HLA and antigen presentation

Department of Immunology
Charles University, 2nd Medical School
University Hospital Motol

MHC in adaptive immunity

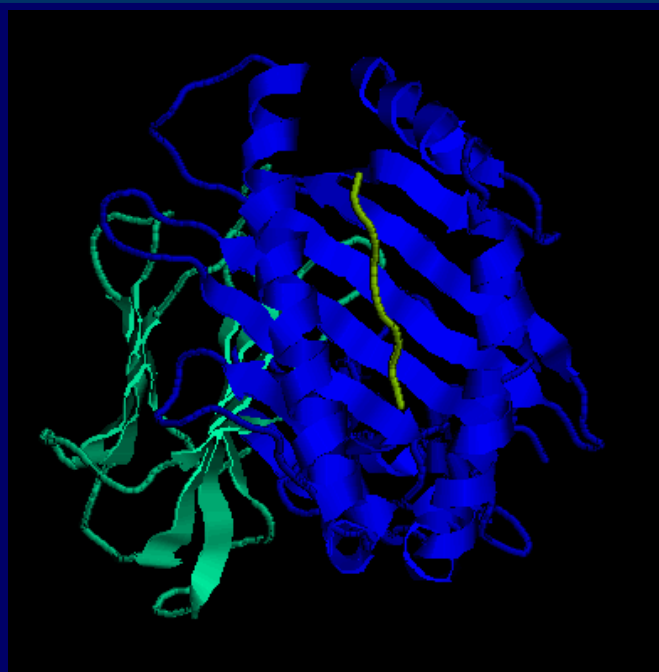
	Innate	Adaptive
Characteristics		
Specificity	For structures shared by groups of related microbes	For antigens of microbes and for nonmicrobial antigens
Diversity	Limited; germline-encoded	Very large; receptors are produced by somatic recombination of gene segments
Memory	None	Yes
Nonreactivity to self	Yes	Yes
Components		
Physical and chemical barriers	Skin, mucosal epithelia; antimicrobial chemicals	Lymphocytes in epithelia; antibodies secreted at epithelial surfaces
Blood proteins	Complement	Antibodies
Cells	Phagocytes (macrophages, neutrophils), natural killer cells	T and B Lymphocytes

T cells recognise cell-associated antigens displayed on

MHC = major histocompatibility complex

Outline

- Adaptive immunity, role of MHC (HLA)
- discovery of HLA genes
- structure of HLA genes and molecules
- polymorphism of HLA molecules
- nomenclature of HLA system
- HLA association with disease
- antigen presentation



MHC GLYCOPROTEINS

CENTRAL MOLECULES OF IMMUNITY

MHC gp I – EXPRESSION NA ALL CELLS

MHC gp II – EXPRESSION ON APC

FUNCTION – "EXHIBIT" ON CELL SURFACE SAMPLES OF FRAGMENTS OF ENDOGENOUS (MHC gp I) RESP. EXOGENOUS (MHC gp II) PROTEINS.

THESE COMPLEXES ARE THEN RECOGNIZED BY T-LYMPHOCYTES (T_h, T_c)

HLA – MHC: basic facts

- Two groupes of MHC genes:

  structurally and functionally distinct

  class I recognition by CD8+ T cells

  class II recognition by CD4+ T cells

- HLA molecules are responsible for the compatibility of the tissues of genetically different individuals and for the rejection of transplant

- MHC genes are codominantly expressed in each individual
- monozygotic twins have the same histocompatibility molecules on their cells
- MHC genes are the most polymorphic genes present in the genome!

  (Up to 250 alleles identified for some loci)

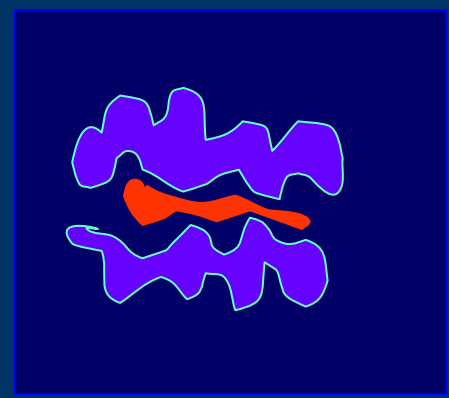
MHC expression

Class I

On all nucleated cells (no MHC on red blood cells, weak expression on cells in CNS)

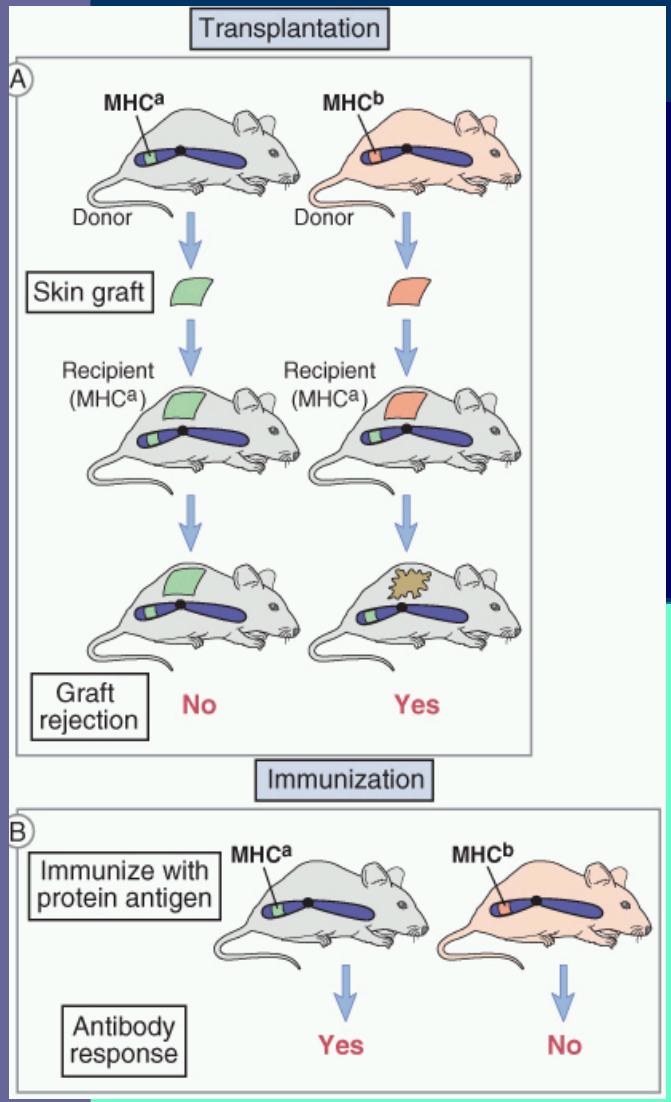
Class II

Found on antigen presenting cells



HUMAN		MOUSE	
MHC I	HLA-A, -B, -C	MHC I	H-2K, D, L
Ib	E, F, G CD1)	Ib	Qa, TL, H-2M3 (CD1)
MHC II	DR, DQ, DP	MHC II	I-A, I-E
	(DM)		(DM)

Discovery of Human MHC



- Recognition of a graft as self or foreign is an inherited trait
- histocompatibility genes: differences between self and foreign were attributed to their genetic polymorphisms
- Mouse study: identification of MHC locus

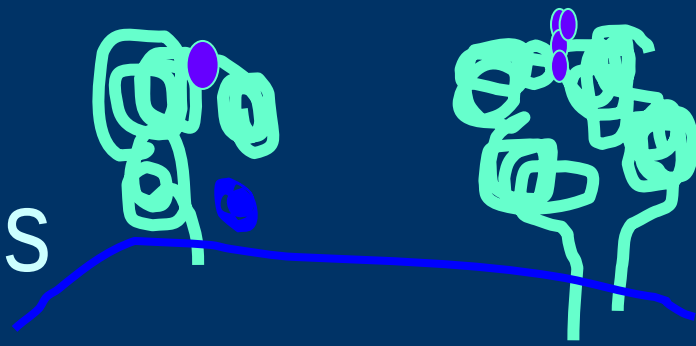
- Human MHC

  In study with transplanted patients discovered „human leukocyte antigens“ HLAs

  HLA-A, HLA-B, HLA-C (class I MHC genes)

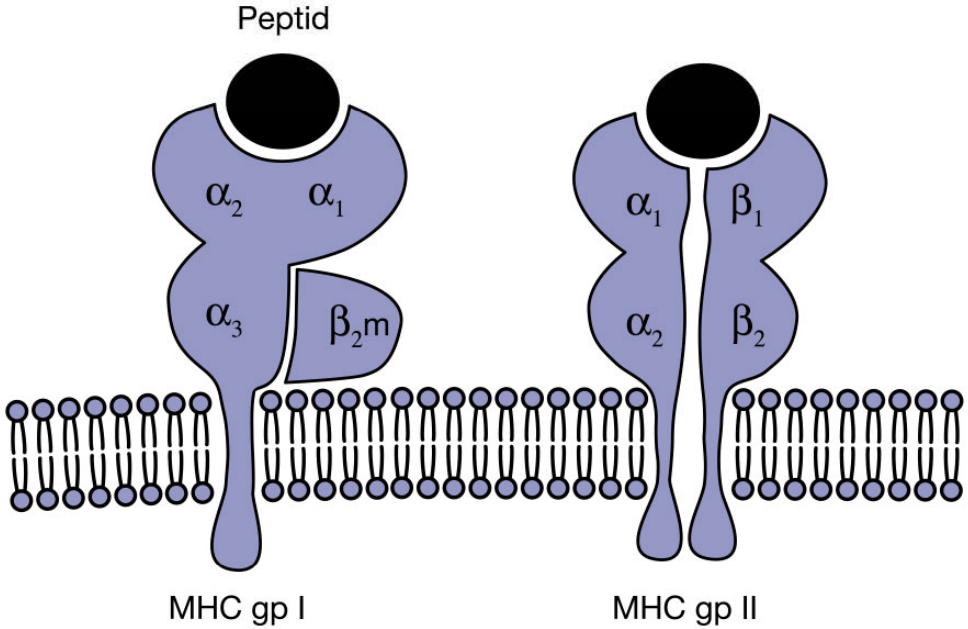
  In study of mixed leukocyte reaction identified HLA-DR, HLA-DP, HLA-DQ (class II MHC genes

structure of HLA molecules

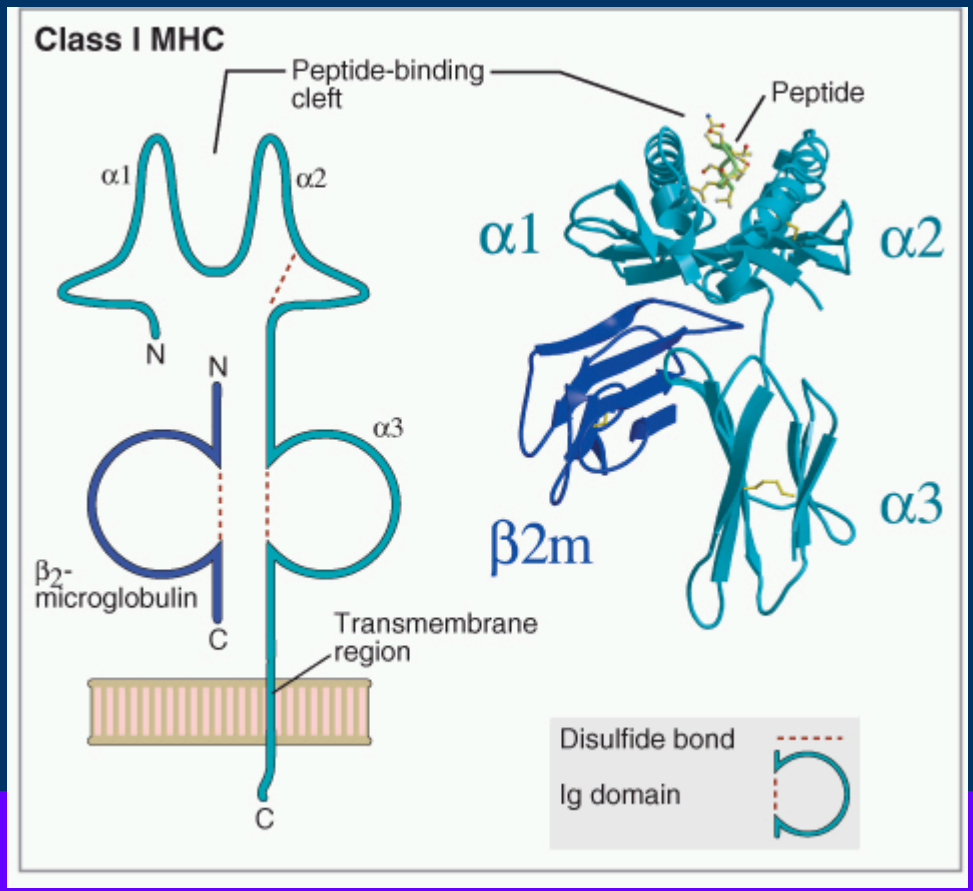


- glycoproteins, heterodimers (two chains)
- Structure of HLA molecules of both classes enables antigen binding and contact with T cell receptors. Extracellulary located peptide binding cleft
- polymorphic (predominantly in the cleft).
- Nonpolymorphic part of the molecule contains binding sites for the T cell molecules CD4 and CD8

