Università degli Studi di Padova

Corso di Laurea in
BIOTECNOLOGIE
Piano di studi Farmaceutico

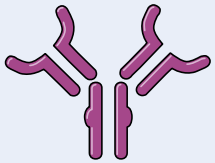
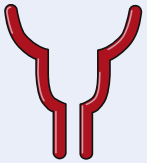
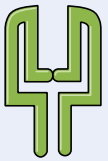
Anno Accademico 2024-2025

Insegnamento di

Immunologia Farmaceutica

I recettori che legano l'antigene nell'immunità adattativa

TABLE 5-1 Features of Antigen Binding by the Antigen-Recognizing Molecules of the Immune System

Feature	Antigen-Binding Molecule		
	Immunoglobulin (Ig) 	T cell receptor (TCR)* 	MHC molecules* 
Antigen-binding site	Made up of three CDRs in V_H and three CDRs in V_L domains	Made up of three CDRs in V_α and three CDRs in V_β domains	Peptide-binding cleft made of α1 and α2 domains (class I MHC) and α1 and β1 domains (class II MHC)
Nature of antigen that may be bound	Macromolecules (proteins, lipids, polysaccharides) and small chemicals	Peptide-MHC complexes	Peptides
Nature of antigenic determinants recognized	Linear and conformational determinants of various macromolecules and chemicals	Linear determinants of peptides; only 2 or 3 amino acid residues of a peptide bound to an MHC molecule	Linear determinants of peptides; only some amino acid residues of a peptide
Affinity of antigen binding	K_d 10^{-7}–10^{-11} M; average affinity of Igs increases during immune response	K_d 10^{-5}–10^{-7} M	K_d 10^{-6}–10^{-9} M; extremely stable binding
On-rate and off-rate	Rapid on-rate, variable off-rate	Slow on-rate, slow off-rate	Slow on-rate, very slow off-rate

CDR, complementarity-determining region; K_d, dissociation constant; *MHC,* major histocompatibility complex; (only class II molecules depicted); V_H, variable domain of heavy chain Ig; V_L, variable domain of light chain Ig.

*The structures and functions of MHC and TCR molecules are discussed in Chapters 6 and 7, respectively.

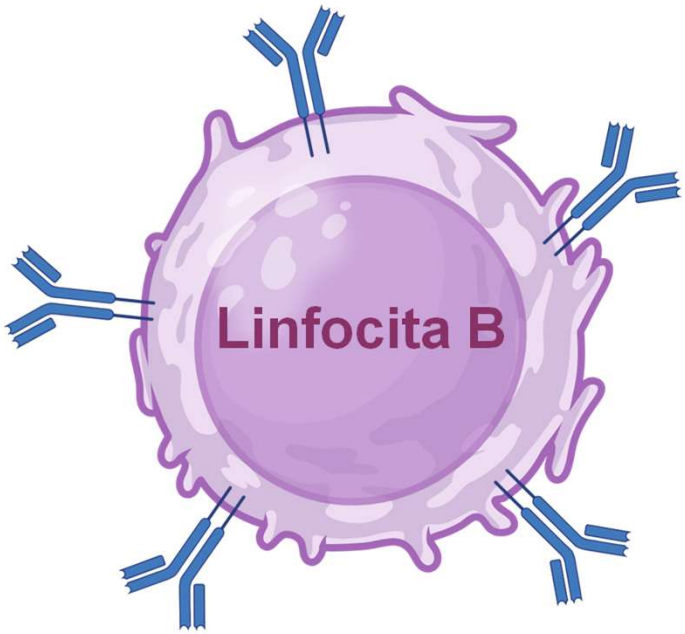
ANTIGEN RECOGNITION:

-B cells and Ig ⇒ Native antigen
(sugars, lipids, hormones, carbohydrates, phospholipids, nucleic acids and proteins)

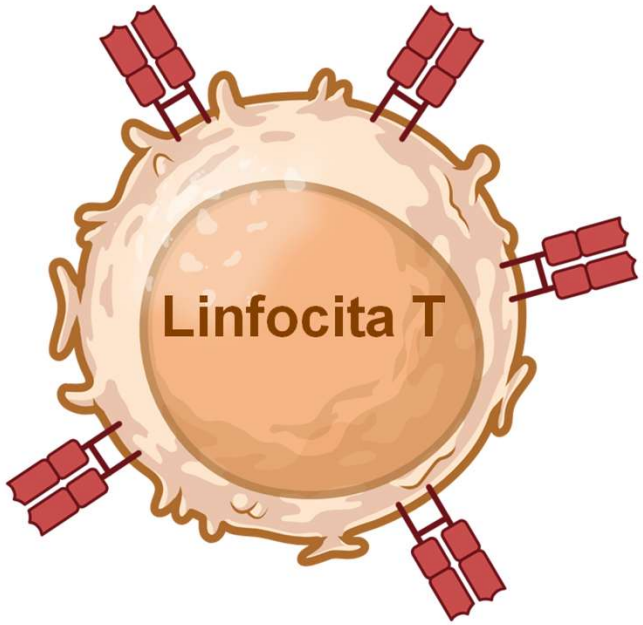
-T cells ⇒ Antigen processed by Antigen presenting cells:
Only peptides

