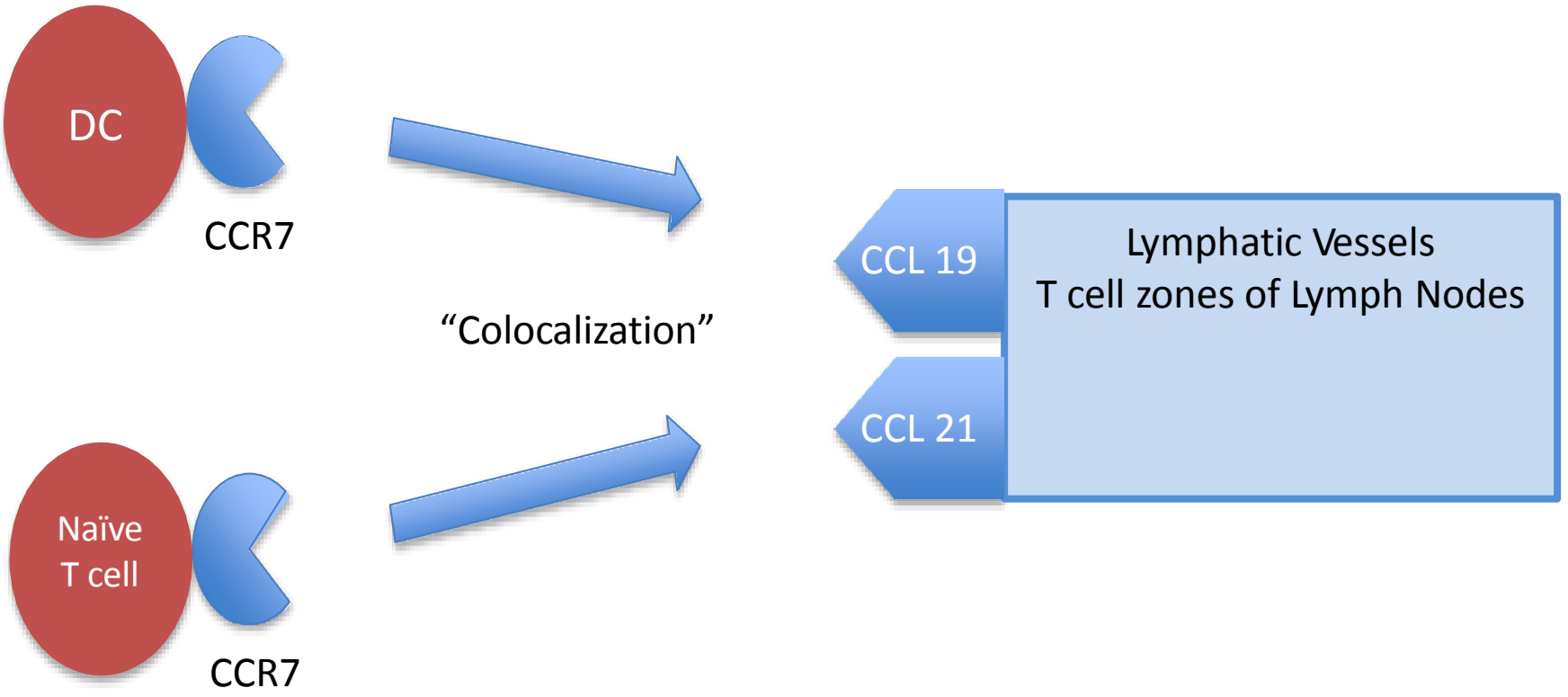


FIGURE 6-5 Role of dendritic cells in antigen capture and presentati

Microbial products recognized by TLR

Signals and Cytokines activate DC (TNF)

Activated DC lose adhesiveness and migrate to lymph nodes


DC
CCR7
Naïve
T cell
CCR7
"Colocalization"
CCL 19
CCL 21
Lymphatic Vessels
T cell zones of Lymph Nodes

B

	Tissue resident dendritic cell	Activated dendritic cell
Principal function	Antigen capture	Antigen presentation to T cells
Expression of Fc receptors, mannose receptors	++	–
Expression of molecules involved in T cell activation: B7, ICAM-1, IL-12	– or low	++
Class II MHC molecules		
Half-life	~10 hr	>100 hr
Number of surface molecules	~10^6	~7×10^6

• Properties that make DC the most efficient APCs for initiating T cell responses

1. Strategically located at common sites of entry
2. Express receptors that enable capture and response
3. Migrate from epithelia and tissues via lymphatics to T cell zones of LN
4. Mature DC express high levels of peptide-MHC complexes, costimulators, and cytokines

• Dendritic cells can also ingest infected cells and present antigens to CD8+ T lymphocytes

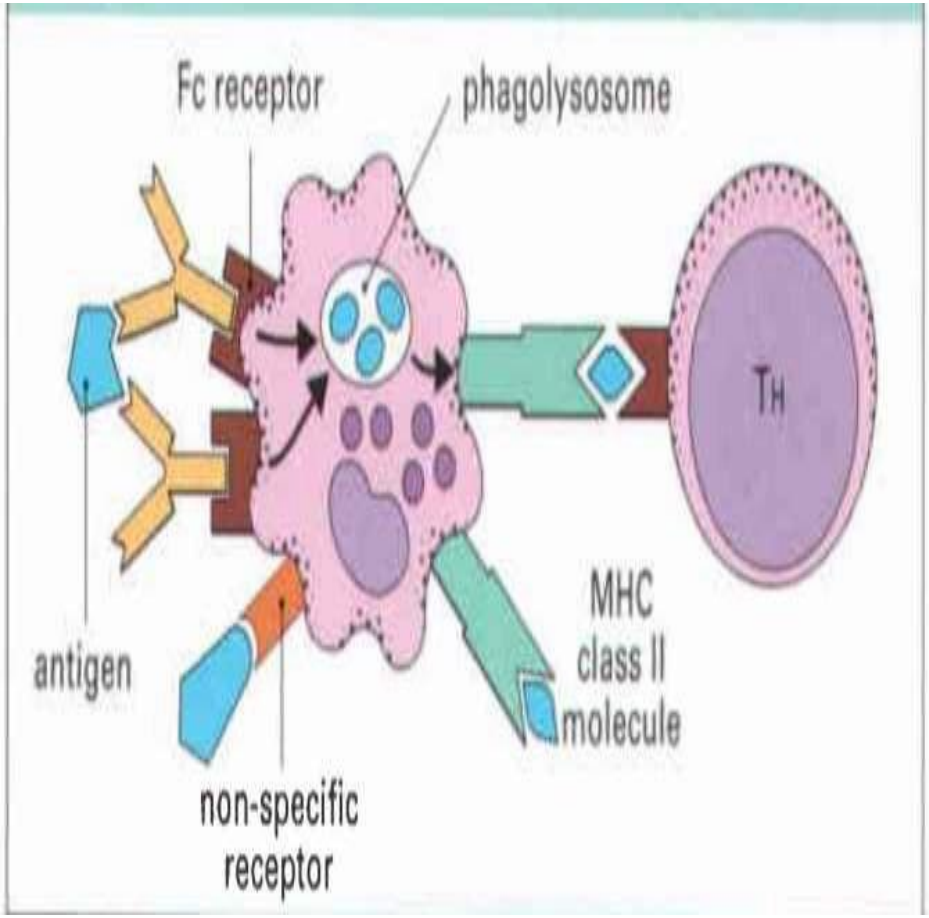
- Peptide antigens must be derived from proteins in the cytosol of DC
- Specialized DC: able to ingest virus-infected cells and deliver viral proteins into their cytosol
- **"CROSS-PRESENTATION or CROSS-PRIMING"**

So, DC act as messenger between innate and adaptive immune systems

2-Macrophages:

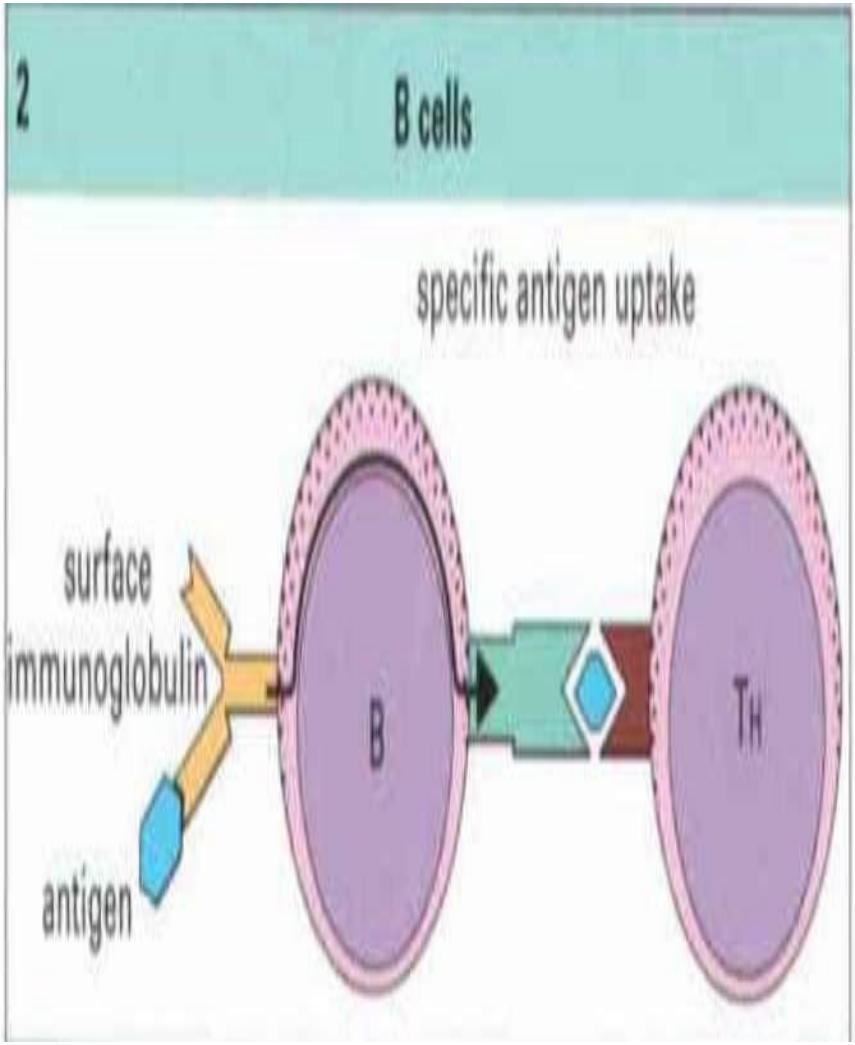
Macrophages are antigen presenting cells that recognize, engulf and destroy target cells.

- After the activation, macrophages are able to express MHC class II and co-stimulatory molecules , including the B7 complex and can present phagocytosed peptide fragments to helper T cells.



3-B Cells:

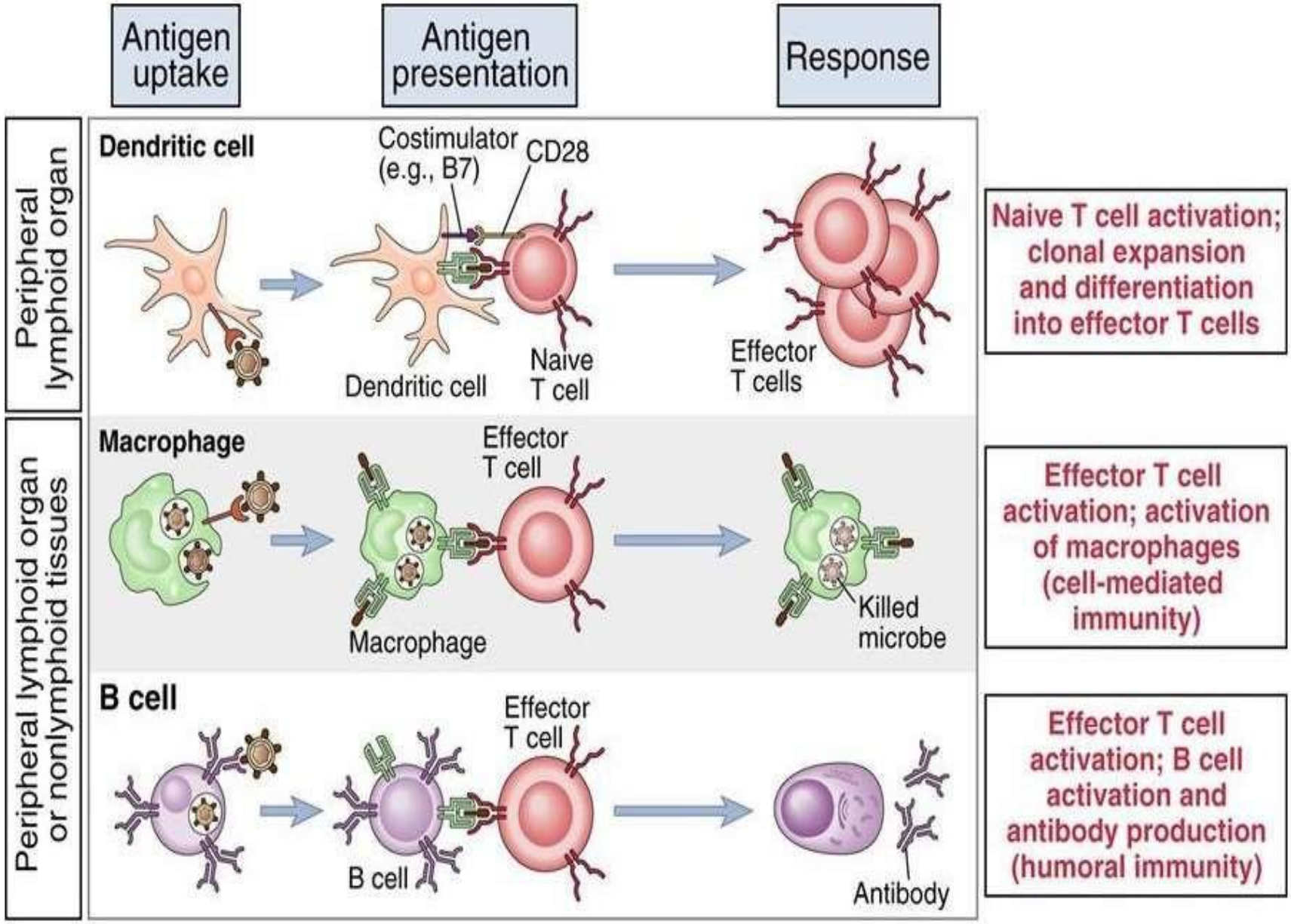
B cells , also known as B lymphocytes, are a type of white blood cell of small lymphocyte subtype.



B cells can

- Internalize antigen;
- Recognize soluble antigen; then process the antigen and present peptides by using MHC class II molecules

Function of different Antigen Presenting cells



Comparative characteristics of some APCs

	Dendritic cell	Macrophage	B lymphocyte
Antigen uptake	Endocytosis Phagocytosis	phagocytosis	Receptor- mediated endocytosis
Class II MHC expression	Constitutive (+++)	Inducible (++)	Constitutive (+++)
Co-stimulatory activity	Constitutive B 7 (+++)	Inducible B 7 (++)	Inducible B 7 (++)
T-cell activation	Native T cells Effector T cells Memory T cell	Effector T cell Memory T cell	Native T cell Effector T cell Memory T cell

Cell type	Expression of		Principal function
	Class II MHC	Costimulators	
Dendritic cells	Constitutive; increases with maturation; increased by IFN-γ	Constitutive; increases with maturation; increased by TLR ligands, IFN-γ, and T cells (CD40-CD40L interactions)	Antigen presentation to naive T cells in the initiation of T cell responses to protein antigens (priming)
Macrophages	Low or negative; inducible by IFN-γ	Low, inducible by TLR ligands, IFN-γ, and T cells (CD40-CD40L interactions)	Antigen presentation to $CD4^+$ effector T cells in the effector phase of cell-mediated immune responses
B lymphocytes	Constitutive; increased by cytokines (e.g., IL-4)	Induced by T cells (CD40-CD40L interactions), antigen receptor cross-linking	Antigen presentation to $CD4^+$ helper T cells in humoral immune responses (T cell–B cell interactions)

Antigen processing & presentation

- Foreign protein antigen are degraded into small antigenic peptides that form complexes with class I or class II MHC molecule..
- This conversion of proteins into MHC associated peptide fragments is called

”Antigen Processing And Presentation”

Antigen processing and presentation

- The process by which pathogens and their products are degraded to produce peptide antigens is known as Antigen Processing

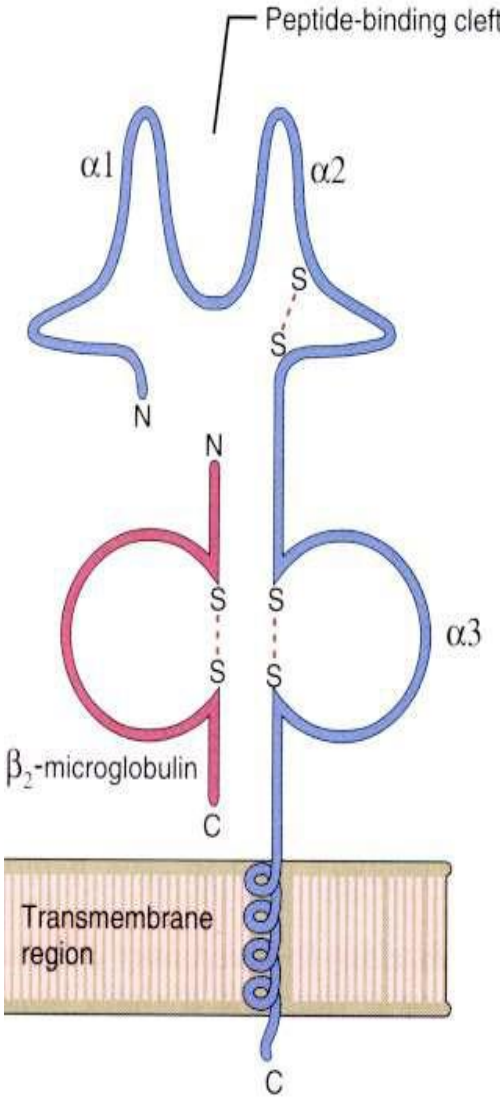
- These peptide fragments combine with MHC molecules inside cell

- The MHC-peptide complex thus formed travels to the cell surface where it displays peptide fragments to T cells. This is known as Antigen Presentation.