(a)



HLA class I. molecules



1. Heavy chain

α1, α2 domain: polymorphic sites

α3 domain: binding of CD8

2. β-2 microglobulin
3. peptide

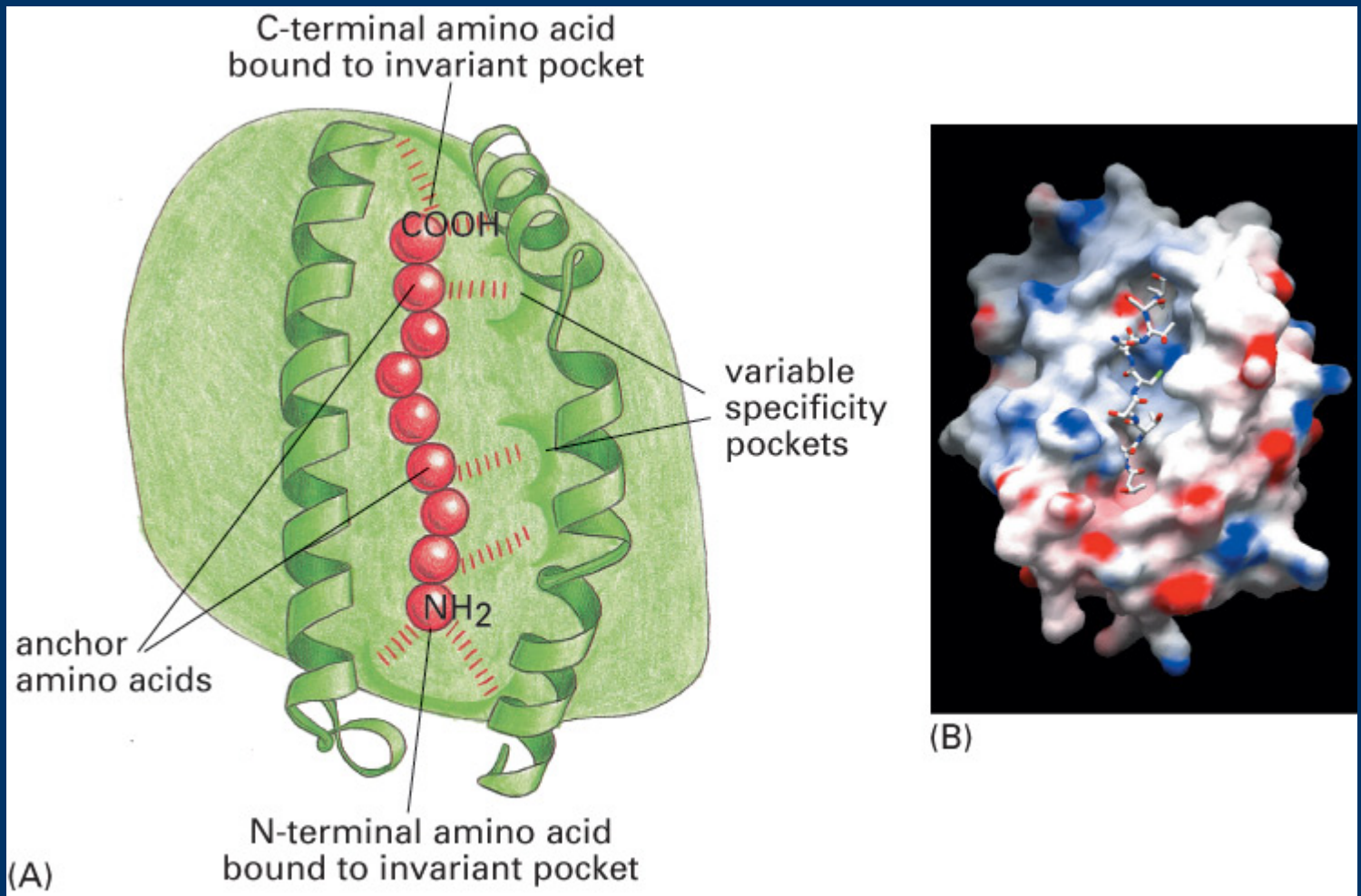


Figure 24–52. Molecular Biology of the Cell, 4th Edition.