Cellule dell'immunità adattativa

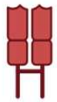
BCR, Ig



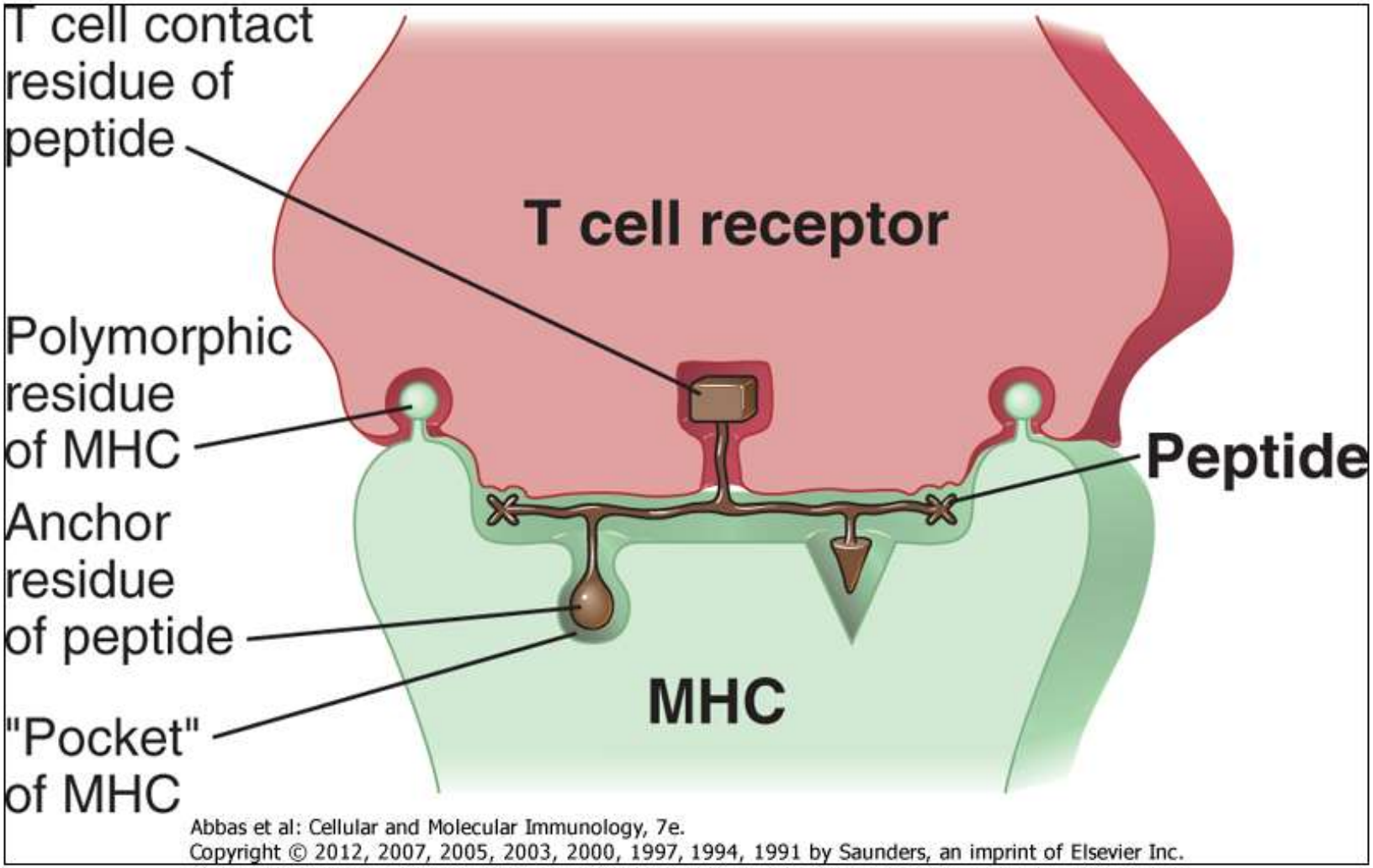
TCR



BCR, Ig, Immunoglobulina di superficie /anticorpo



TCR, Recettore delle cellule T



T cells are needed to control intracellular pathogens and to activate B cell responses

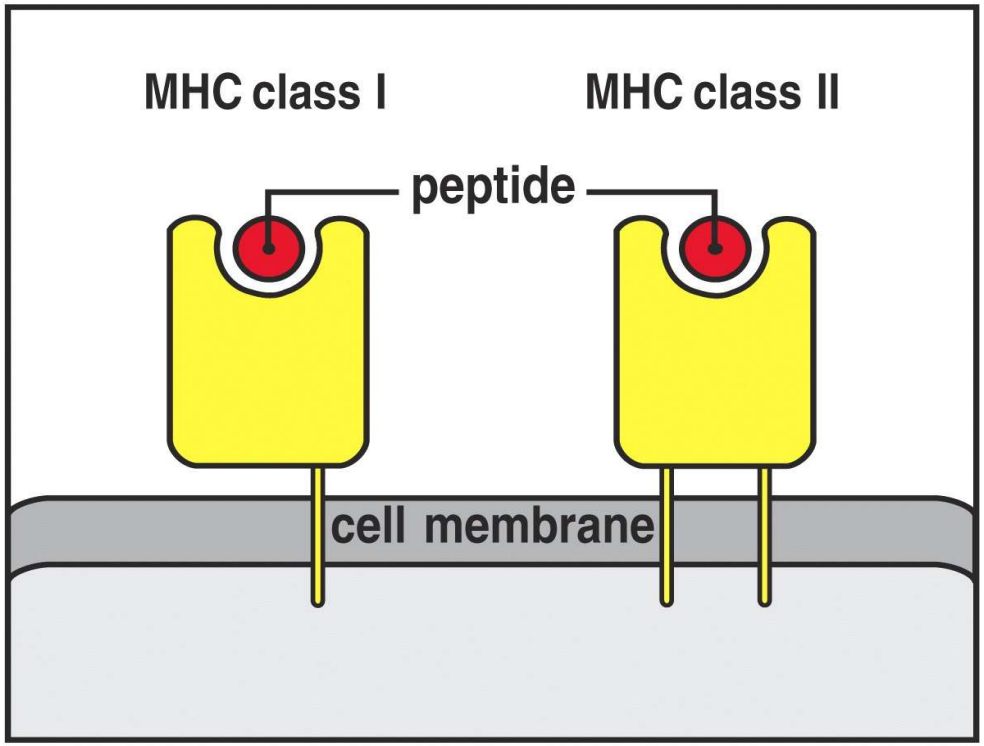


Figure 1-27 Immunobiology, 6/e. (© Garland Science 2005)

MHC/HLA

Complesso genico identificato a metà degli anni ’30 nel topo (complesso H2) da George Snell perché coinvolto nell’attecchimento dei trapianti di cute e d’organo.

Negli anni ’60 e ’70 è stato scoperto che i geni di questa regione sono di fondamentale importanza per tutte le risposte immunitarie agli antigeni proteici

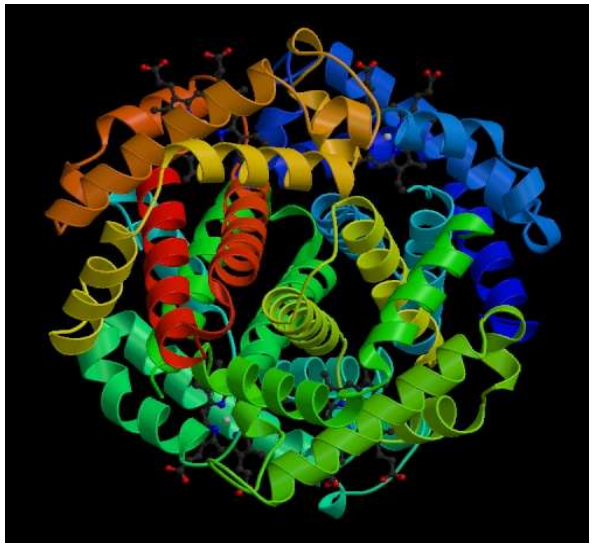
Inbred strains of mice

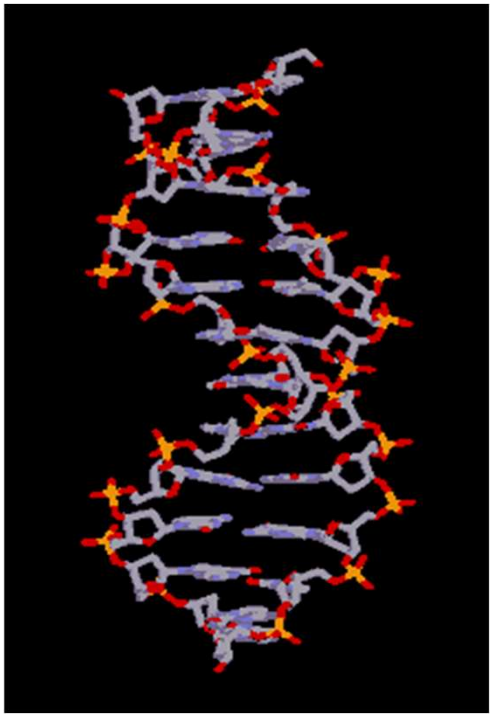
- By inter-crossing mice between brother and sister for at least 20 generations, mice homozygous at more than 98% of loci are obtained. They are named **syngeneic**.

- Two distinct syngeneic strains are defined **allogenic** if expressing different alleles.

- Two distinct strains are defined as **congenic** if they differ for the alleles at one single locus.
- Generando ceppi congenici di topi che rigettavano trapianti da altri ceppi, ma che erano identici per tutti gli altri geni, i ricercatori hanno dimostrato che una singola regione genica collocata sul cromosoma 17 è responsabile del rigetto rapido dei trapianti di tessuto.
- Questa regione è stata chiamata *locus maggiore di istocompatibilità.*

MHC genes and proteins





POLIMORFISMO DELLE MOLECOLE MHC

•La maggior parte dei geni sono rappresentati da una sequenza unica di DNA in tutti i membri di una specie: tali geni sono definiti **non polimorfi** e la loro sequenza è presente è generalmente presente su entrambi i cromosomi di ogni individuo di quella specie.