MHC molecules

- Major Histocompatibility Complex
  --Cluster of genes found in all mammals
  --Its products play role in discriminating self/non-self
  --Participate in both humoral and cell-mediated immunity
  --Act as antigen presenting structures
- Polymorphic)genetically diverse) glycoproteins
- Alleles are co-dominantly expressed
- In Humans -Chromosome & 6referred to as HLA complex
- In Mice -Chromosome & 17referred to as H-2 complex

MHC class I molecules



-Class I MHC is found in almost all nucleated cell.

-Present endogenous antigen in the cell cytosol

-Structure of MHC class I molecules

MHC class I encoded

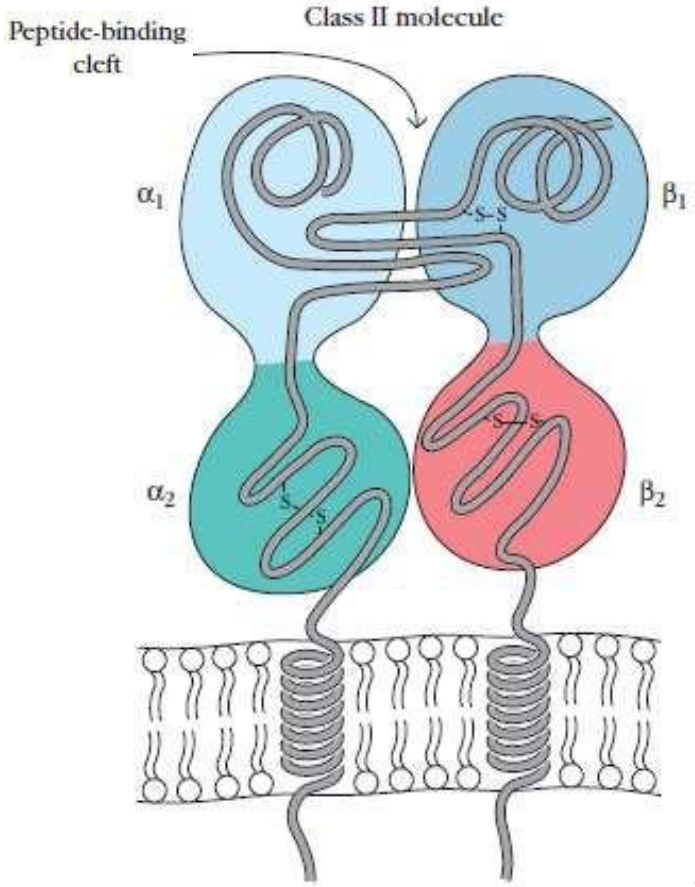
A]α-chain of 43kDa

α Chain Made Up Of 3 Domains (α1, α2 and α3)

- α1 And α2 Form Peptide Binding Cleft that fits peptide of about 8-10 a/a long
- α3-chain anchored to the cell membrane and it serves as a binding site to CD8 molecule of T_{Cyt} cell

B]β2-microglobulin, 12kDa, non-MHC encoded, non-transmembrane, non covalently bound to α3-chain

MHC class II molecules



- Found in APCs,
- presents exogenous antigen.
- Structure of MHC class II molecules

Class II MHC-encoded α-chain of 34kDa & β-chain of 29kDa

-α and β chains anchored to the cell membrane

-α chain and β chain associate non-covalently

-α and β chains Made Up Of Domains

- α1 and α2 (α chain)
- β1 and β2 (β chain)

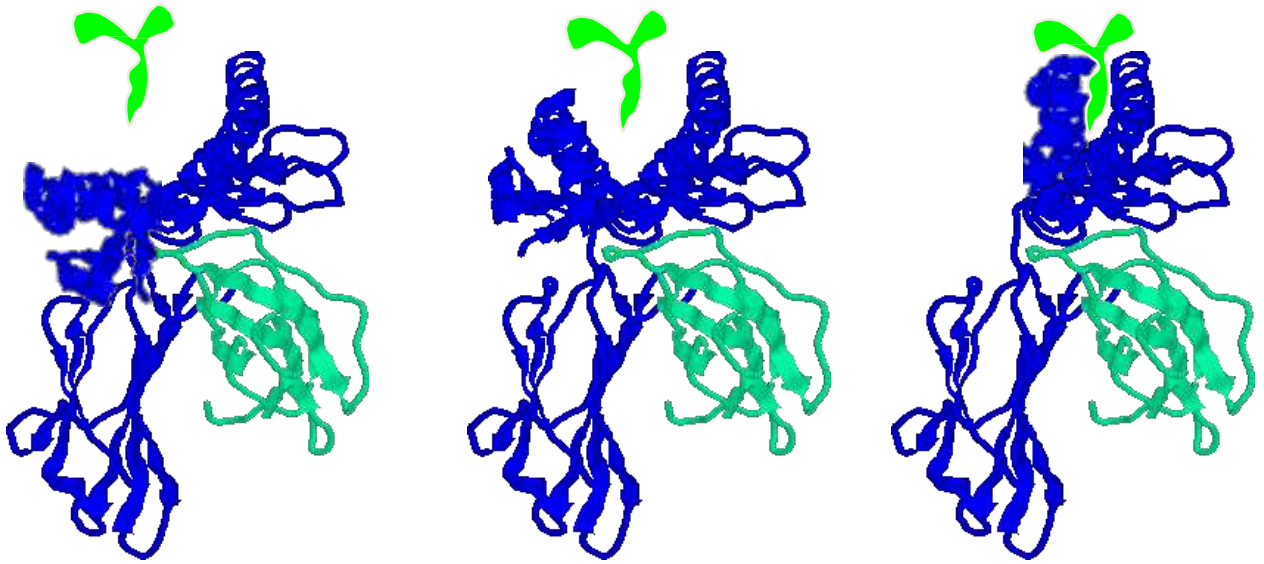
α1and β1 Form Antigen Binding Cleft

MHC class II accommodate peptides of >13 amino acids

CD4 Molecule Binds α2/β2 domains

A flexible binding site?

A binding site that is flexible at an early, intracellular stage of maturation Formed by folding the MHC molecules around the peptide.



Floppy → Compact

Allows a single type of MHC molecule to

- bind many different peptides
- bind peptides with high affinity
- form stable complexes at the cell surface
- Export only molecules that have captured a peptide to the cell surface

Anchor residue: The peptide residue that anchor the antigenic peptide into the MHC groove.