Structure of HLA class II. molecules

1. α chain

α1: polymorphic sites

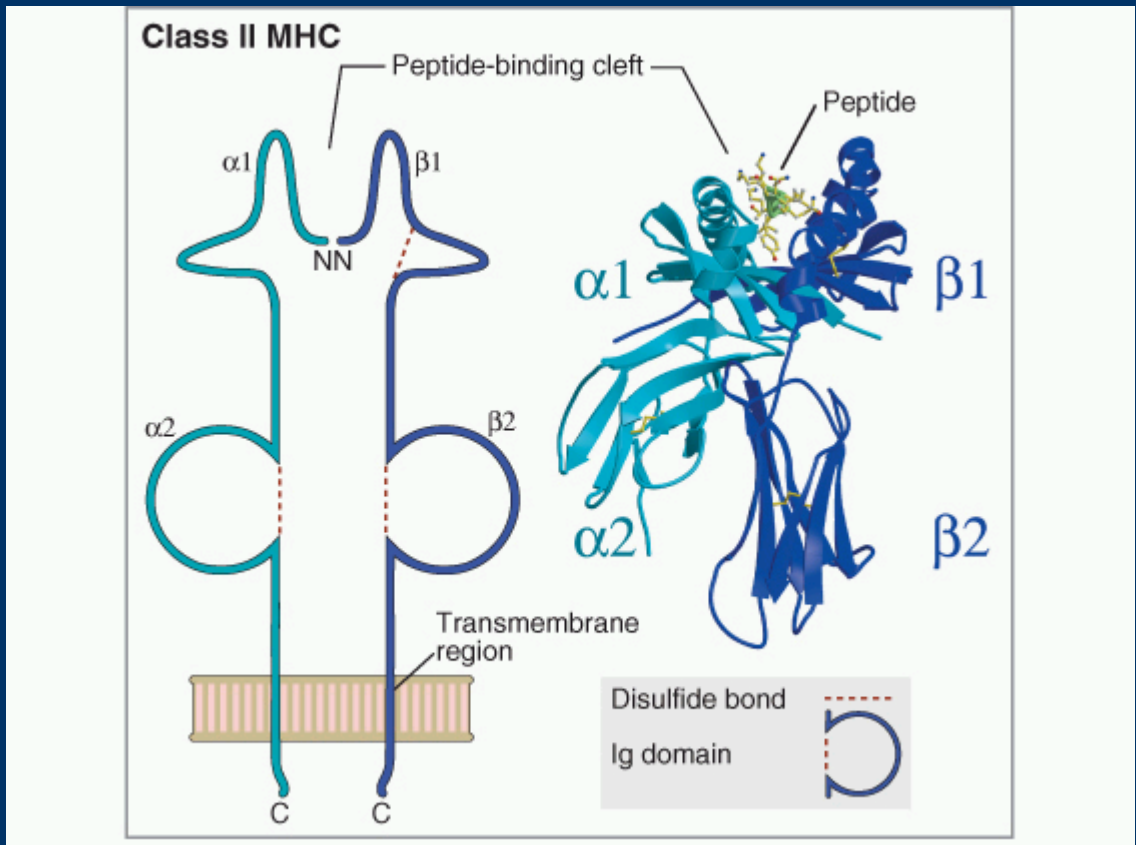
α2: binding of CD4

2. β chain

β1: polymorphic sites

β2: binding of CD4

3. peptide



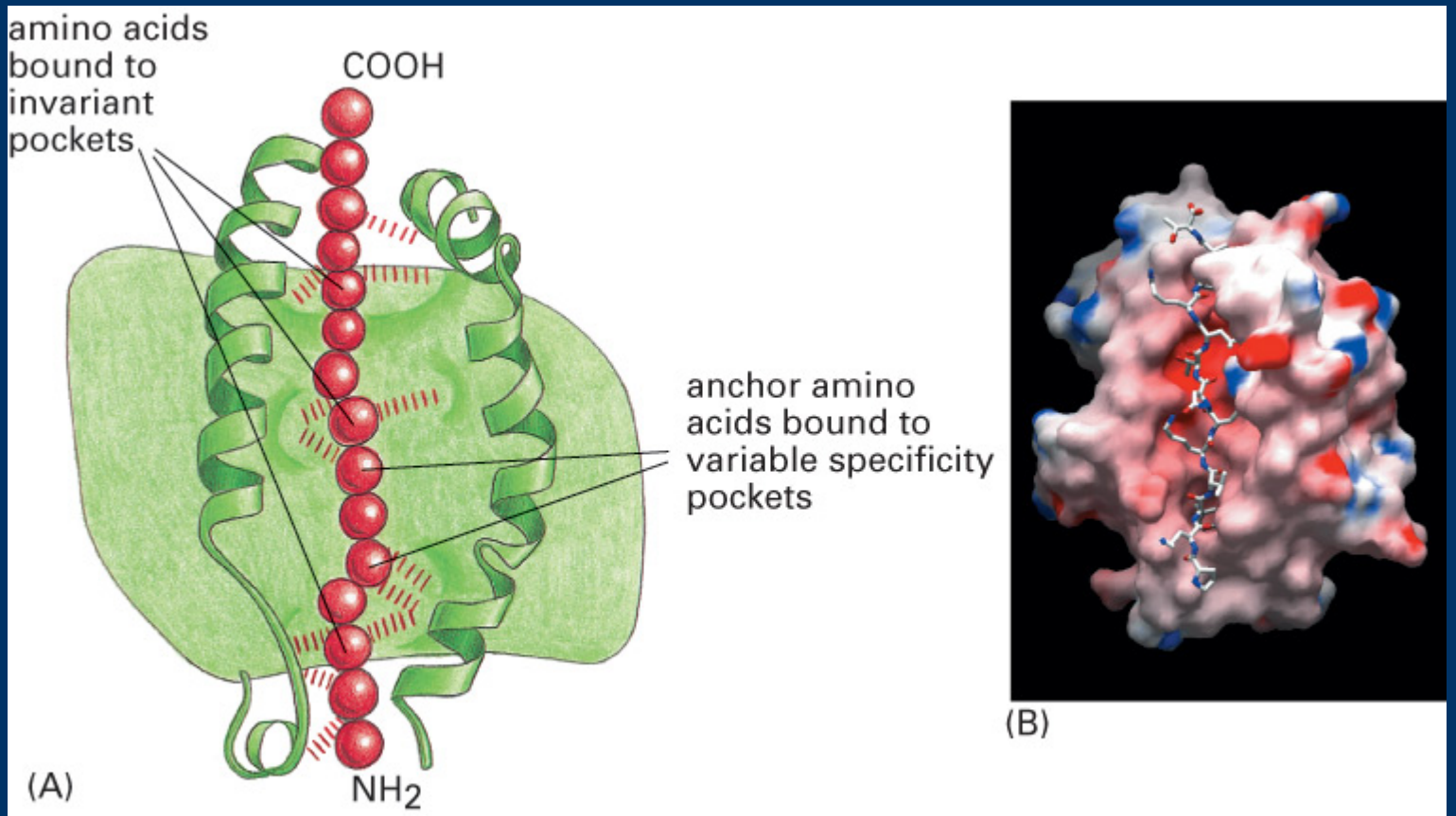


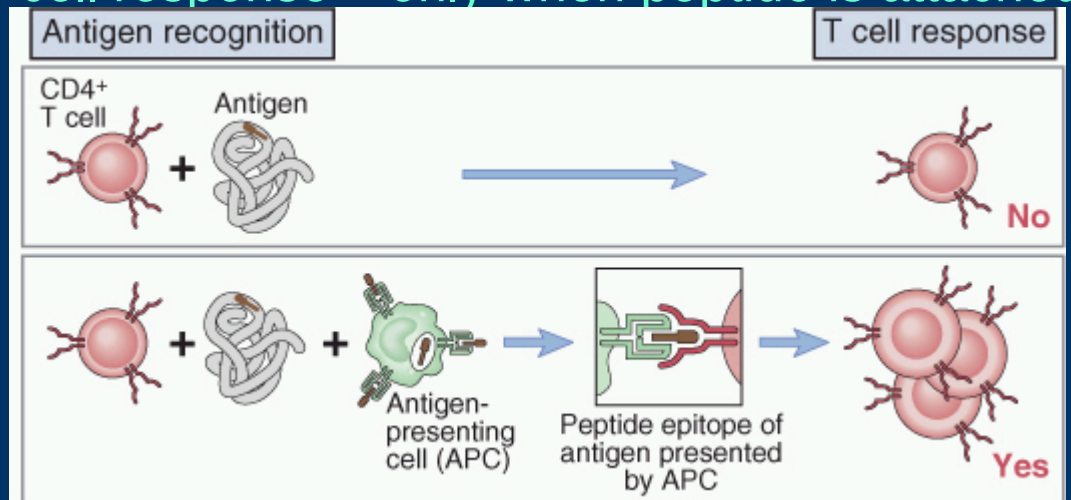
Figure 24–53. Molecular Biology of the Cell, 4th Edition.

HLA and antigens

- Most T lymphocytes recognize only peptides
- T cells are specific for amino acid sequences of peptides - TCR
- Intracellular antigens are presented in connection with HLA class I. - CD8+ T cells recognition
- Extracellular antigens are presented in connection with HLA class II.
  – CD4+ T cells recognition

Experiment:

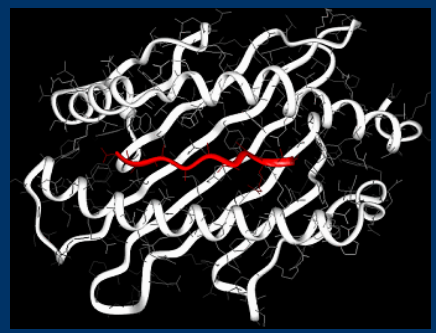
- T cell response – only when peptide is attached to APC

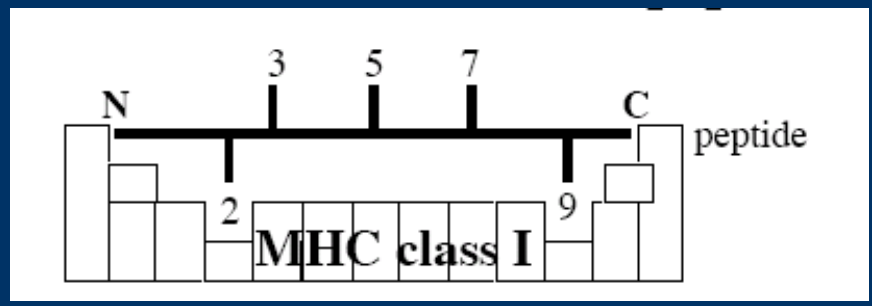


HLA and peptides

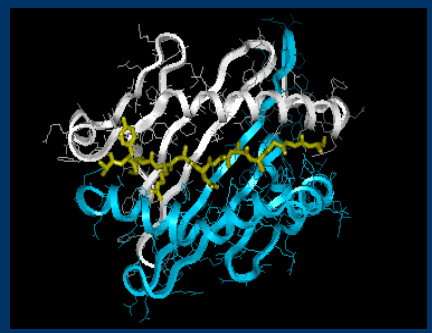
- antigenic peptides in the binding sites of HLA molecules
- One MHC - many peptides sharing structural features can bind
- Interaction has a very slow on- and off-rate (very stable)

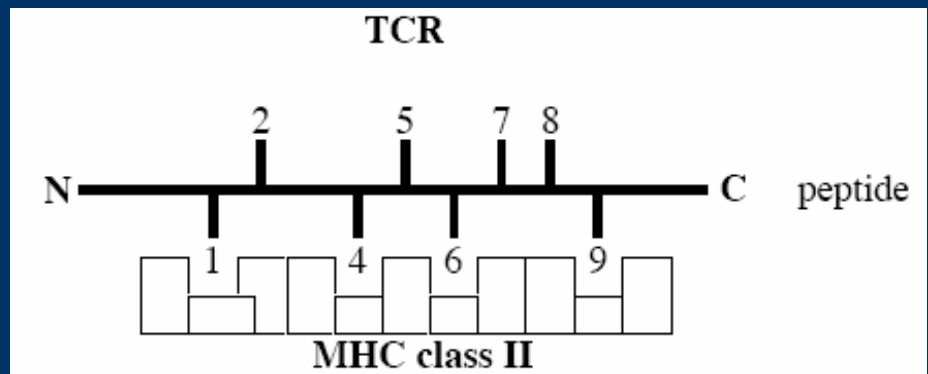
- class I.

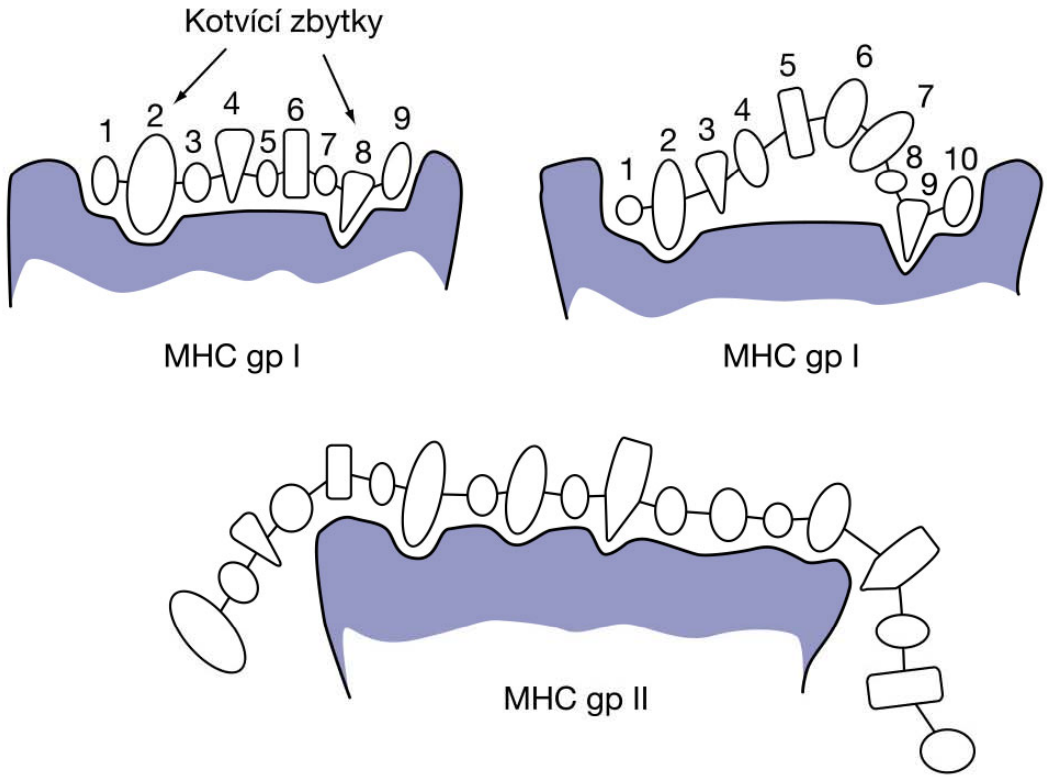




- class II.



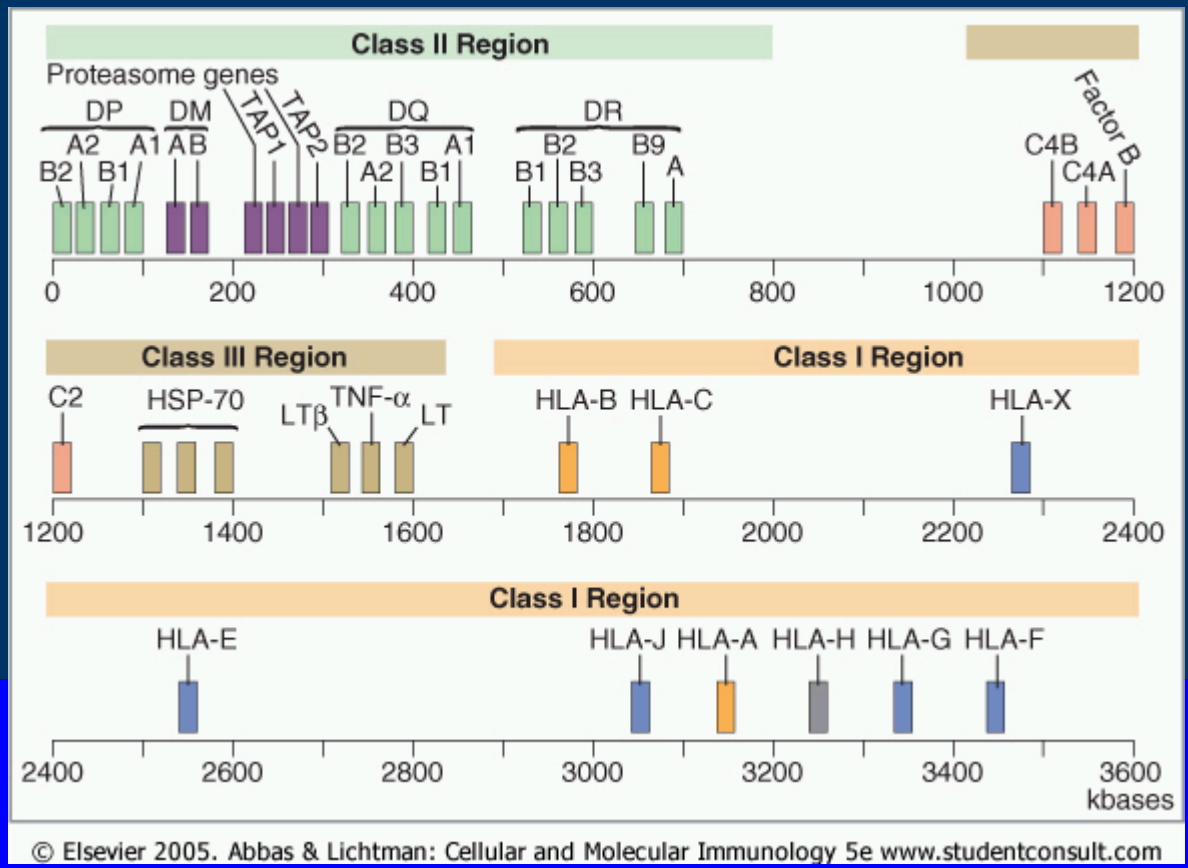



Kotvící zbytky
1
2
3
4
5
6
7
8
9
MHC gp I
1
2
3
4
5
6
7
8
9
10
MHC gp I
MHC gp II

HLA genes

MHC locus

- On chromosome 6
- HLA class III. are soluble molecules as complement, TNF, HSP
- Many other proteins involved in antigen presentation



HLA nomenclature

Nomenclature: The genetic "unit" of the HLA system is the allele, with each defined by its own DNA nucleotide sequence

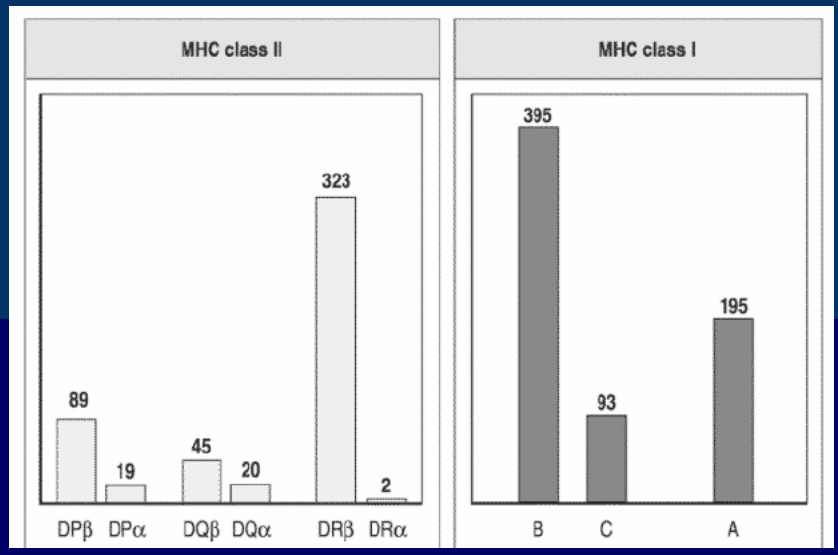
Allele	E.g. HLA-B*0801	}	"Specificity"
	*0802		HLA-B8
	…		
	*0821		
	*2701	}	
	*2702		
	*2703		HLA-B27
	…		
	*2725		

But to make things "simpler", alleles can be grouped in families, e.g. HLA-B*27

"specificity", is an old nomenclature used when human alloantibodies were used to first detect HLA serologic "specificities" or "antigens"

HLA polymorphism

- most polymorhic structures from all known systems.