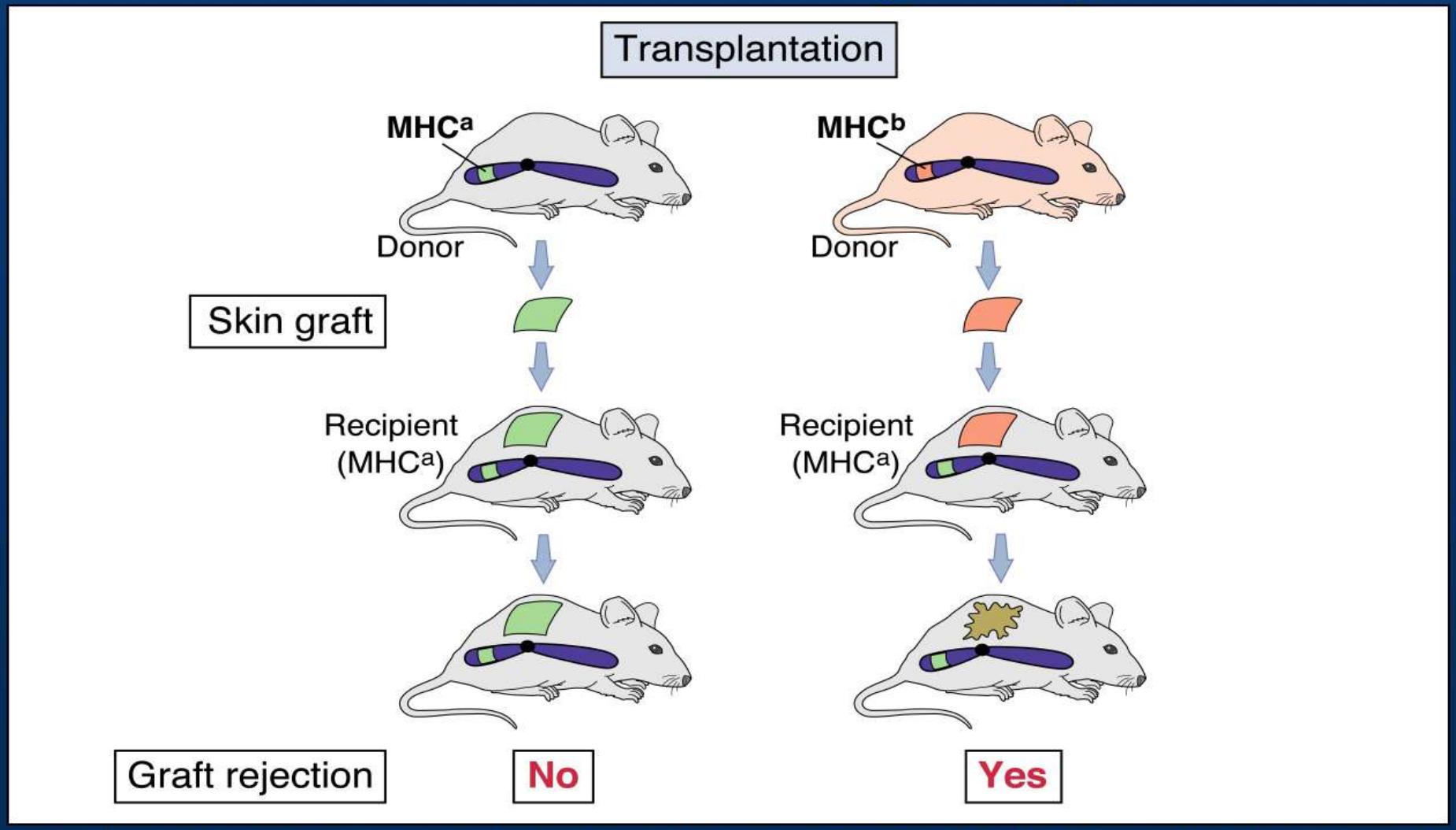
•Un allele ad un locus genetico che abbia una frequenza < 0.01 è denominato "rara variante allelica".

• Un allele con una frequenza ≥ 0.01 (ma < 1) è definito come **polimorfico**".

• Gli alleli ai loci MHC sono polimorfici poichè presentano sempre frequenze elevate nella popolazione.

• Il polimorfismo è concentrato negli esoni che codificano per i domini che legano il peptide e il TCR.

MHC alleles control allograft rejection

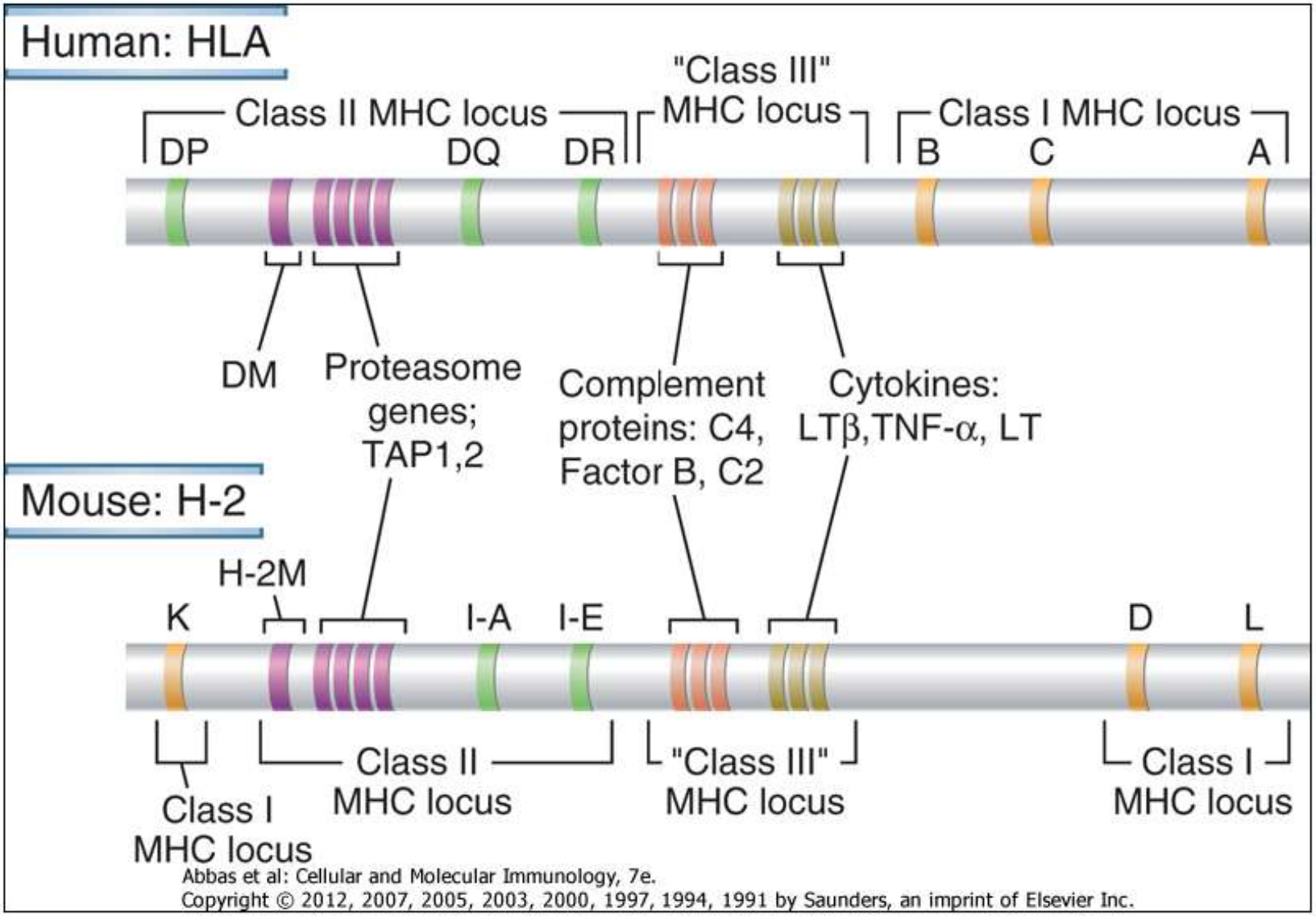


From Abbas, Lichtman, & Pober: Cellular and Molecular Immunology. W.B. Saunders, 1999, Fig. 4-2a

Organizzazione del complesso HLA

- E' localizzato sul cromosoma 6 nell'uomo e sul cromosoma 17 nel topo
- HLA comprende più di 200 geni codificanti
- Il locus contiene i geni che codificano per le proteine HLA «classiche» di I e II classe e le proteine di III classe:

geni HLA di classe I (A, B, C)
geni HLA di classe II (DP, DQ, DR)
geni HLA di classe III (C2, C4, TNF)



Caratteristiche del sistema **HLA**

1. è **poligenico** - il locus comprende circa 200 geni che codificano per le proteine di **I** e **II** classe con una diversa specificità per i peptidi.