MHC I	MHC II
Composed of an α)or heavy) chain in a non-covalent complex with a β2-microglobulin	Contain two MHC-encoded polymorphic chains, an α chain and a β chain.
Recognized by CD8+ T cells	Recognized by CD4+ T cells
Accommodate peptides that are 6to 16amino acid residues in length	Allows larger peptides)up to 30amino acid residues in length or more) to bind
Expressed on all nucleated cells	Expressed mainly on specialized APCs
Cytosolic proteins are proteolytically degraded in the proteasome	Extracellular proteins are internalized into endosomes

Genetic Map Of MHC

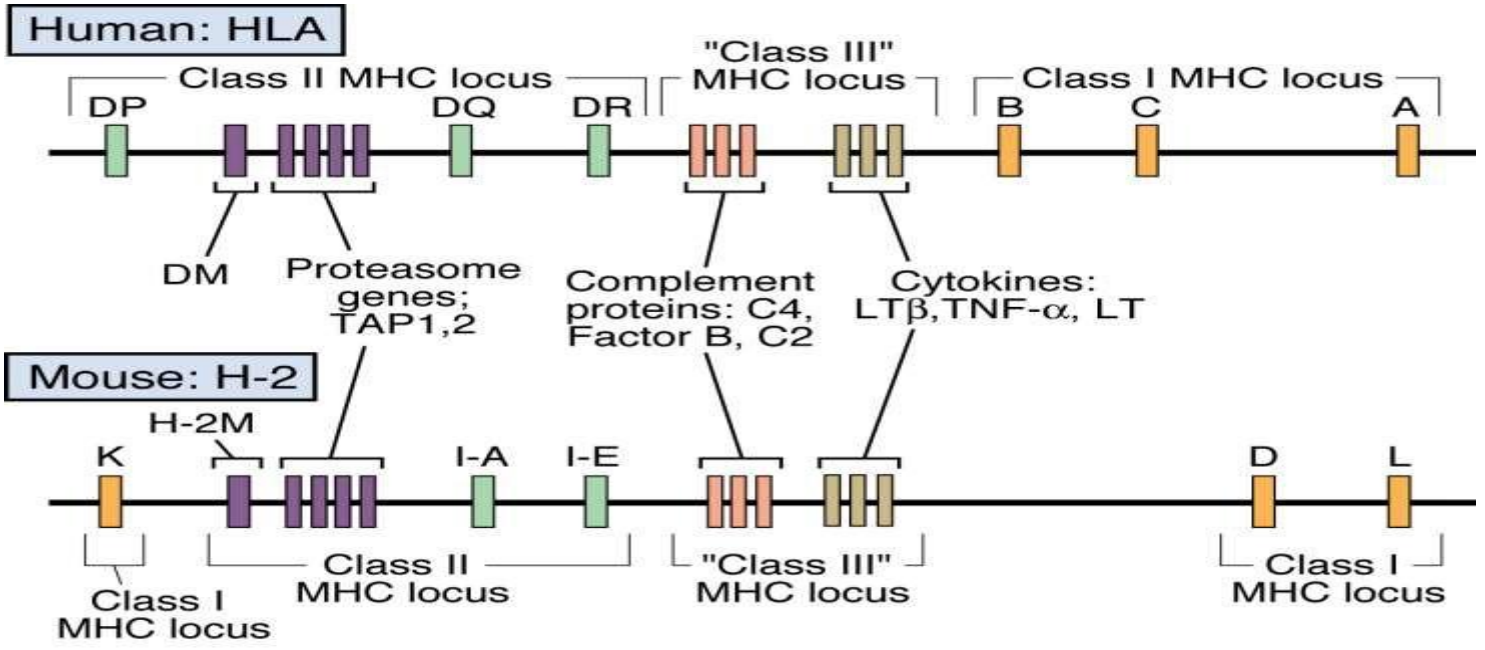
Human Leukocyte Antigen (HLA(

- In humans, there are three MHC Class I α-chain genes, called:

  HLA -A, HLA-B, and HLA-C

- There are also three pairs of MHC Class II α- and β-chain genes, called :**HLA-DR, HLA-DP, and HLA-DQ.**

The Genes Of The MHC Locus



Molecular basis of MHC types and variants

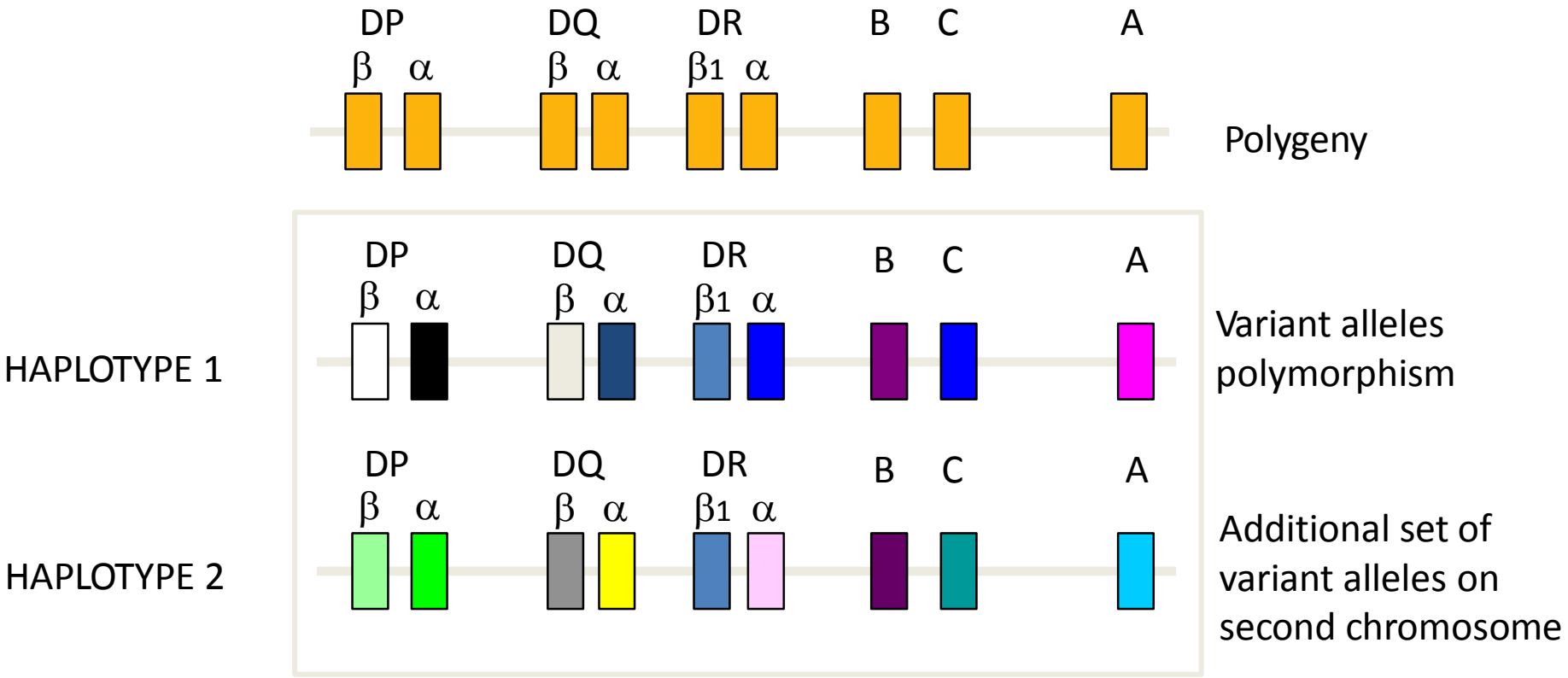
POLYMORPHISM

Variation >1% at a single genetic locus in a population of individuals
MHC genes are the most polymorphic known genes

The type and variant MHC molecules do not vary in the lifetime of the individual

Diversity in MHC molecules exists at the population level
This sharply contrasts diversity in T and B cell antigen receptors which are in a constant state of flux within the individual.

Diversity of MHC molecules in the individual



MHC molecules are CODOMINANTLY expressed
Two of each of the six types of MHC molecule are expressed

Genes in the MHC are tightly LINKED and usually inherited in a unit called an MHC HAPLOTYPE

Populations need to express variants of each type of MHC molecule

- Populations of microorganisms reproduce faster than humans
- Mutations that change MHC-binding antigens or MHC molecules can only be introduced to populations after reproduction
- The ability of microorganisms to mutate in order to evade MHC molecules will always improve counter evasion measures that involve mutations in the MHC
- The number of types of MHC molecules are limited

To counteract the superior flexibility of pathogens:

Human populations possess many variants of each type of MHC molecule

Variant MHC may not protect every individual from every pathogen.