- human study: 1000 donors, HLA-A, B genotyping
  - Over half the group had a combination that was unique.
  - Another 111 donors had a set of these molecules that they shared with only one other person in the group.
  - The most frequent phenotype (HLA-A1, HLA-A3, HLA-B7, and HLA-B8) was found in 11 donors.

EXTRAORDINARY POLYMORPHISM OF MHC PROTEINS:

HUNDREDS OF ALLELIC FORMS

IMPORTANT FOR BETTER PROTECTION OF BOTH AN INDIVIDUAL AND POPULATION

(COMPLICATION – TRANSPLANTATIONS)

HLA polymorphism – why?

Pathogen driven mechanisms

- Pathogens tend to escape
- Heterozygotes have advantage
- Frequency-dependent selection: the individual with the rarest allele has the best chance to survive an infection

cheetahs (low polymorphism): extremly susceptible to infectious diseases

vertebrate species can detect MHC genotype by smell!

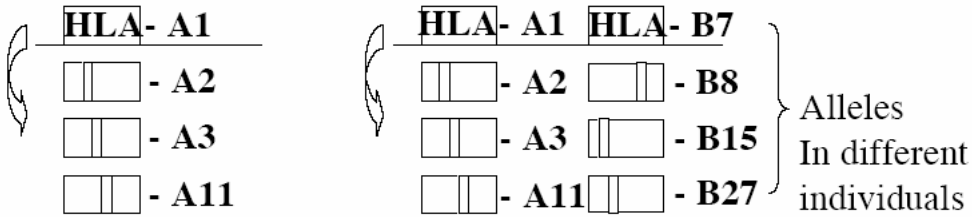
Diversity of MHC class I and II genes

Diversity of MHC class I and II genes

Arises from two mechanisms:

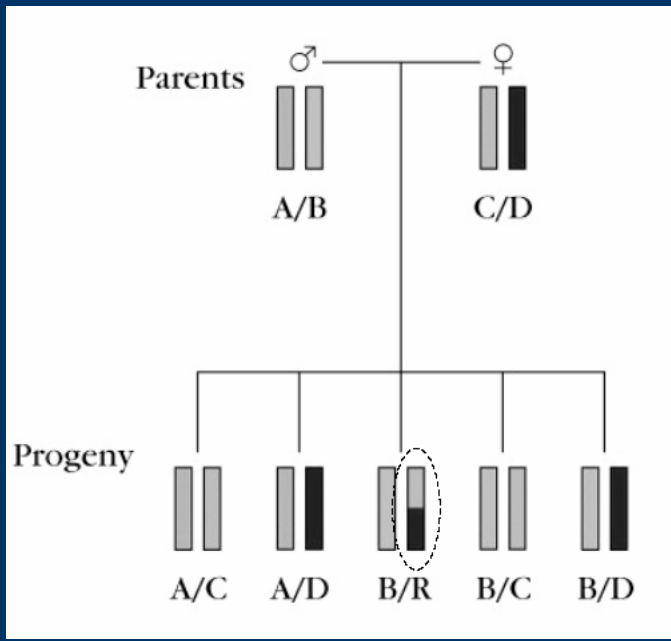
Duplication of a gene locus in an individual resulting in multiple loci, *polygeny*

Development of multiple alleles at a locus among individuals in the species, *polyallelism*



HLA haplotypes in a typical family

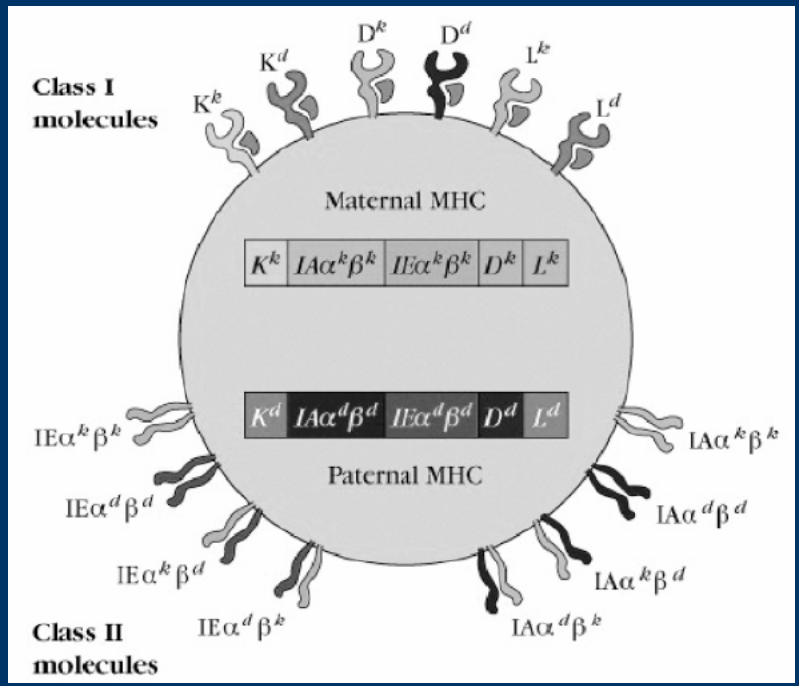
- Haplotype is combination of allelic forms of HLA molecules on one chromosome.
- We inherit 3 types of heavy chains for HLA class I. molecules from each parent .
- Everybody expresses 6 different types of HLA class I. molecules unless honmozygous status for some of the types was inherited.



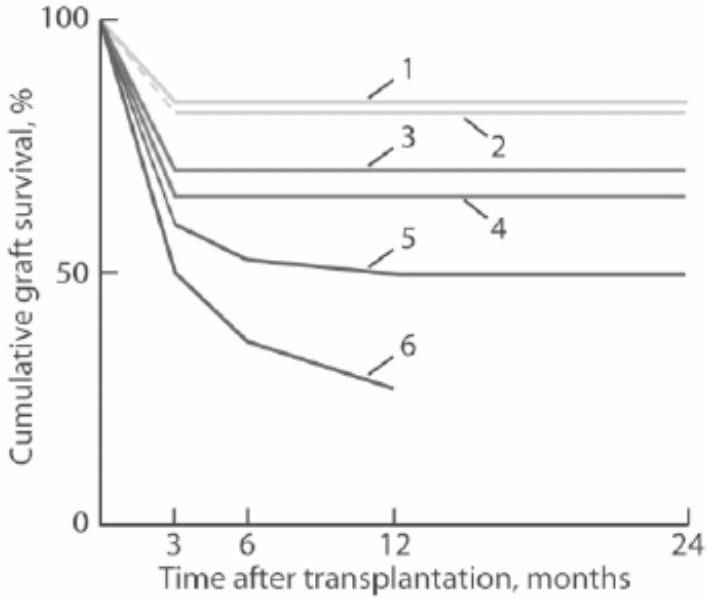
Meiotic recombination scrambles HLA haplotypes

Haplotypes	HLA Alleles					
	A	B	C	DR	DQ	DP
	1	7	w3	2	1	1
B	2	8	w2	3	2	2
C	3	44	w4	4	1	3
D	**11**	**35**	**w1**	**7**	**3**	**4**
R	3	44	w4	**7**	**3**	**4**

Co-dominant expression of MHC alleles



MHC control transplantat survival



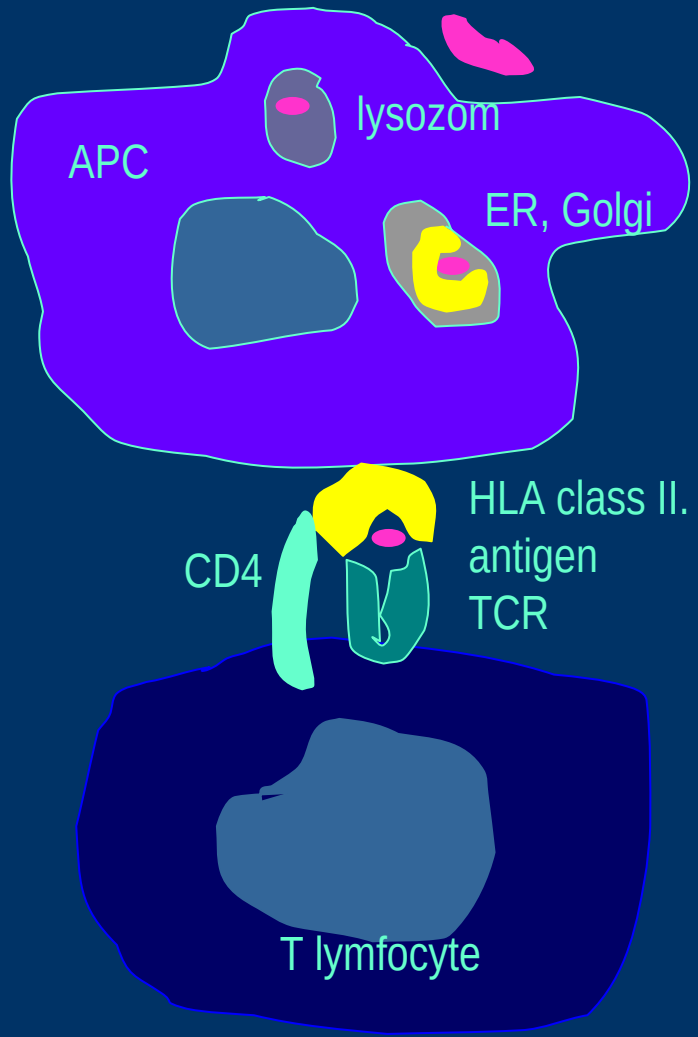
Curve no.	HLA mismatches (no.) Class I	Class II
1	0	0
2	1 or 2	0
3	3 or 4	0
4	0	1 or 2
5	1 or 2	1 or 2
6	3 or 4	1 or 2

- A graft is compatible only if there is a complete match at all MHC alleles, i.e. a two haplotype match for all MHC loci