2. è **polimorfico** - esistono numerose varianti alleliche per ogni gene.

3. è **codominante** - in ogni individuo sono espressi i prodotti di entrambi gli alleli ereditati dai genitori.

2. HLA E' POLIMORFICO

Le molecole MHC espresse da un individuo non si modificano nel tempo ma possono differire da quelle espresse da un altro individuo della stessa specie

↓

POLIMORFISMO

MHC è uno dei complessi genetici più polimorfici dei vertebrati più evoluti e possiede un numero straordinariamente elevato di alleli diversi per ciascun locus.

Gli alleli differiscono del 5-10% nella sequenza del DNA da un individuo all'altro.

Major Histocompatibility Complex (MHC; Human Leukocyte Antigens [HLA])

Class I , II and III MHC genetic loci (short arm of chromosome 6)

Major Class I genes: ***HLA-A, B, C***
Minor Class I-like genes: *HLA-E, F, G, H, J, X*

Major Class II genes: ***HLA-D* region**
DP*, *DQ*, *DR

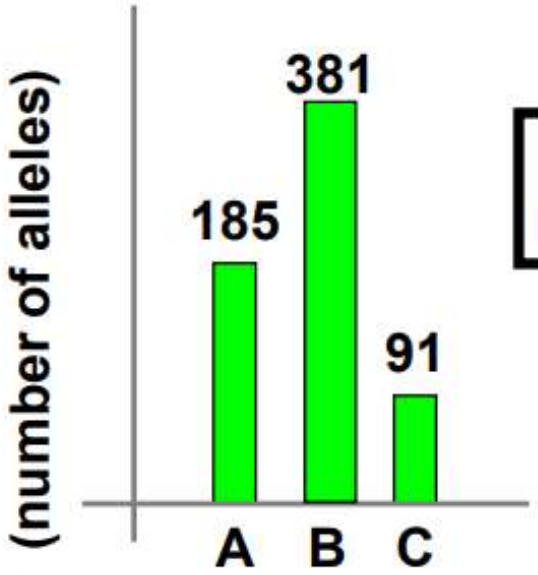
Major Class III genes: ***Diverse (non-antigen presenting functions)***

MHC loci and numbers of alleles:

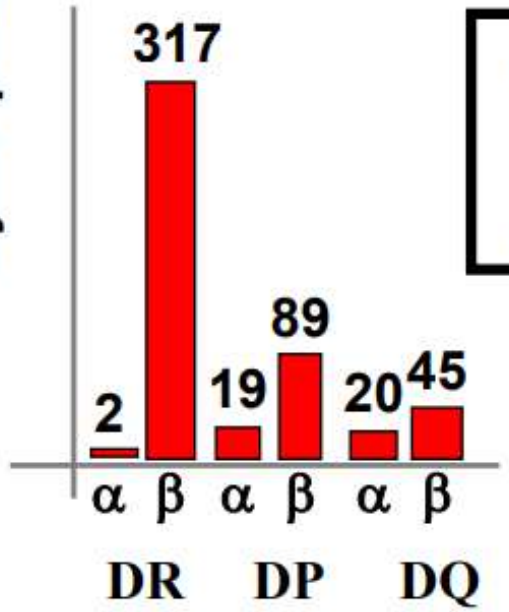
HLA Class I										
Gene	A	B	C	E	F	G	H	J	K	L
Alleles	489	830	266	9	21	23	12	3	6	5
Proteins	390	711	210	3	4	6	0	0	0	0
Nulls	37	28	6	0	0	1	0	0	0	0

HLA Class II										
Gene	DRA	DRB	DQA1	DQB1	DPA1	DPB1	DMA	DMB	DOA	DOB
Alleles	3	545	34	78	23	125	4	7	12	9
Proteins	2	451	25	57	14	112	4	7	3	4
Nulls	0	7	1	1	0	2	0	0	1	0

DISTRIBUZIONE DEGLI ALLELI MHC NELLA POPOLAZIONE UMANA



Class I
657 alleles



Class II
492
alleles

- Gli alleli non sono distribuite in modo uniforme nella popolazione

- Gli alleli sono segregati **nelle popolazioni**

SELEZIONE

HLA e ereditarietà

Aplotipo

Ogni individuo possiede nel proprio patrimonio genetico due copie di geni di ciascun locus HLA, uno di origine paterna e uno di origine materna.

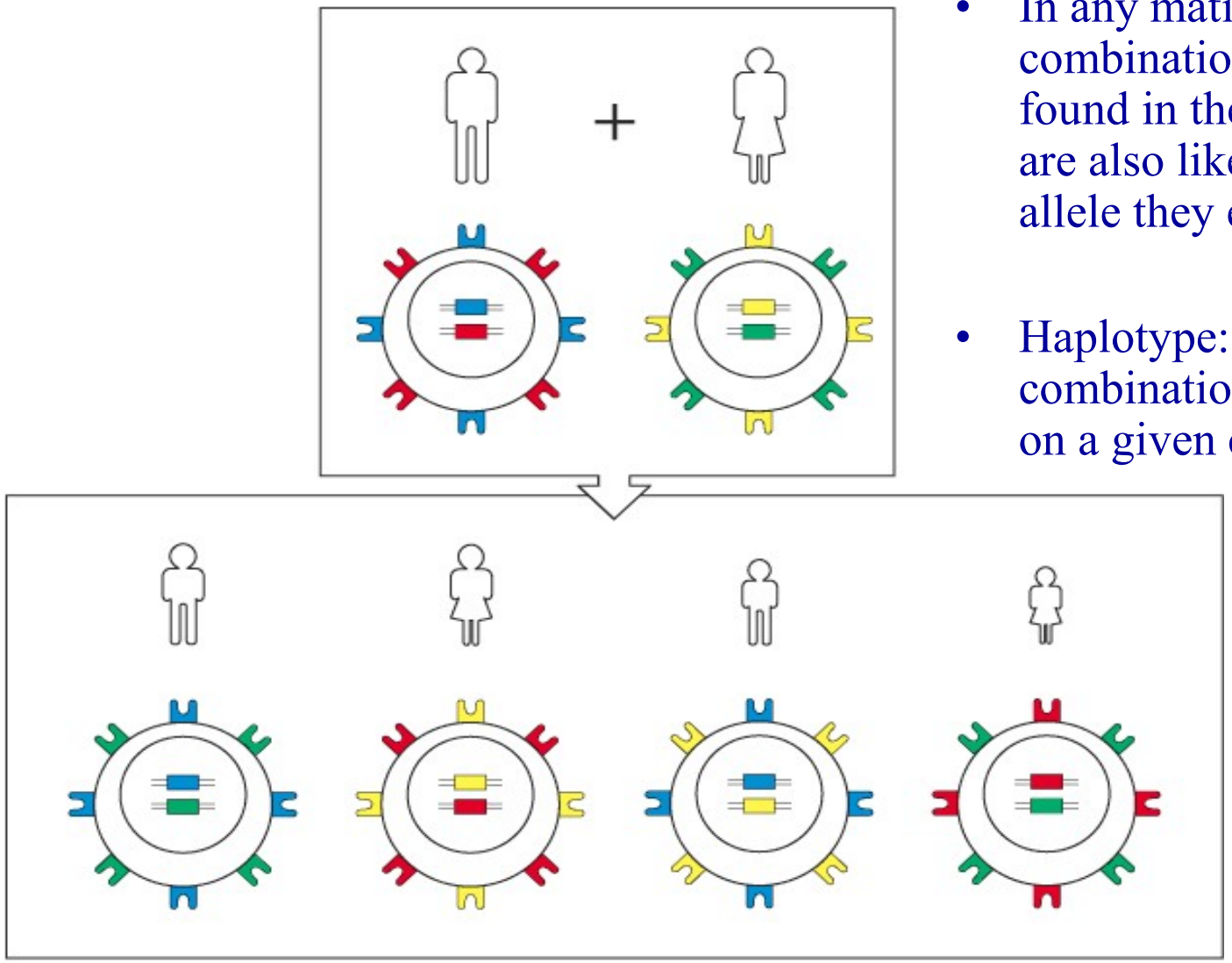
Il set di geni continui ereditato dai genitori viene definito **aplotipo**.