However, the existence of a large number of variants means that the population is prevented from extinction

MHC Expression

- Expression is increased by cytokines such as IFNα, IFNβ, IFNγ and TNF
- Transcription factors like CIITA)Transactivator), RFX)Transactivator) increase MHC gene expression
- Some viruses)CMV, HBV, Ad12) decrease MHC expression
- Reduction of MHC may allow for immune system evasion

MHC-restricted antigen recognition by T cells

- Any T cell can recognize an antigen on an APC only if that antigen is displayed by MHC molecules
  - Antigen receptors of T cells have dual specificities:

    1.for peptide antigen (responsible for specificity of immune response) and

    2. for MHC molecules (responsible for MHC restriction)
  - During maturation in the thymus, T cells whose antigen receptors see MHC are selected to survive and mature; therefore, mature T cells are "MHC-restricted"

Only fragments of proteins)peptides) associated with MHC molecules on surface of cells

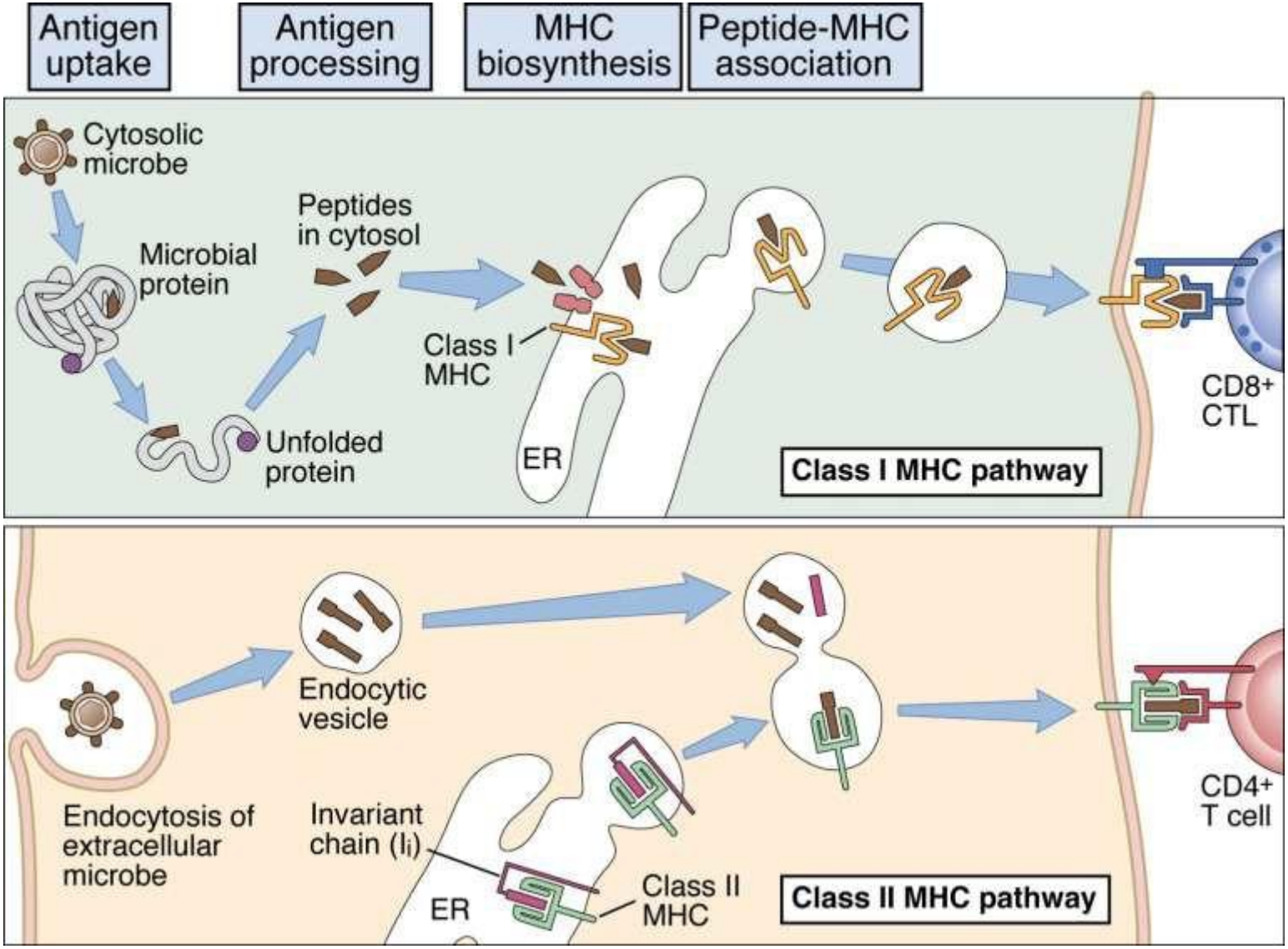
- Helper T cells CD4+(Th) recognize peptide associated with MHC class II molecules
- Cytotoxic T cells CD8+)Tcyt) recognize peptide associated with MHC class I molecules.

Antigen processing

- Conversion of native antigen (large globular protein) into peptides capable of binding to MHC molecules

- Occurs in cellular compartments where MHC molecules are synthesized and assembled (ER)
  - Determines how antigen in different cellular compartments generates peptides that are displayed by class I or class II MHC molecules

Pathways of antigen processing



Protein antigen in cytosol)cytoplasm) <--class I MHC pathway

Protein antigen in vesicles <--class II MHC pathway

Class I MHC molecules
Endogenous Antigens: The Cytosolic Pathway

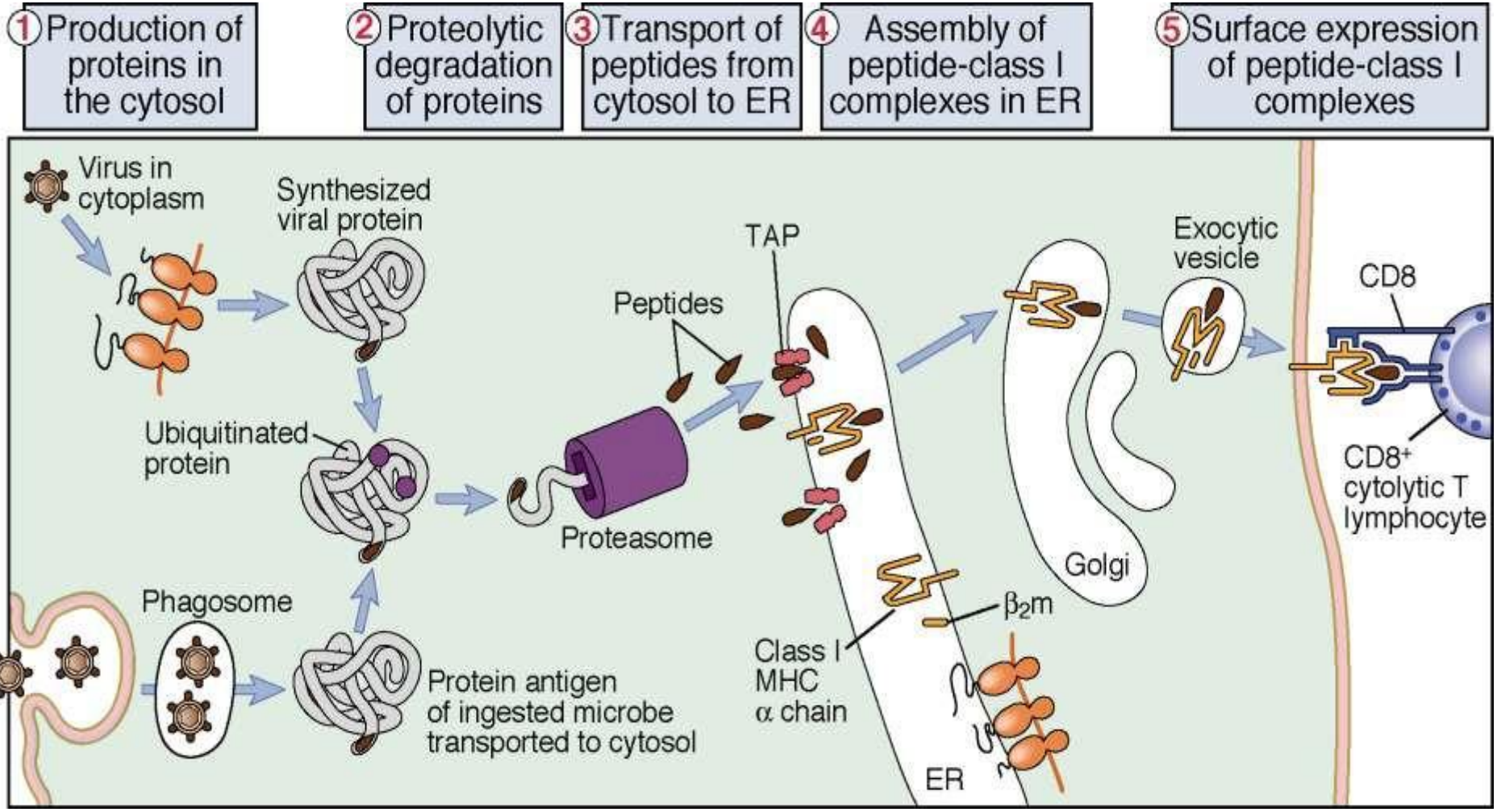
- Cytotoxic T lymphocytes need to kill cells containing cytoplasmic microbes, and tumor cells (which contain tumor antigens in the cytoplasm)

- Cytosolic proteins are processed into peptides that are presented in association with class I molecules

- Most cytosolic peptides are derived from endogenously synthesized (e.g. viral, tumor) proteins

- All nucleated cells (which are capable of being infected by viruses or transformed) express class I