Function of MHC

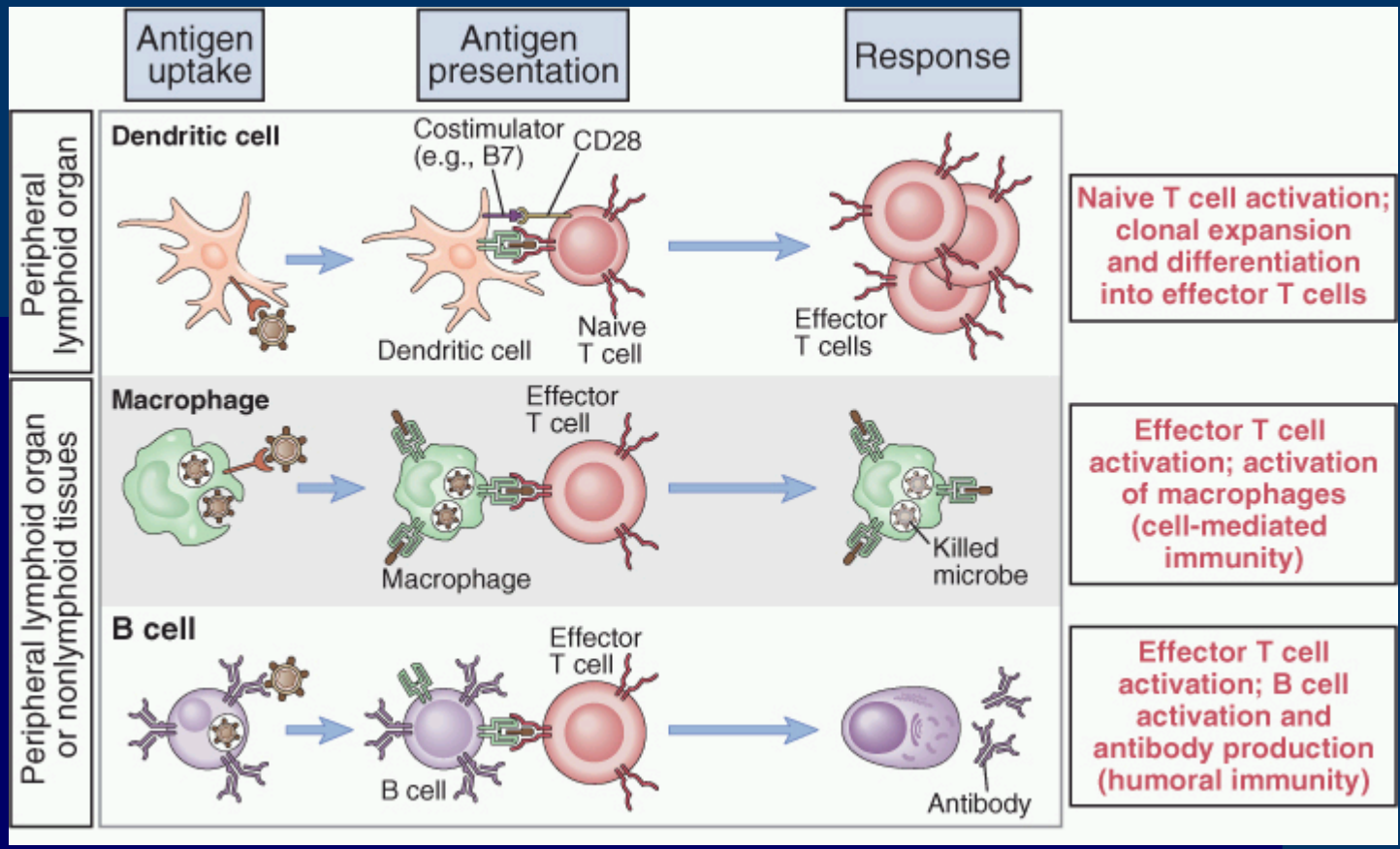
- Recognition of antigen by T cells is necessary for induction of the immune response.

  – exogenous antigen presentation



Antigen presenting cells

Most important population of APC



Other population of APC

- Vascular endothelial cells: function as APC is **inducible**
- Various epithelial and mesenchymal cells: **inducible**

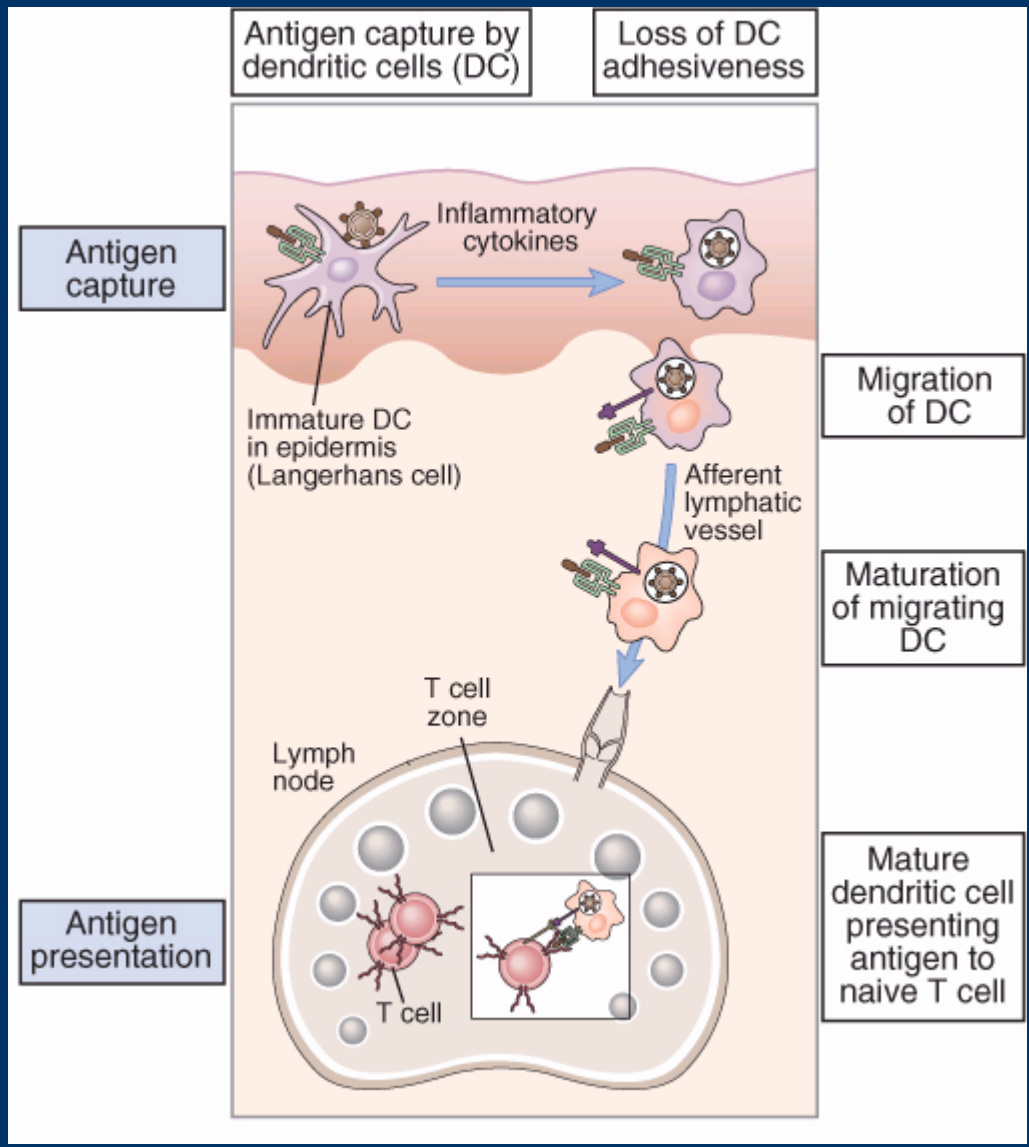
Dendritic cells in antigen presentation

Dendritic cells

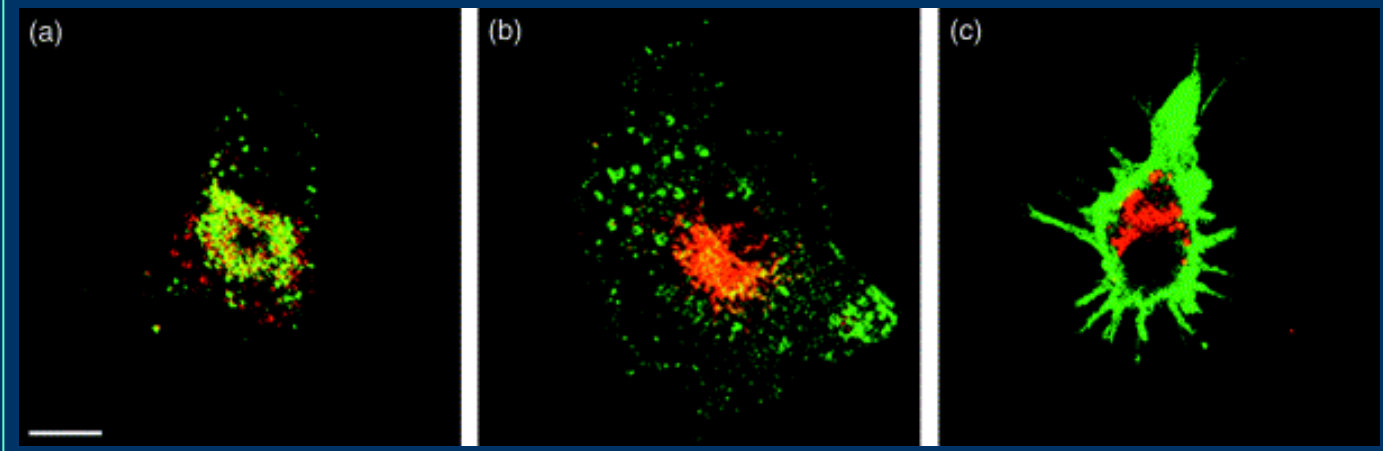
- most effective population in T cell activation
- used as immunoterapeutic tools in cancer vaccines

Immature DC: capture antigens in periphery

Mature DC: activation of T lymphocytes in lymphatic nodes



Antigen Presentation and Dendritic Cells



From Mellman et al., TICB 8: 231 (1998)

Immature (a), maturing (b), and mature (c) dendritic cells stained for MHC class II (green) and lysosomal marker Lamp-1 (red).

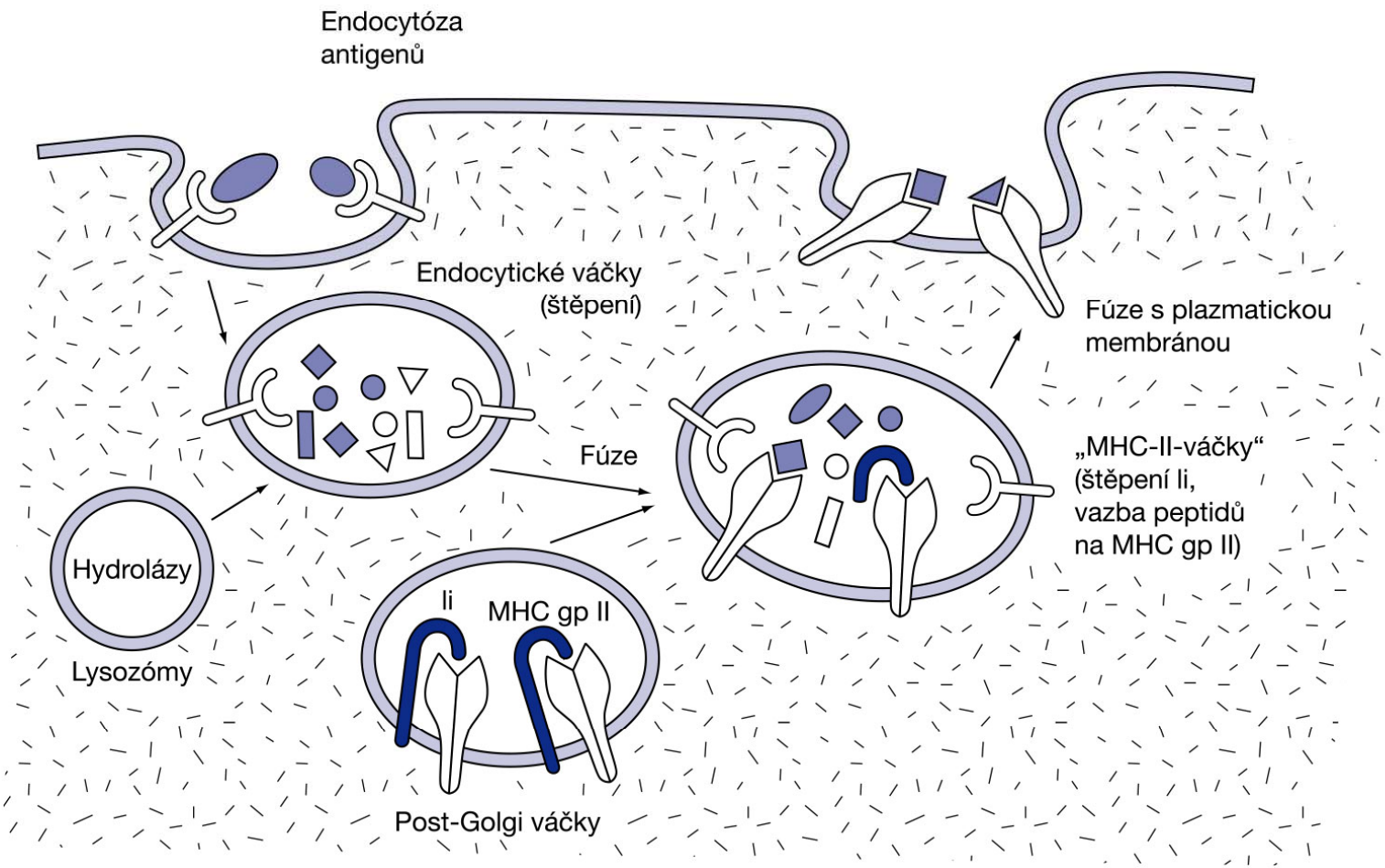
Exogenous antigens

- Exogenous antigens (inhaled, ingested, or injected) are taken up by "professional" antigen-presenting cells
- These include:

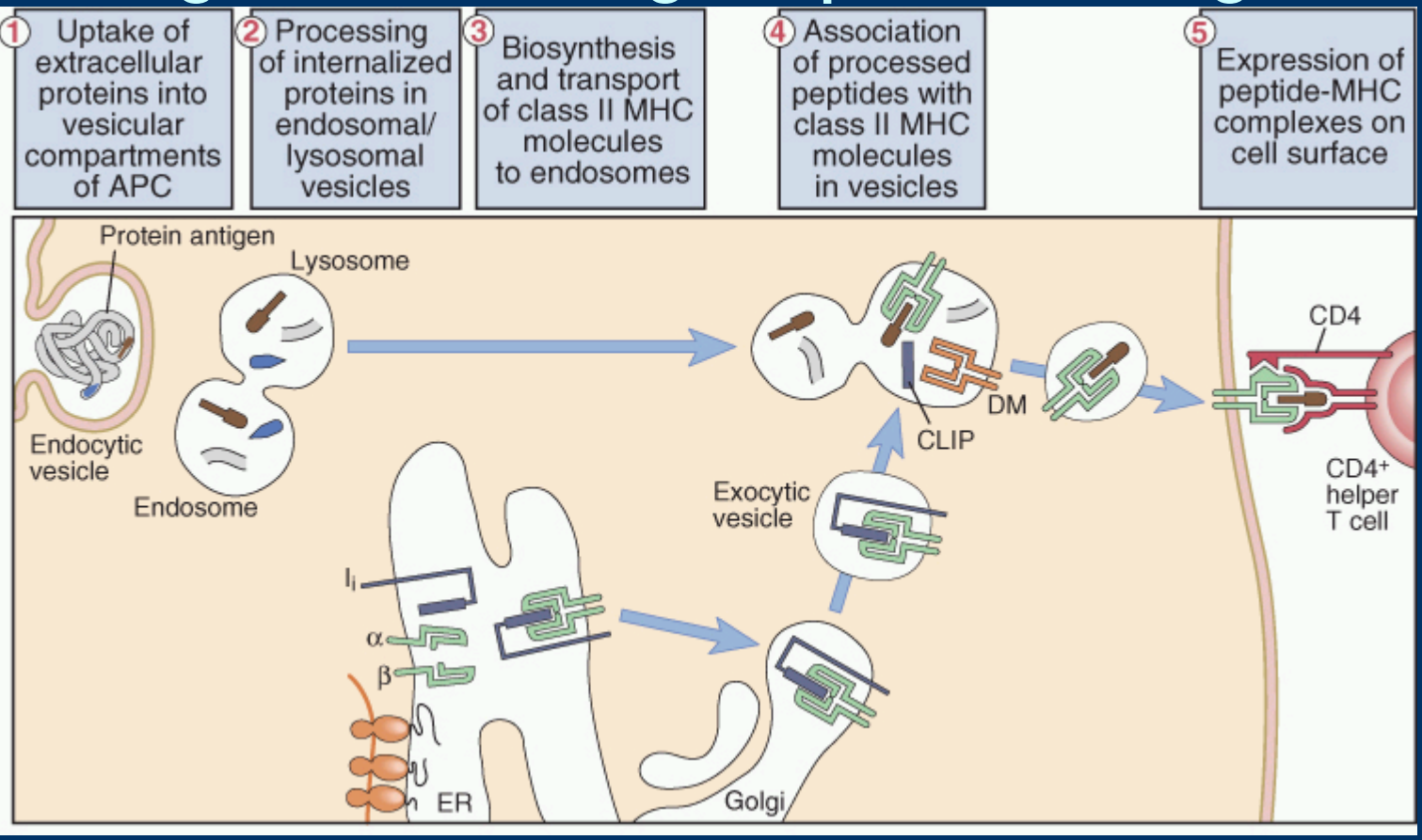
  phagocytic cells like macrophages and dendritic cells

  B lymphocytes which are responsible

  for producing antibodies against the antigen.
- All these cells express HLA class II. molecules


Endocytóza
antigenů
Endocytické váčky
(štěpení)
Fúze s plazmatickou
membránou
Fúze
„MHC-II-váčky“
(štěpení Ii,
vazba peptidů
na MHC gp II)
Hydrolázy
Lysozómy
Ii
MHC gp II
Post-Golgi váčky

Exogenous antigen processing



Invariant Chain (Ii)-Contributes to:

1.proper folding of the alpha and beta chains in the ER

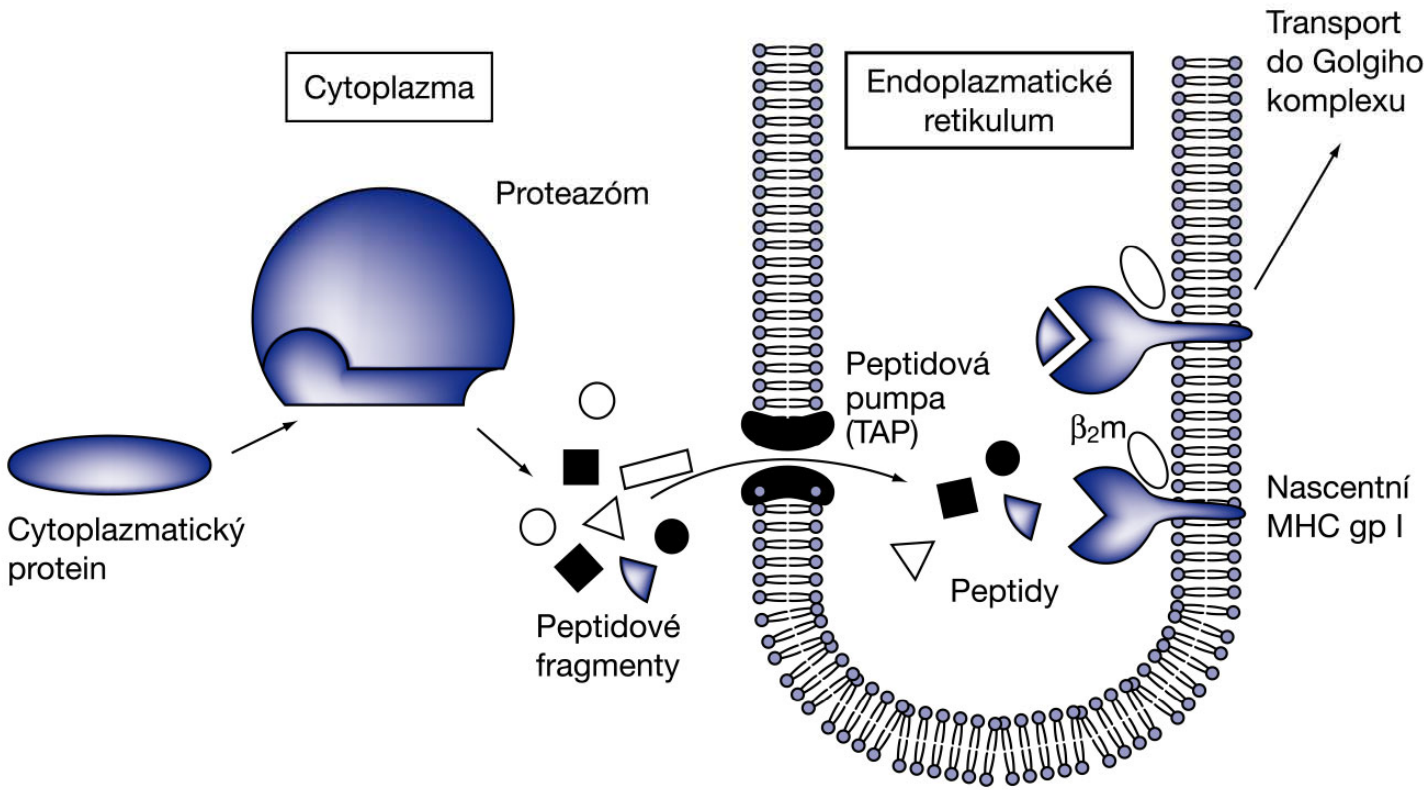
2.preventing in advertent association with endogenous peptides

3.directing immature MHC II through the secretory pathway

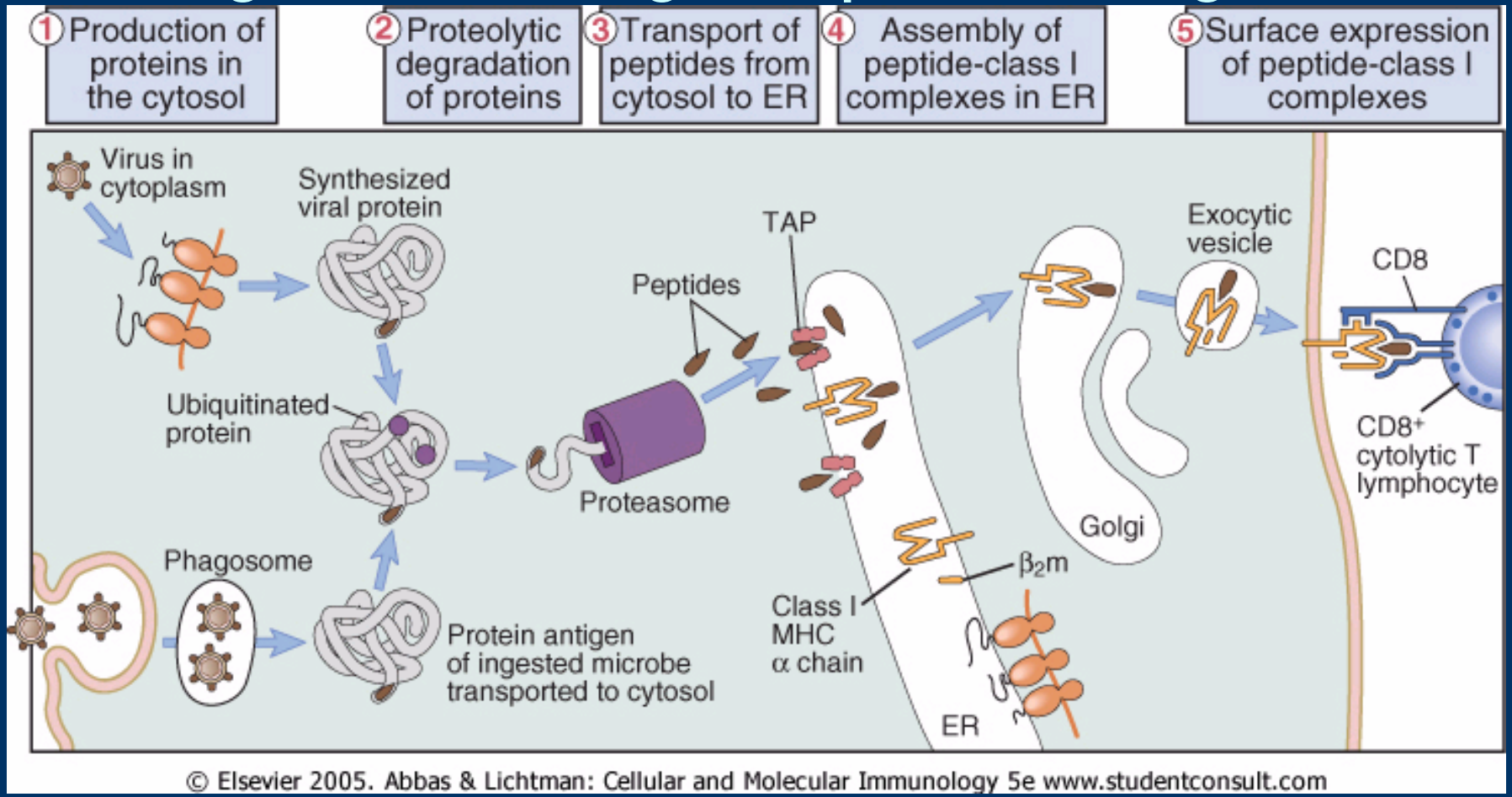
CLIP (Class II-associated invariant chain peptide)-Degradation product derived from the Invariant chain.