Daddy

A*0101 B*0801 C*0701 DRB1*0301 DQB1*0201

A*0302 B*3501 C*0401 DRB1*0401 DQB1*0301

Mom



- In any mating, four possible combinations of haplotypes can be found in the offspring; thus siblings are also likely to differ in the MHC allele they express.

- Haplotype: The particular combination of MHC alleles found on a given chromosome 6.

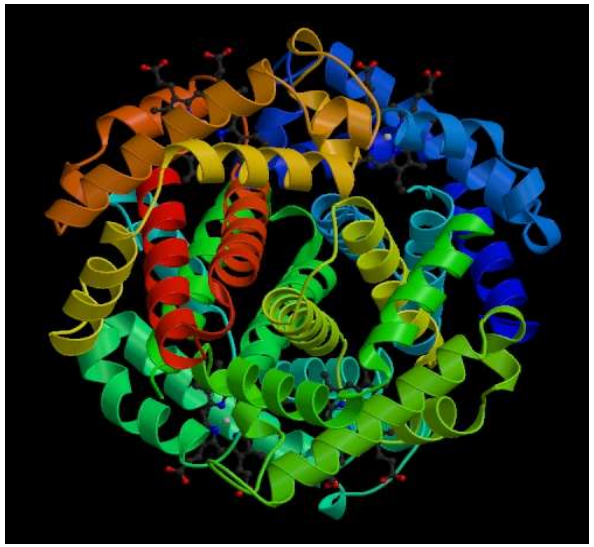
Important aspects of MHC

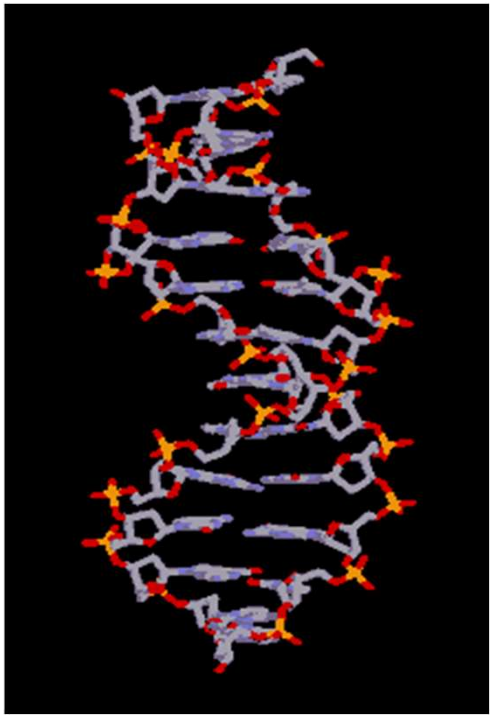
- Individuals have a limited number of MHC alleles for each class
- High polymorphism in MHC for a species
- Alleles for MHC genes are co-dominant
  - Each MHC gene product is expressed on surface of individual cell

MHC: things to remember

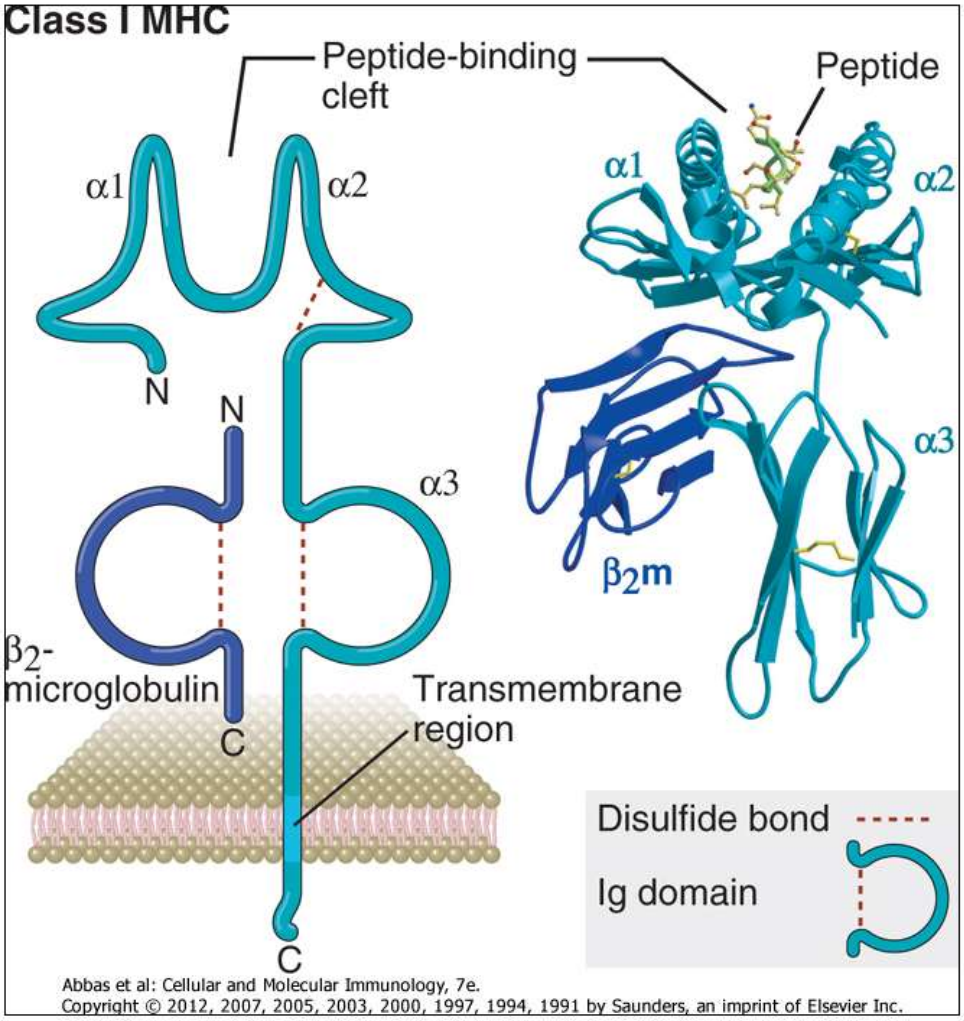
- MHC molecules in humans is also called HLA (human leukocyte antigen)
- Class I and II locus.
- HLA-DR alpha chain is monomorphic
- HLA-DRB1 is most polymorphic in MHC II genes
- HLA-DRB1 is always present in any individual
- HLA-DRB3/4/5 is present in some but not all people.
- A heterozygote person (most people) expresses two haplotypes.
- **A person can express 3-6 class I and 3-8 class II isoforms.**
- **2406 possible class II isoforms in the human population.**
- **753 MHC I isoforms in the human population.**