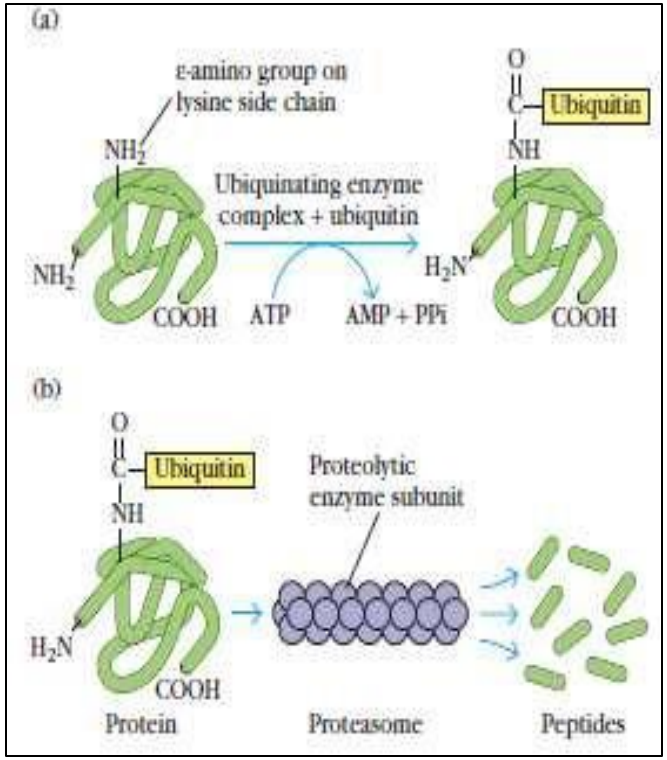
The class I MHC pathway of processing of endogenous cytosolic protein antigens

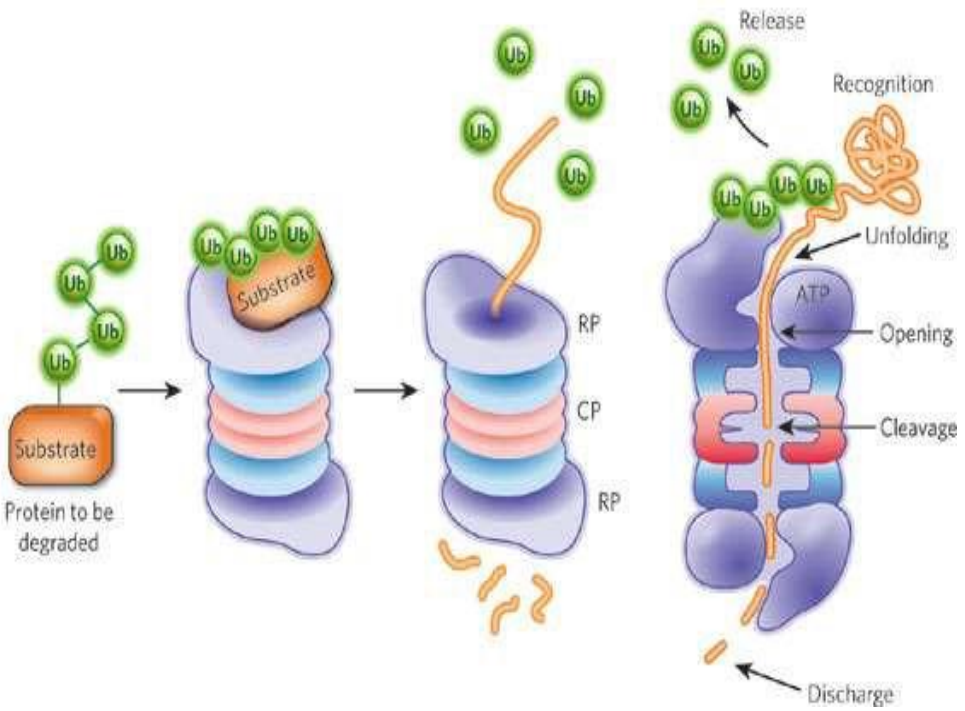


Cytoplasmic peptides are actively transported into the ER; class I MHC molecules are available to bind peptides in the ER

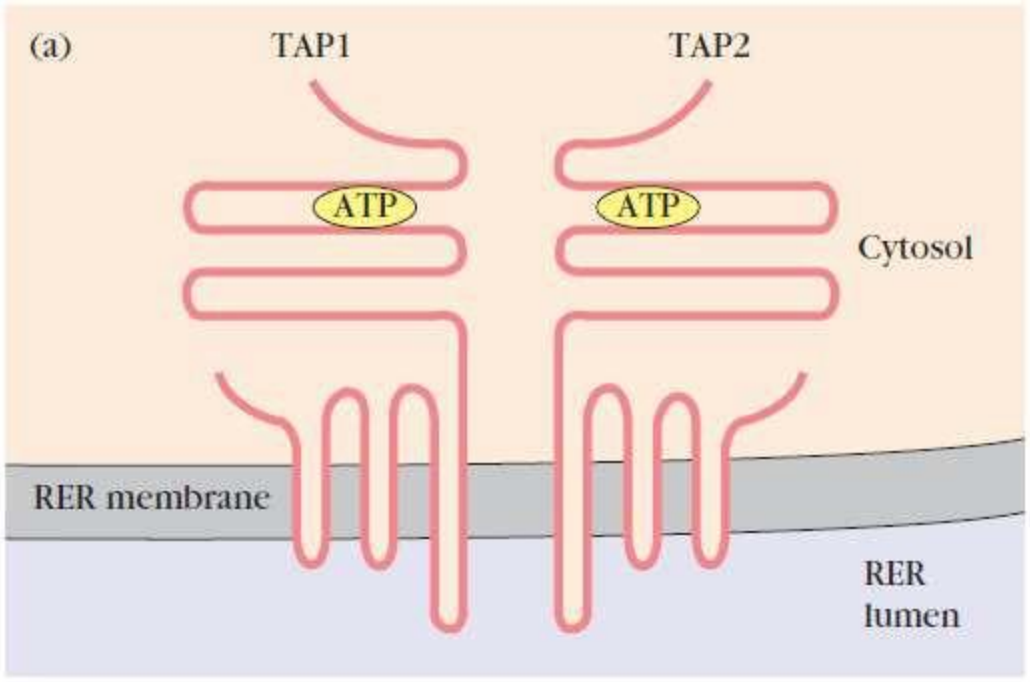
Cytosolic proteolytic system for degradation of intracellular proteins.

- Proteins to be degraded are often covalently linked to a small protein called ubiquitin.
- Degradation of ubiquitin-protein complexes occurs within the central channel of proteasomes, Proteasomes are large cylindrical particles whose subunits catalyze cleavage of peptide bonds.
- In the cytosol,association of LMP2, LMP7, and LMP10 with a proteasome changes its catalytic specificity to favor production of peptides that bind to class I MHC molecules. All three are induced by increased levels of the T-cell cytokine IFN-γ

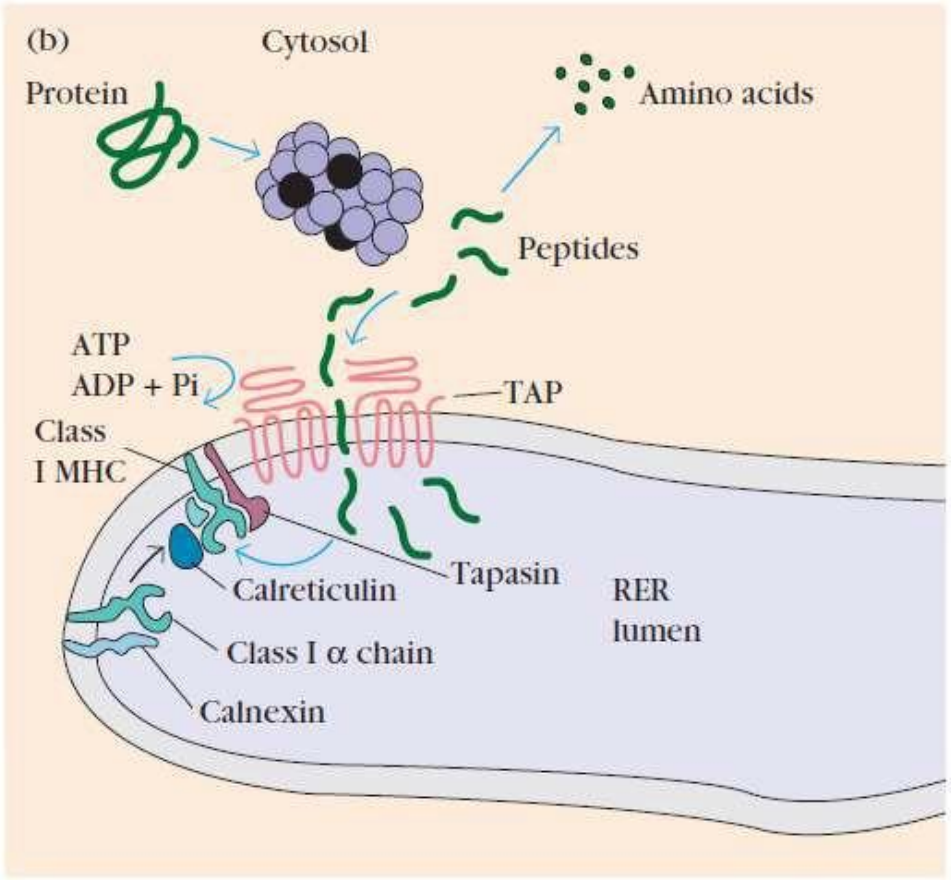




- Peptides generated in the cytosol by the proteasome are translocated by TAP) Transporter Associated With Antigen Processing (into the RER by a process that requires the hydrolysis of ATP.
- TAP has the highest affinity for peptides containing 10–8amino acids, which is the optimal peptide length for class I MHC binding.