HLA-DM Responsible for displacing CLIP and allowing peptide to bind once MHC has reached the endosome.

HLA-DO Negative regulator of HLA-DM activity

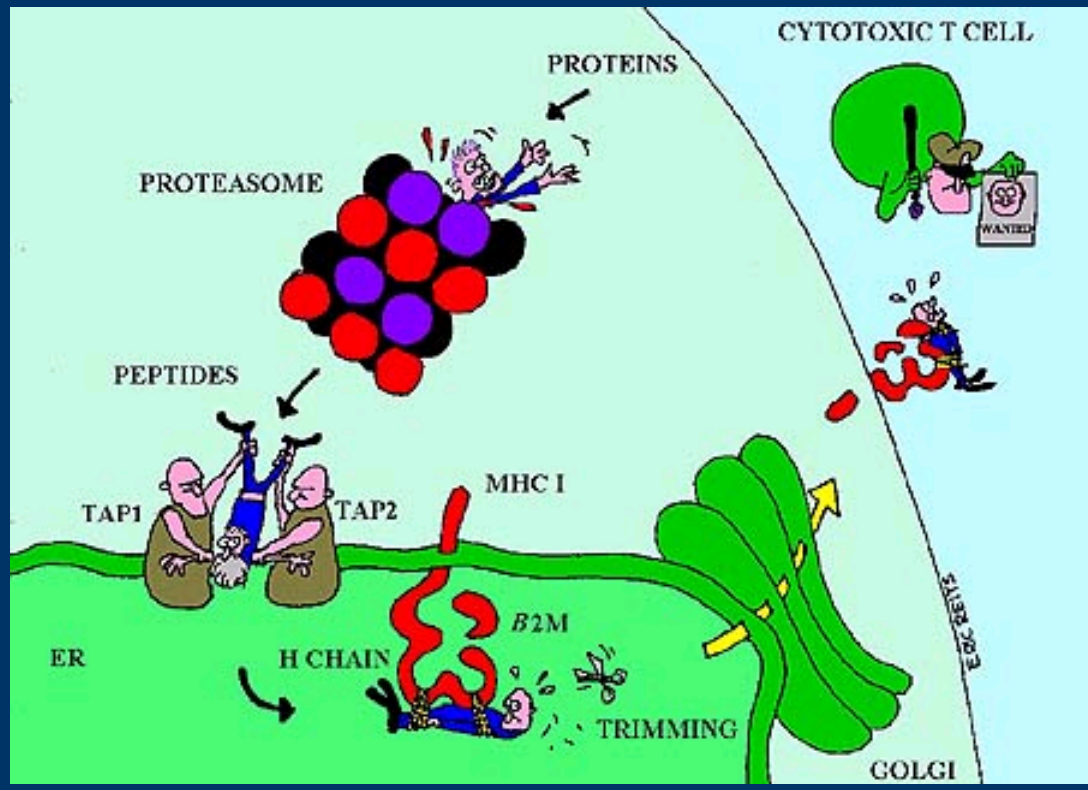

Cytoplazma
Endoplazmatické retikulum
Transport do Golgiho komplexu
Proteazóm
Peptidová pumpa (TAP)
β_2m
Cytoplazmatický protein
Peptidové fragmenty
Peptidy
Nascentní MHC gp I

Endogenous antigens processing

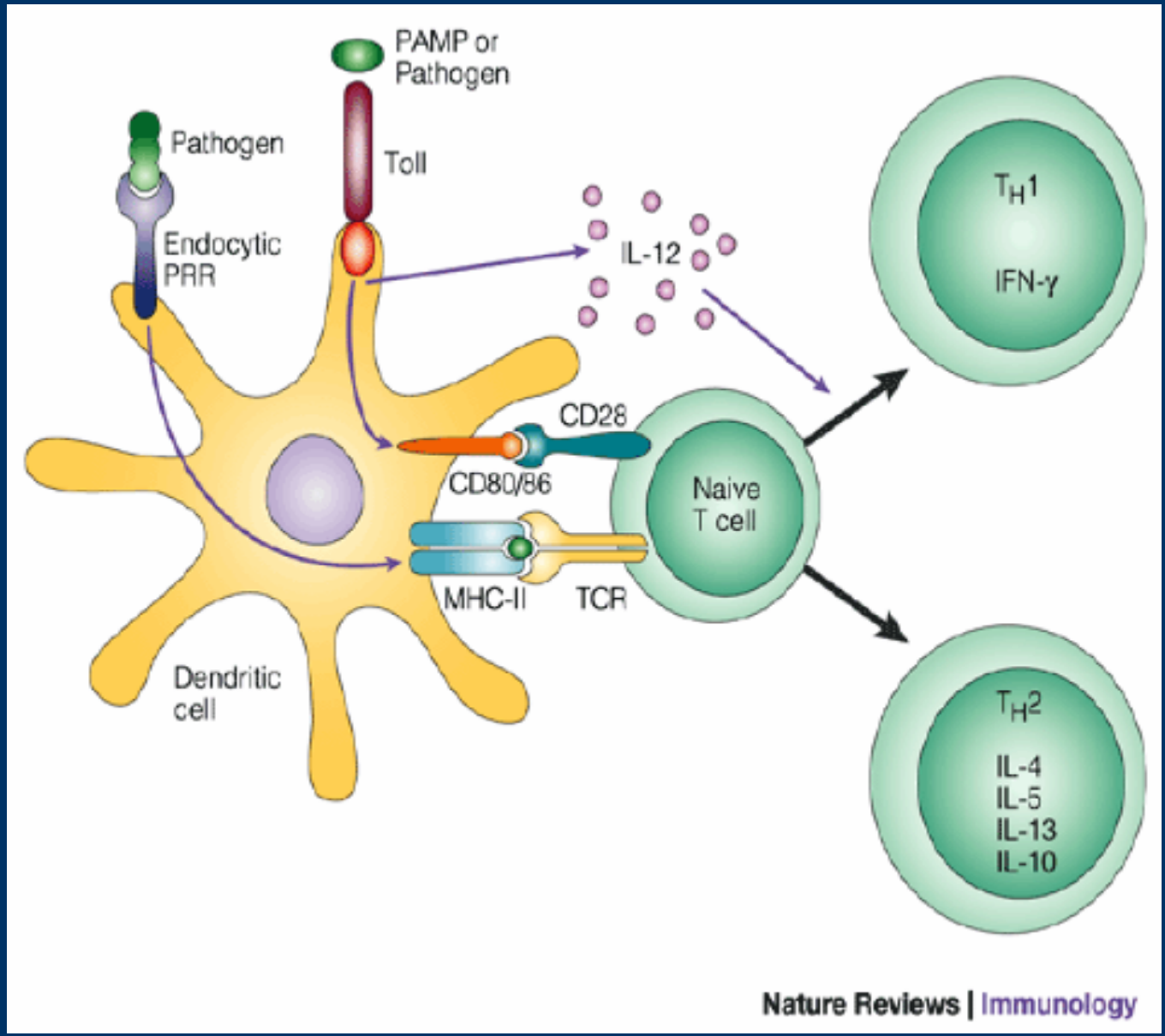


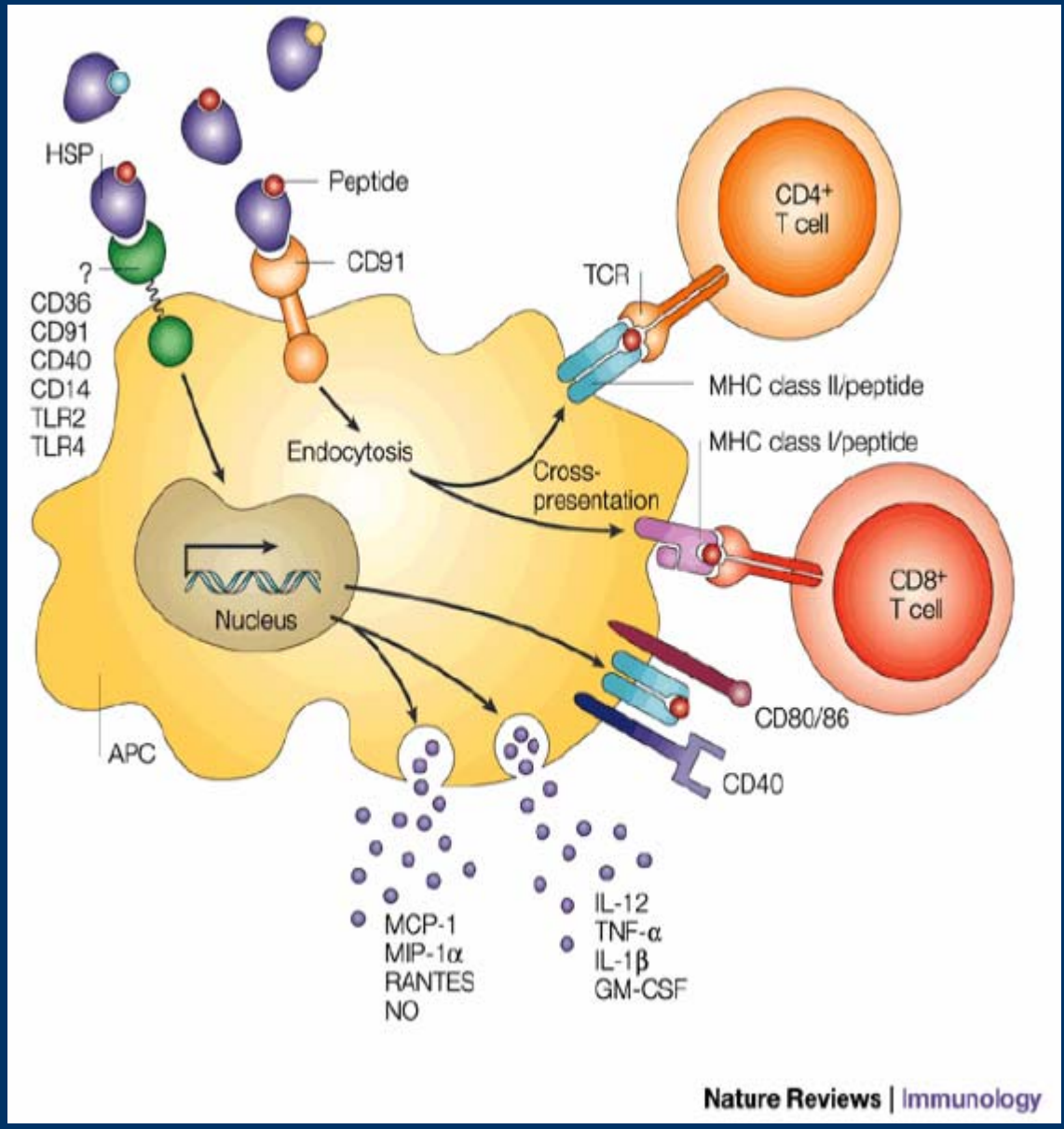
TAP (transporter associated with antigen presentation)