MHC: protein structure

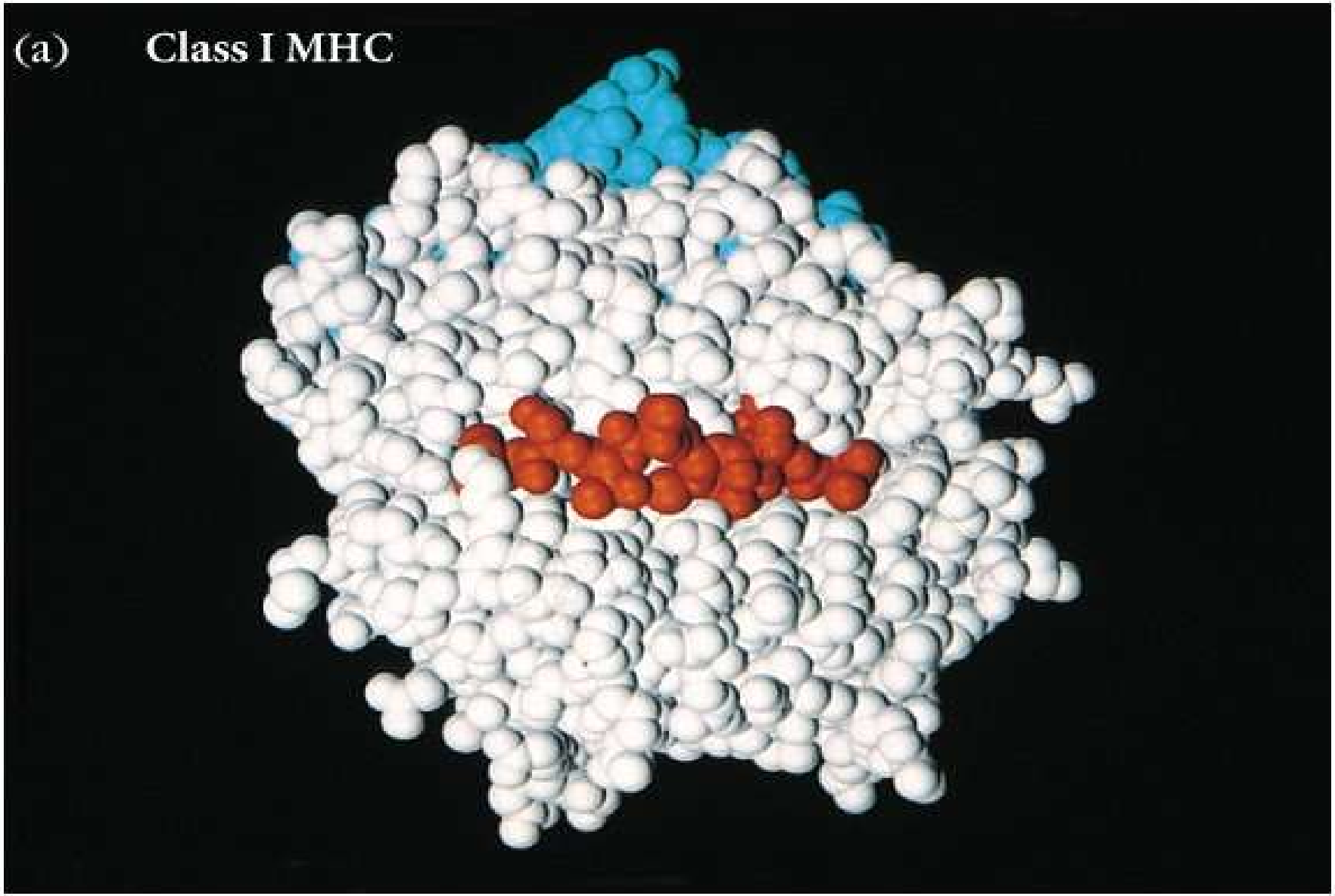




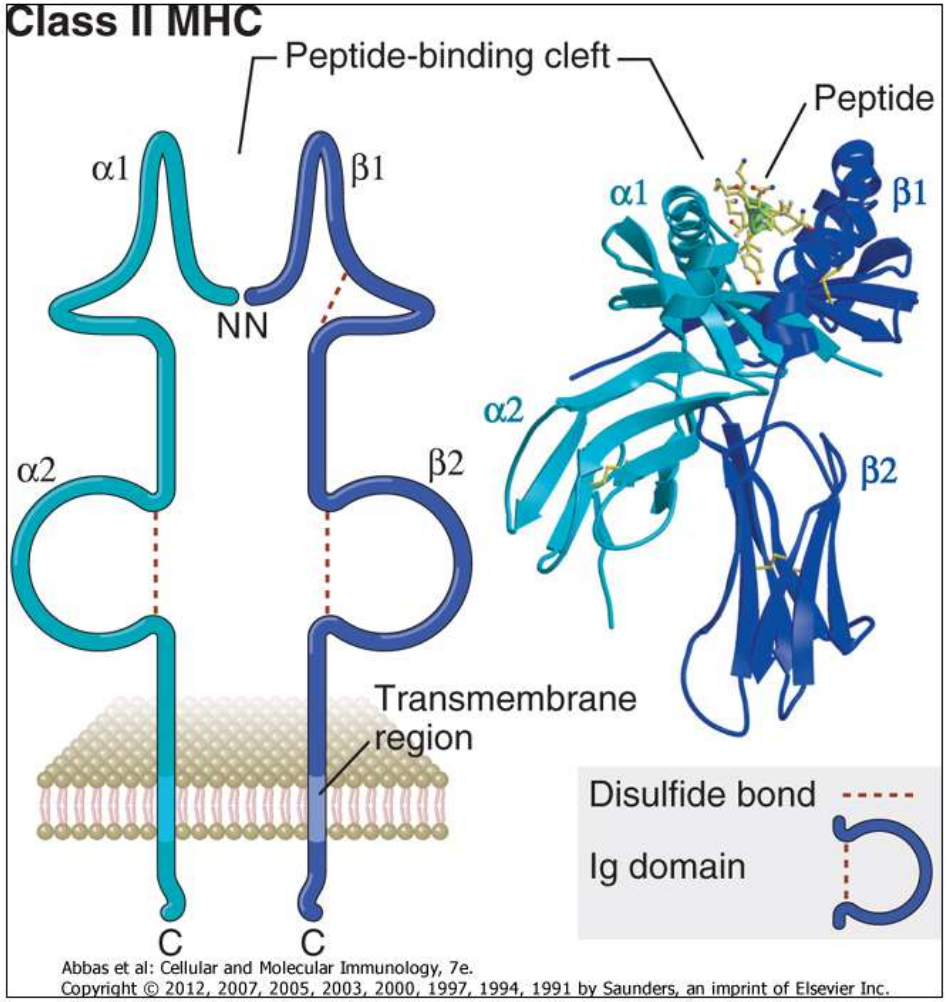
Structure of a Class I MHC molecule



Class I:Peptide Binding



Structure of a Class II MHC molecule



The peptide-binding groove of MHC molecules

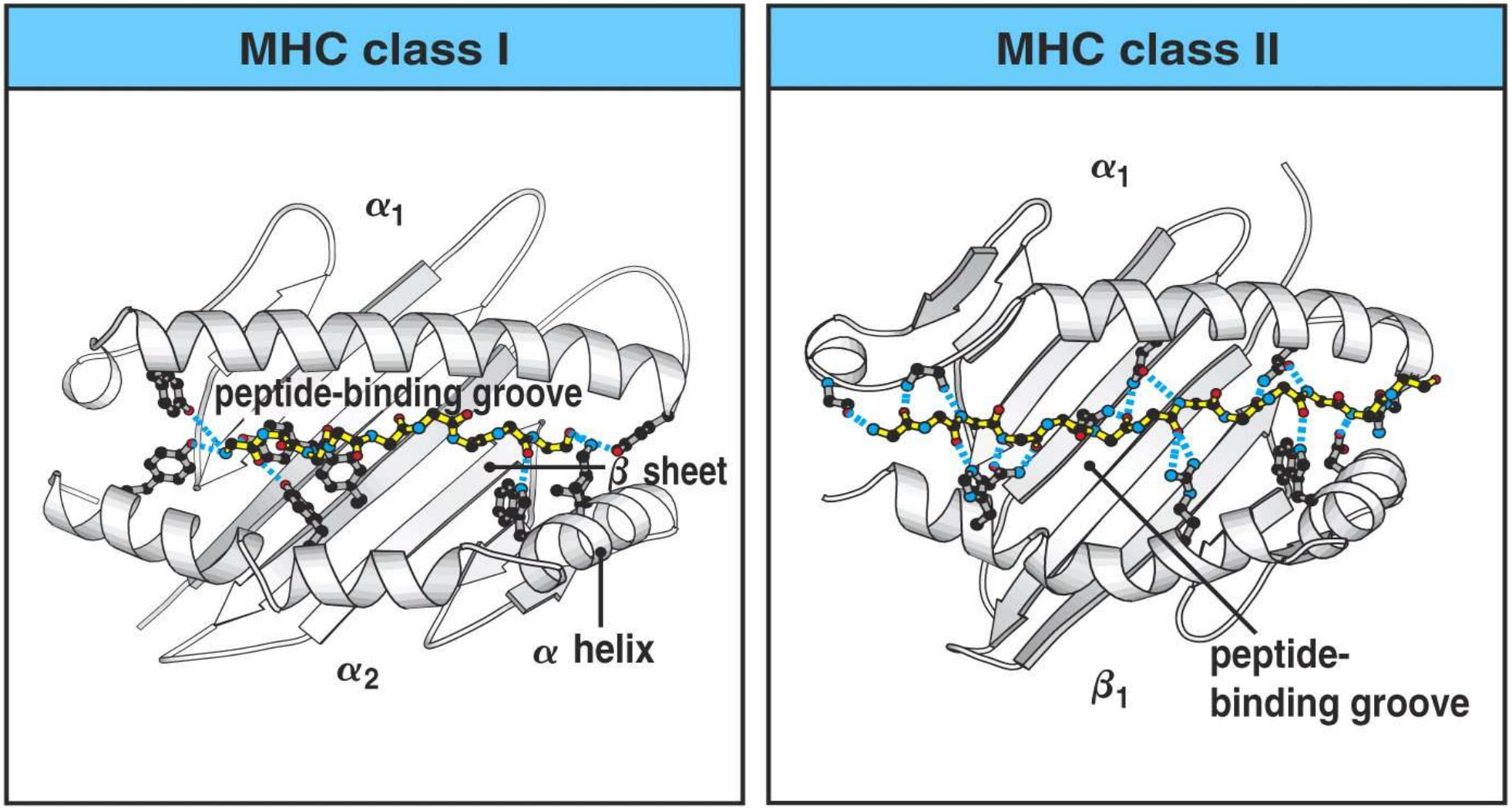


Figure 3-15 The Immune System, 2/e (© Garland Science 2005)

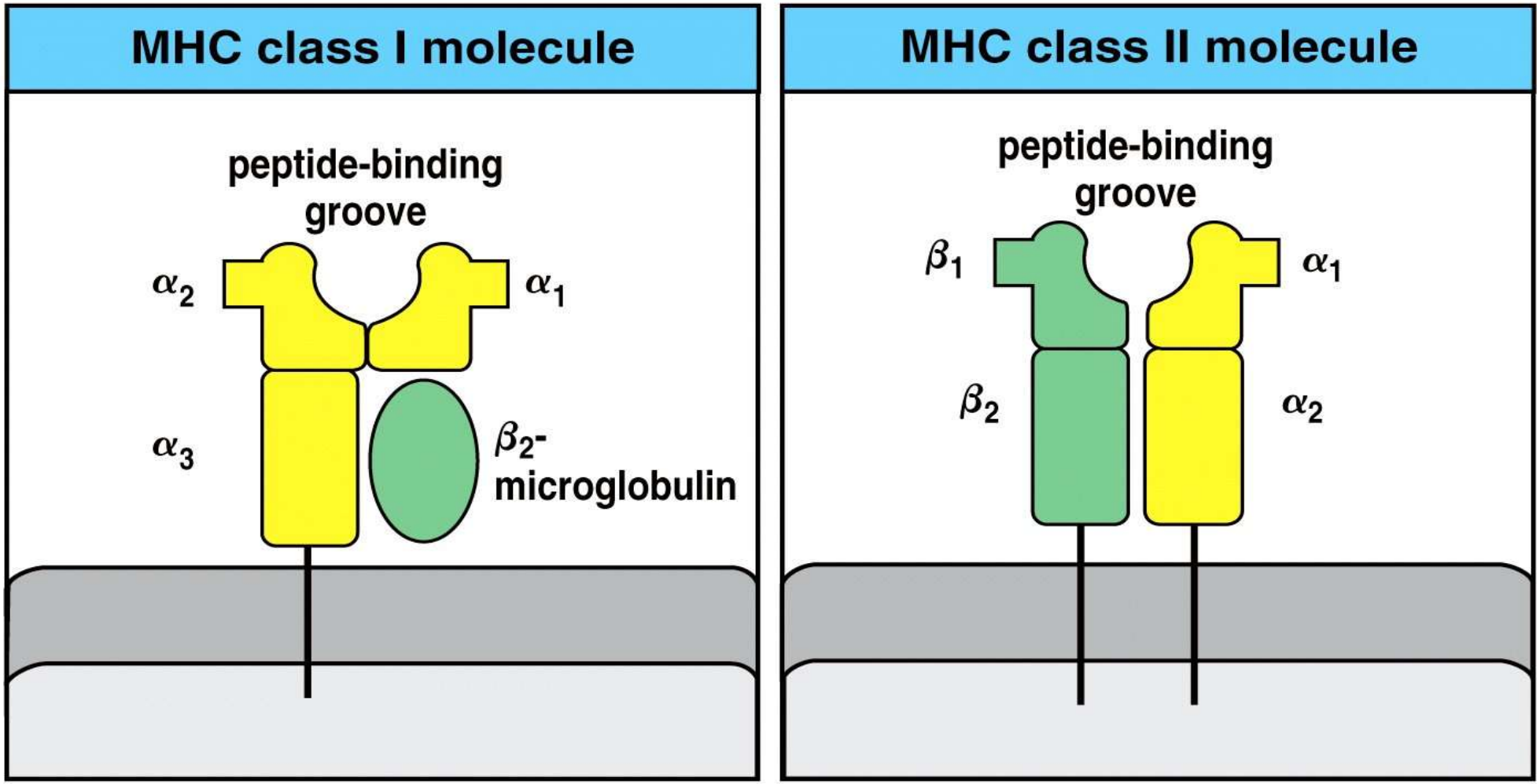


Figure 3-13 part 1 of 2 The Immune System, 2/e (© Garland Science 2005)

Important aspects of MHC

- Each MHC has ONE peptide binding site
  - But each MHC can bind many different peptides
  - Only one at a time
  - Peptide binding is "degenerate"
- MHC polymorphism is determined in germline
  - NO recombination mechanisms for creating diversity in MHC
- Peptide must bind with individual's MHC to induce immune response

Table 4–1. Features of Class I and Class II MHC Molecules

Feature	Class I MHC	Class II MHC
Polypeptide chains	α (44-47 kD) β_2-Microglobulin (12 kD)	α (32-34 kD) β (29-32 kD)
Locations of polymorphic residues	α1 and α2 domains	α1 and β1 domains
Binding site for T cell coreceptor	α3 region binds CD8	β2 region binds CD4
Size of peptide-binding cleft	Accommodates peptides of 8-11 residues	Accommodates peptides of 10-30 residues or more
Nomenclature		
Human	HLA-A, HLA-B, HLA-C	HLA-DR, HLA-DQ, HLA-DP
Mouse	H-2K, H-2D, H-2L	I-A, I-E