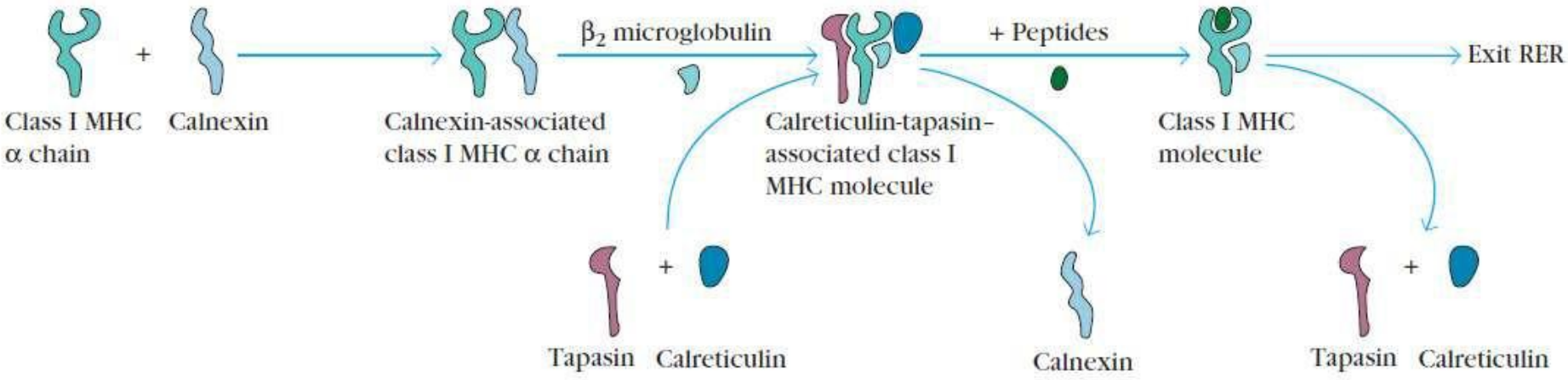


- Within the RER membrane, a newly synthesized class I chain associates with calnexin until β2-microglobulin binds to the chain.
- The class I αchain/β2-microglobulin heterodimer then binds to calreticulin and the TAP-associated protein tapasin.
- When a peptide delivered by TAP is bound to the class I molecule, folding of MHC class I is complete and it is released from the RER and transported through the Golgi to the surface of the cell.



Assembly and stabilization of class I MHC molecules.

- Newly formed class I chains associate with calnexin, a molecular chaperone, in the RER membrane.
- Subsequent binding to β2-microglobulin releases calnexin and allows binding to the chaperone calreticulin and to tapasin, which is associated with the peptide transporter TAP.
- This association promotes binding of an antigenic peptide, which stabilizes the class I molecule–peptide complex, allowing its release from the RER

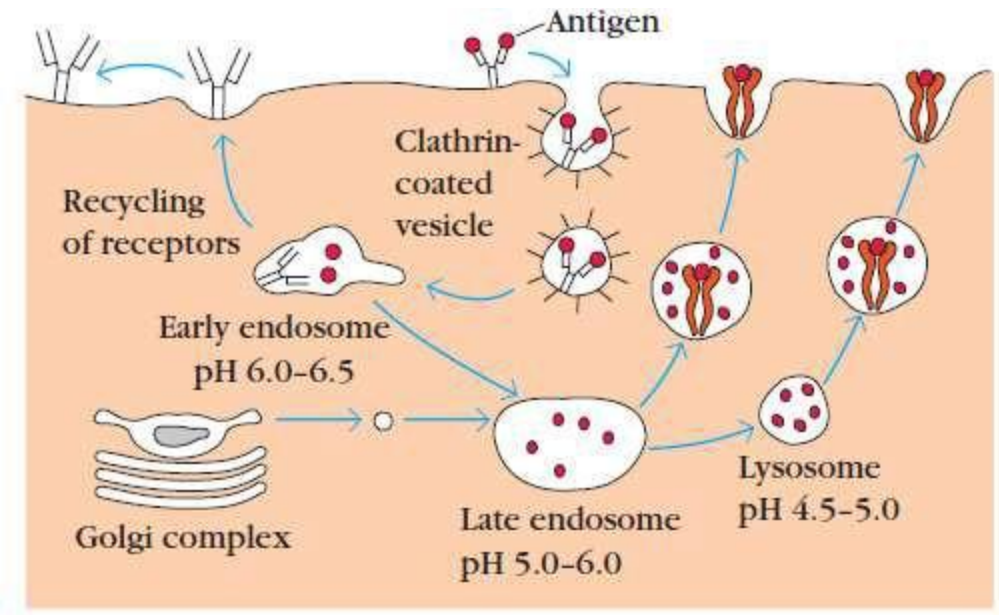


Loading of antigen to MHC class II

Exogenous Antigens: The Endocytic Pathway

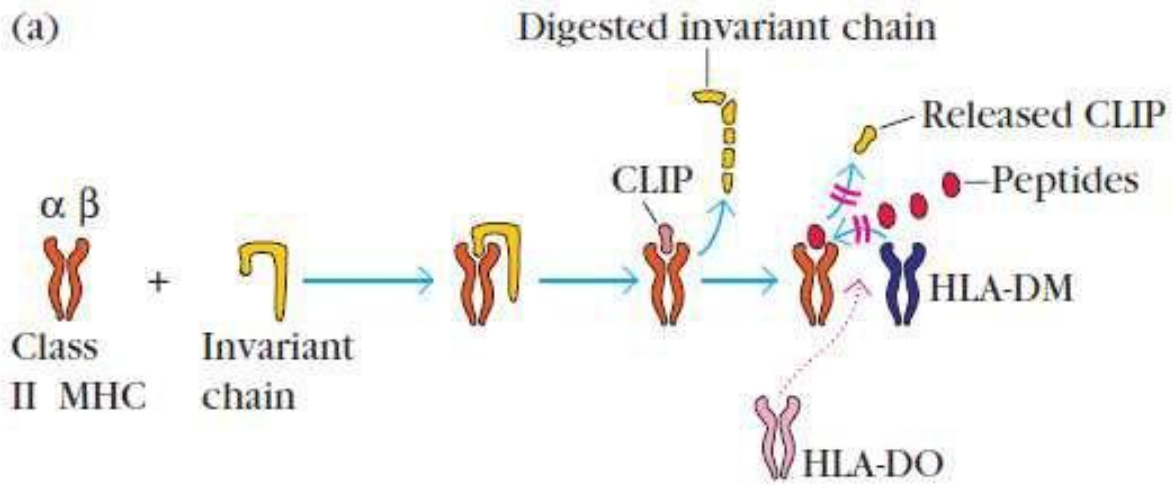
Generation of antigenic peptides in the endocytic processing pathway.