- Transport associated protein - TAP is responsible for the peptide transport from cytoplasm to ER.
- Proteins are degraded to peptide in proteasome.
- The peptides are picked up by TAP proteins and transported from the cytosol into the RER where they assemble with



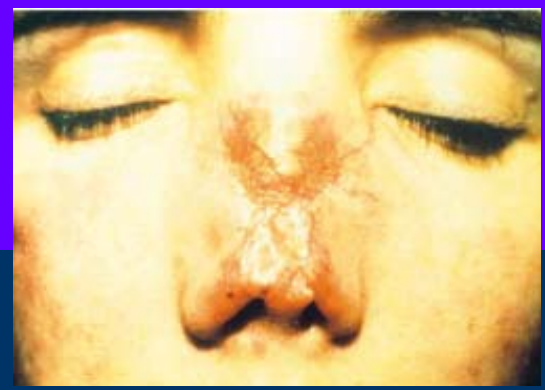
  - the transmembrane polypeptide and beta-2 microglobulin.
  - this trimolecular complex then moves through the Golgi apparatus and is inserted in the plasma membrane


PAMP or
Pathogen
Pathogen
Toll
Endocytic
PRR
IL-12
T_H1
IFN-γ
CD28
CD80/86
Naive
T cell
MHC-II
TCR
Dendritic
cell
T_H2
IL-4
IL-5
IL-13
IL-10
Nature Reviews | Immunology


HSP
Peptide
CD91
?
CD36
CD91
CD40
CD14
TLR2
TLR4
CD4+
T cell
TCR
MHC class II/peptide
MHC class I/peptide
Endocytosis
Cross-
presentation
Nucleus
CD8+
T cell
CD80/86
CD40
APC
MCP-1
MIP-1α
RANTES
NO
IL-12
TNF-α
IL-1β
GM-CSF
Nature Reviews | Immunology

Imunodeficiences - MHC defect

- Bare lymphocyte syndrome:
  mutation in genes regulating class II MHC transcription
  - reduced number of CD4+ T cells in periphery
  - defective activation of CD4+ T cells
  - fatal, treatment: BM transplantation

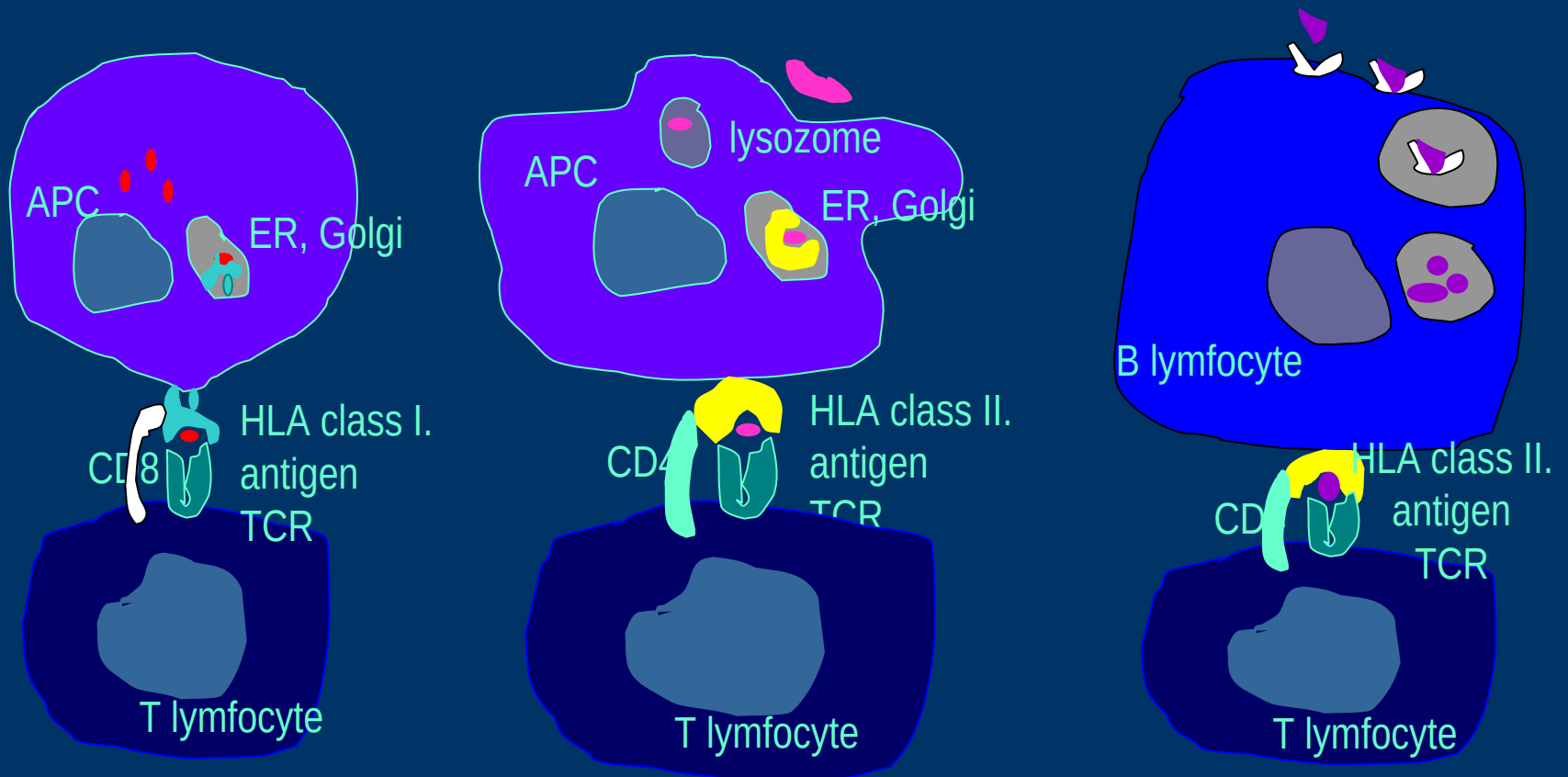


- Class I MHC deficiencies:
  - decreased number of CD4+ T cells in periphery
  - caused by TAP1, TAP2
  - patients suffer from respiratory tract bacterial infection

HLA-associated diseases

	HLA	Patients	Controls
Ankylosing spondylitis	*B27*	90%	9%
Type 1 diabetes	*DR3*	52%	23%
	DR4	74%	24%
	DR3 or *DR4*	93%	43%
Multiple sclerosis	*DR2*	86%	33%
Rheumatoid arthritis	*DR4*	81%	24%
Narcolepsy	*DR2*	>95%	33%

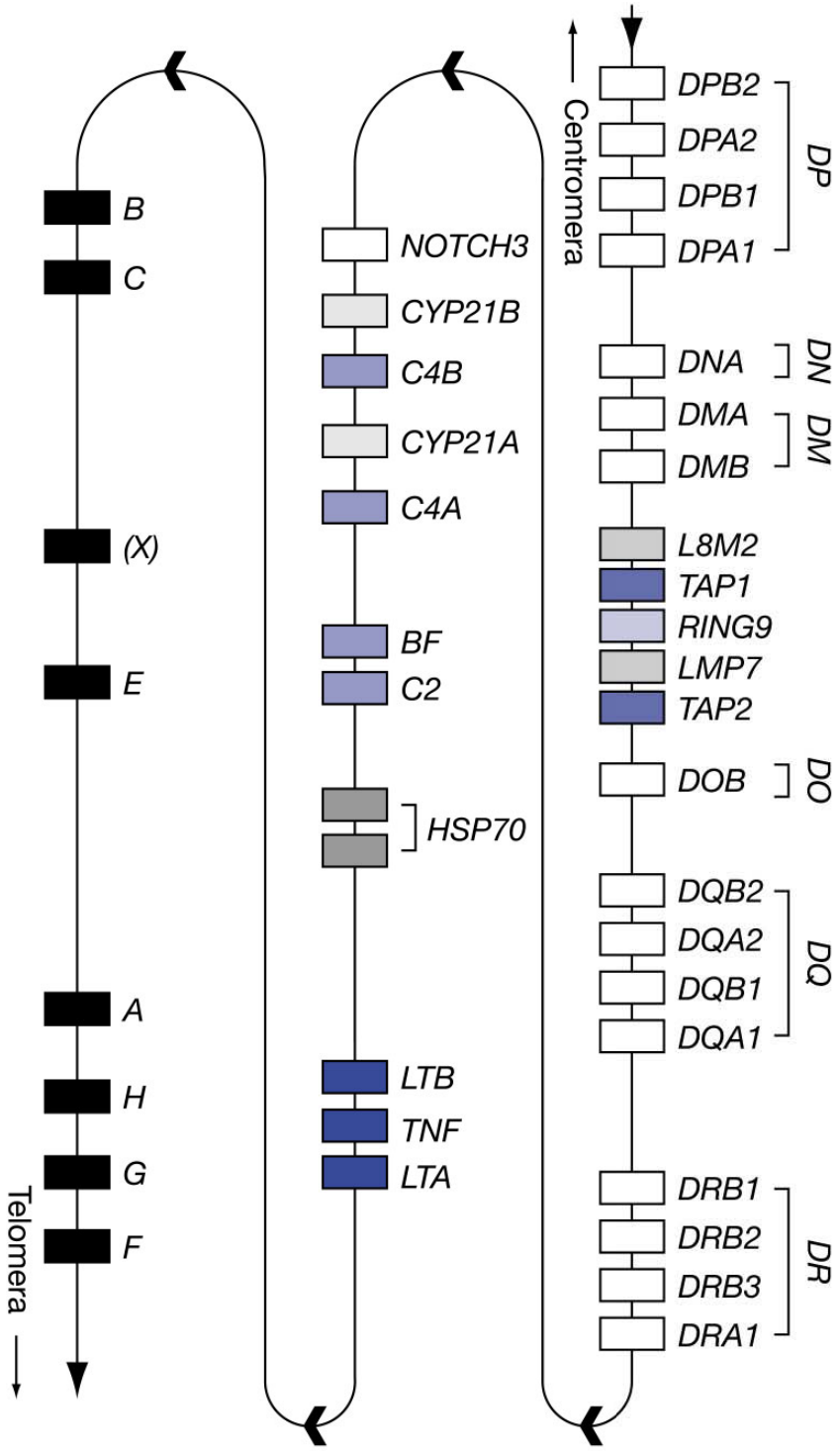
Summary - antigen presentation pathways



endogenous
cell destruction

exogenous
immune response

B lymfocytes
antibody production


Centromera
DPB2
DPA2
DPB1
DPA1
DP
DNA
DN
DMA
DMB
DM
L8M2
TAP1
RING9
LMP7
TAP2
DOB
DO
DQB2
DQA2
DQB1
DQA1
DQ
DRB1
DRB2
DRB3
DRA1
DR
NOTCH3
CYP21B
C4B
CYP21A
C4A
BF
C2
HSP70
LTB
TNF
LTA
B
C
(X)
E
A
H
G
F
Telomera

POLYMORPHISM OF MHC PROTEINS

DRα 1	HLA-A >280
DRβ >400	HLA-B >500
DQβ >50	HLA-C >130

Variability is in the amino acid residues in the peptide binding site!

MHC gp	Motif
HLA-11	xxAspxxxxxTyr
HLA-A2,1	xLeuxxxxxxLeu(Val)
HLA-A11	xValxxxxxxLys
HLA-A24	xTyrxxxxxxLeu(Phe)
HLA-B7	xProArgxxxxxLeu
HLA-B27	xArgxxxxxxLys(Arg)

Disease	HLA	Relative risk*
Ankylosing spondyloarthritis	B27	87.4
Uveitis	B27	10
Goodpasture syndrome	DR2	15.9
Multiple sclerosis	DR2	4.8
Graves-Basedow disease	DR3	3.7
Systemic lupus erythematodes	DR3	5.8
Myasthenia gravis	DR3	2.5
Pemphigus	DR4	14.4
Rheumatoid arthritis	DR4	4.2
Hashimoto thyreoiditis	DR5	3.2