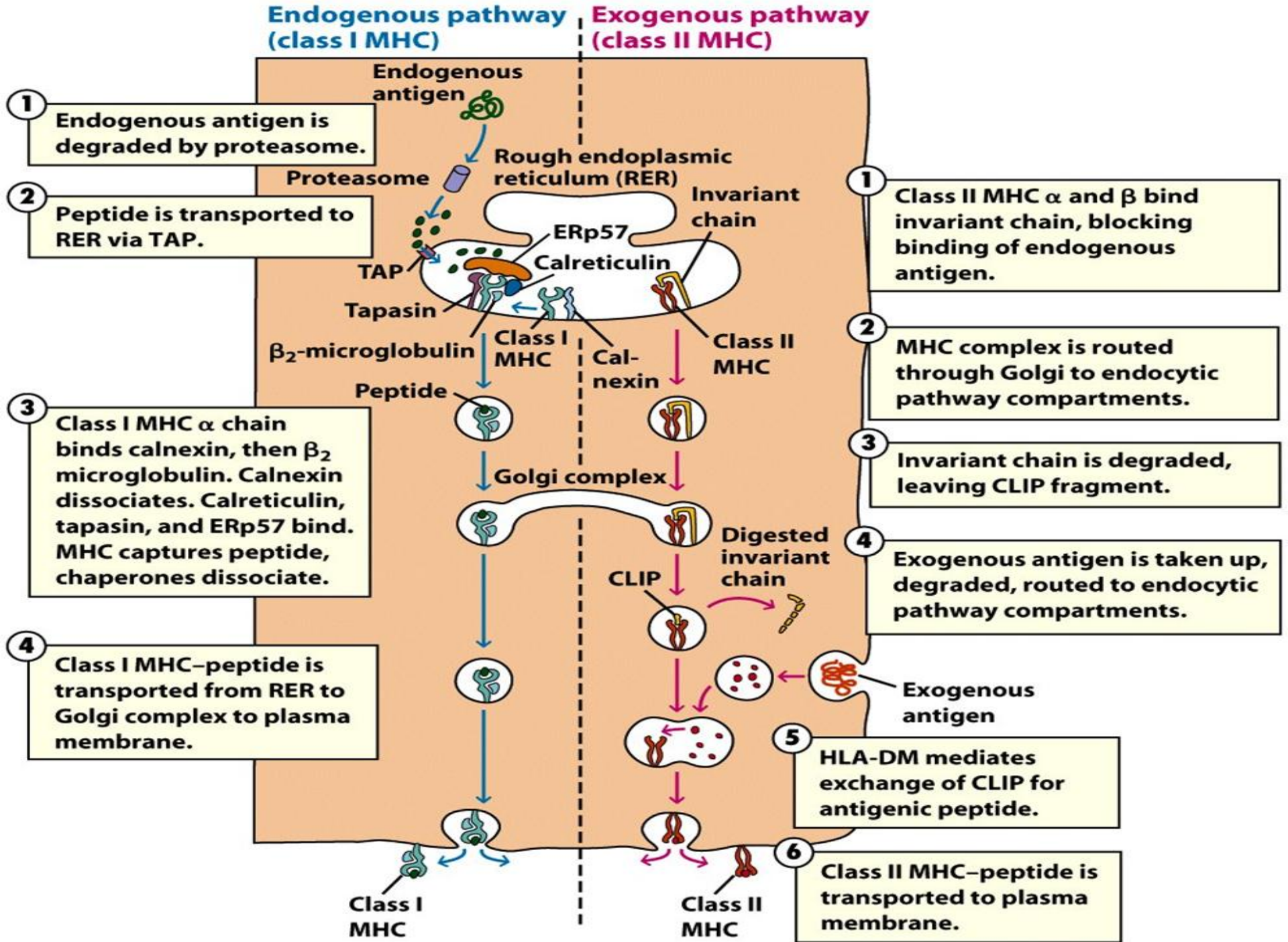
- Internalized exogenous antigen moves through several acidic compartments, in which it is degraded into peptides that ultimately associate with class II MHC molecules transported in vesicles from the Golgi complex.

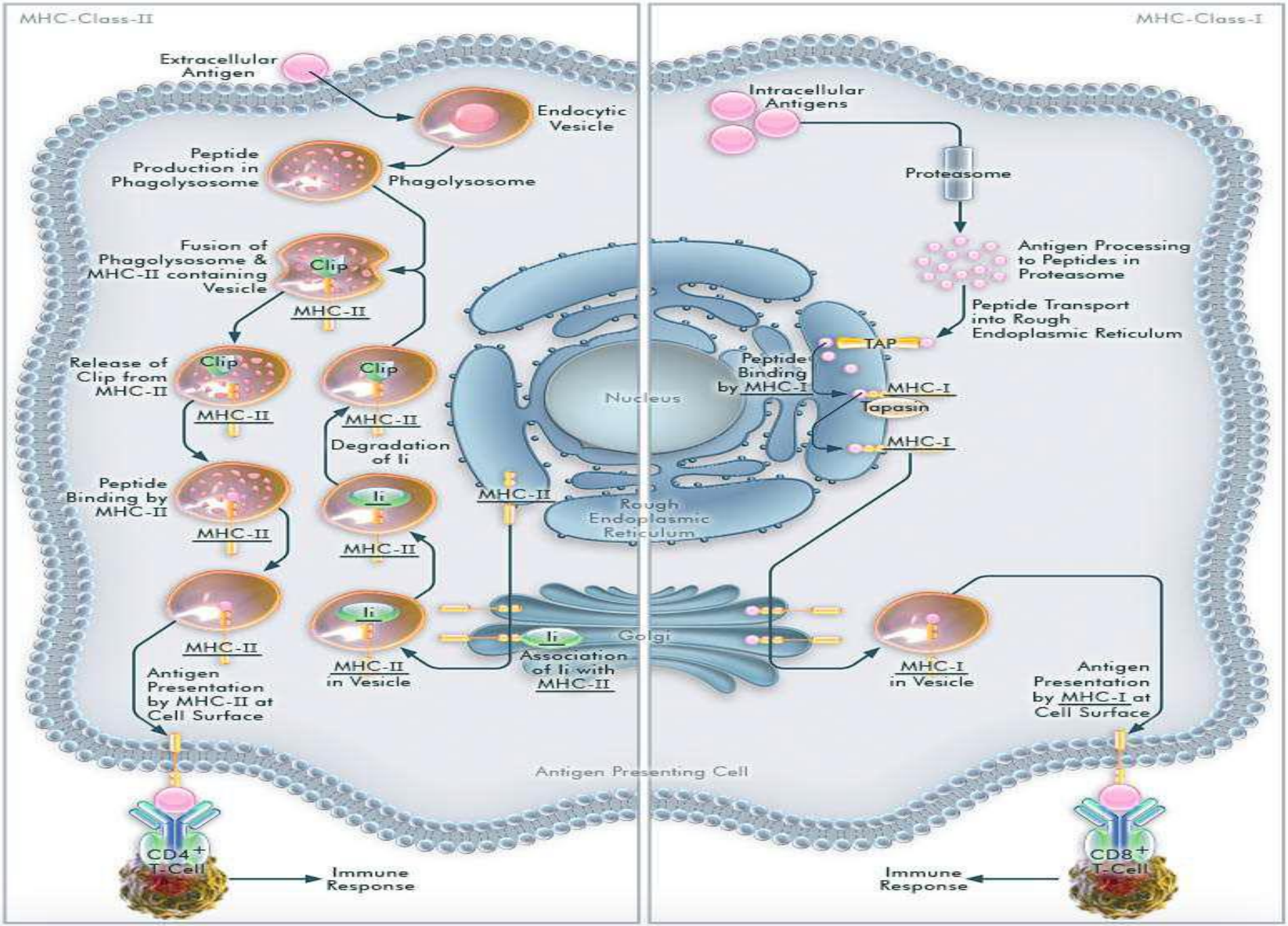


Assembly of class II MHC molecules

- Within the rough endoplasmic reticulum, a newly synthesized class II MHC molecule binds an **invariant chain**.
- The bound invariant chain prevents premature binding of peptides to the class II molecule and helps to direct the complex to endocytic compartments containing peptides derived from exogenous antigens.
- Digestion of the invariant chain leaves **CLIP**, a small fragment remaining in the binding groove of the class II MHC molecule.
- HLA-DM, a nonclassical MHC class II molecule expressed within endosomal compartments, mediates exchange of antigenic peptides for CLIP.




Endogenous pathway (class I MHC)
Exogenous pathway (class II MHC)
Endogenous antigen
1 Endogenous antigen is degraded by proteasome.
Proteasome
Rough endoplasmic reticulum (RER)
2 Peptide is transported to RER via TAP.
Invariant chain
ERp57
Calreticulin
TAP
Tapasin
β2-microglobulin
Class I MHC
Cal-nexin
Class II MHC
Peptide
3 Class I MHC α chain binds calnexin, then β2 microglobulin. Calnexin dissociates. Calreticulin, tapasin, and ERp57 bind. MHC captures peptide, chaperones dissociate.
Golgi complex
Digested invariant chain
CLIP
4 Class I MHC–peptide is transported from RER to Golgi complex to plasma membrane.
Exogenous antigen
5 HLA-DM mediates exchange of CLIP for antigenic peptide.
Class I MHC
Class II MHC
6 Class II MHC–peptide is transported to plasma membrane.
1 Class II MHC α and β bind invariant chain, blocking binding of endogenous antigen.
2 MHC complex is routed through Golgi to endocytic pathway compartments.
3 Invariant chain is degraded, leaving CLIP fragment.
4 Exogenous antigen is taken up, degraded, routed to endocytic pathway compartments.



Antigen Processing and Presentation by MHCs