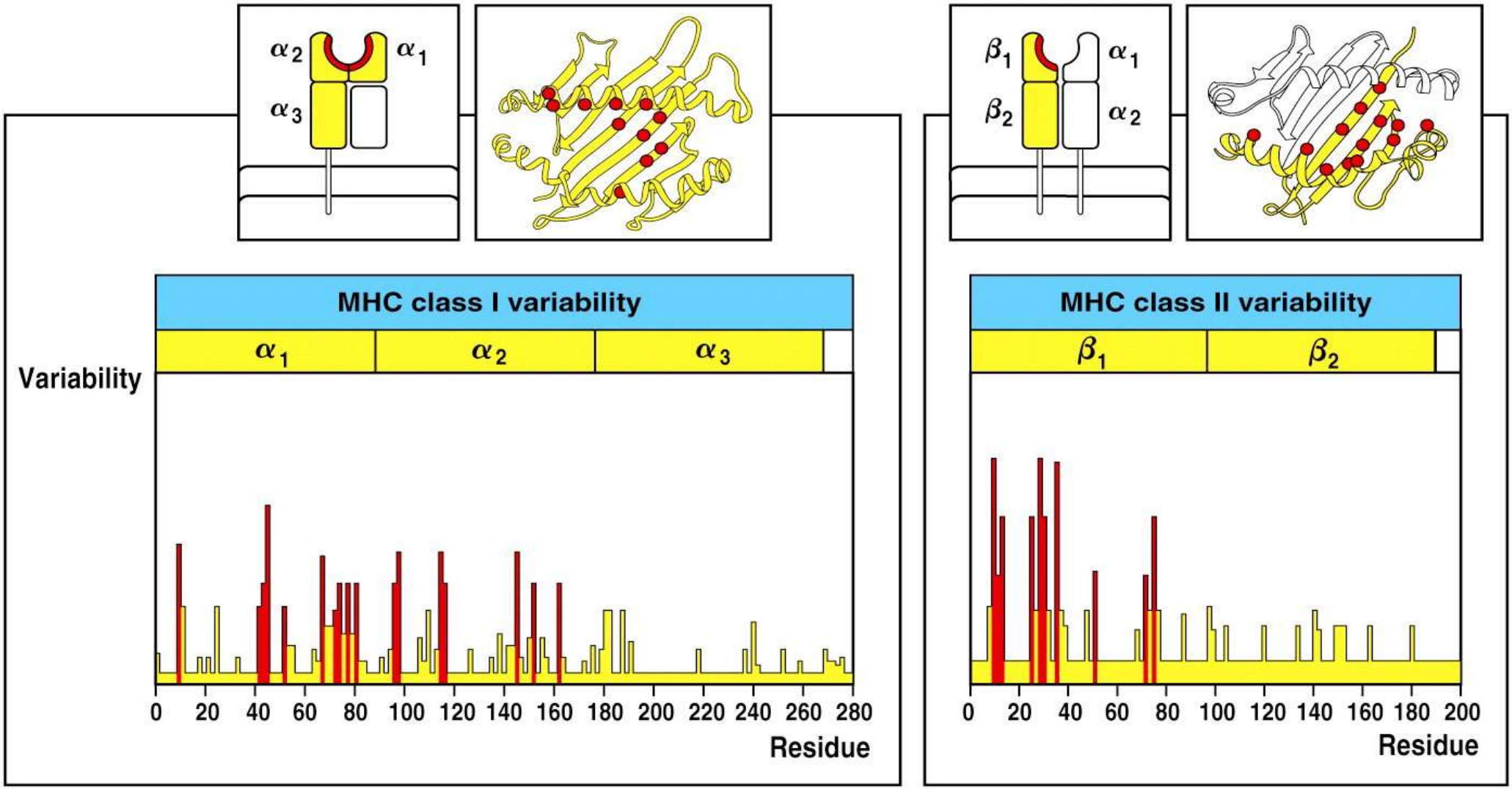


Figure 5-16 Immunobiology, 6/e. (© Garland Science 2005)

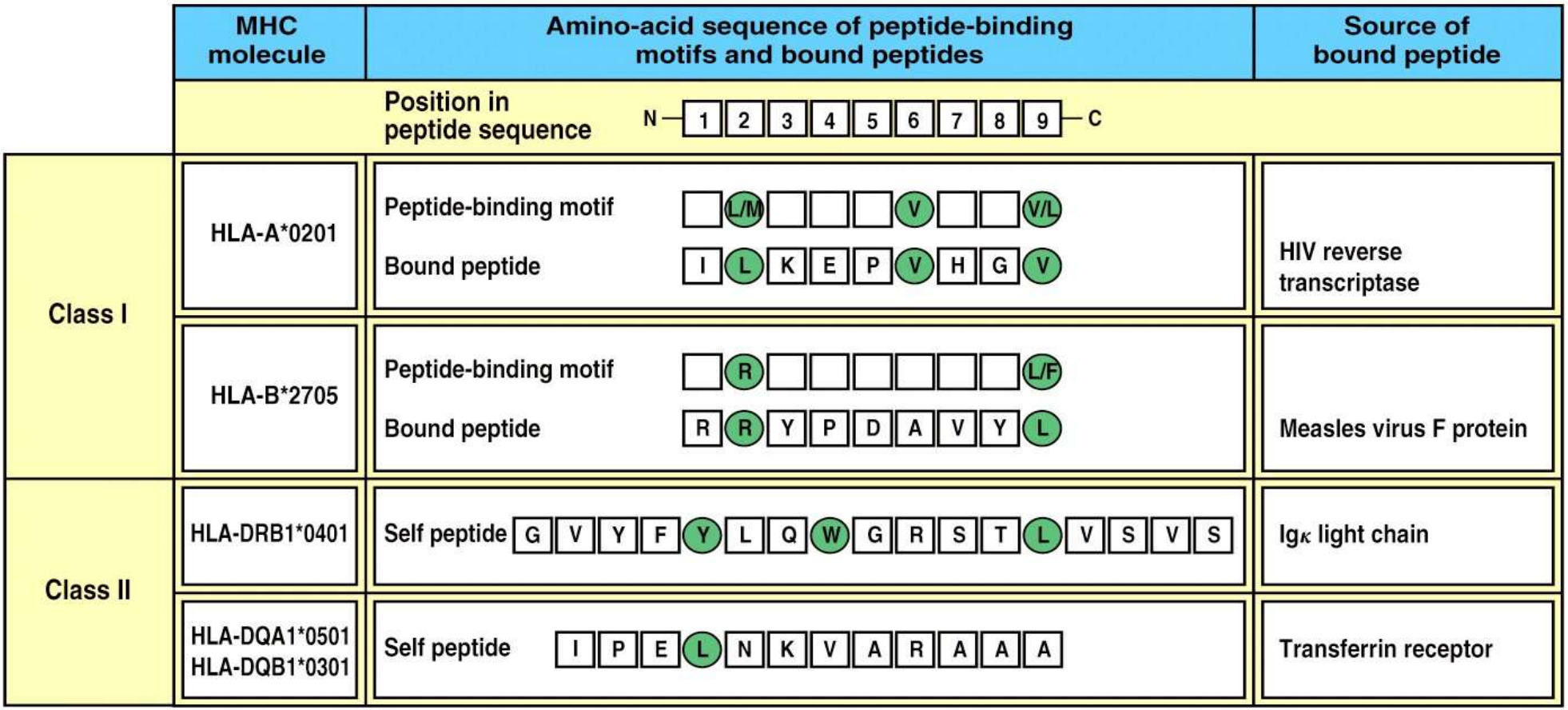


Figure 3-29 The Immune System, 2/e (© Garland Science 2005)

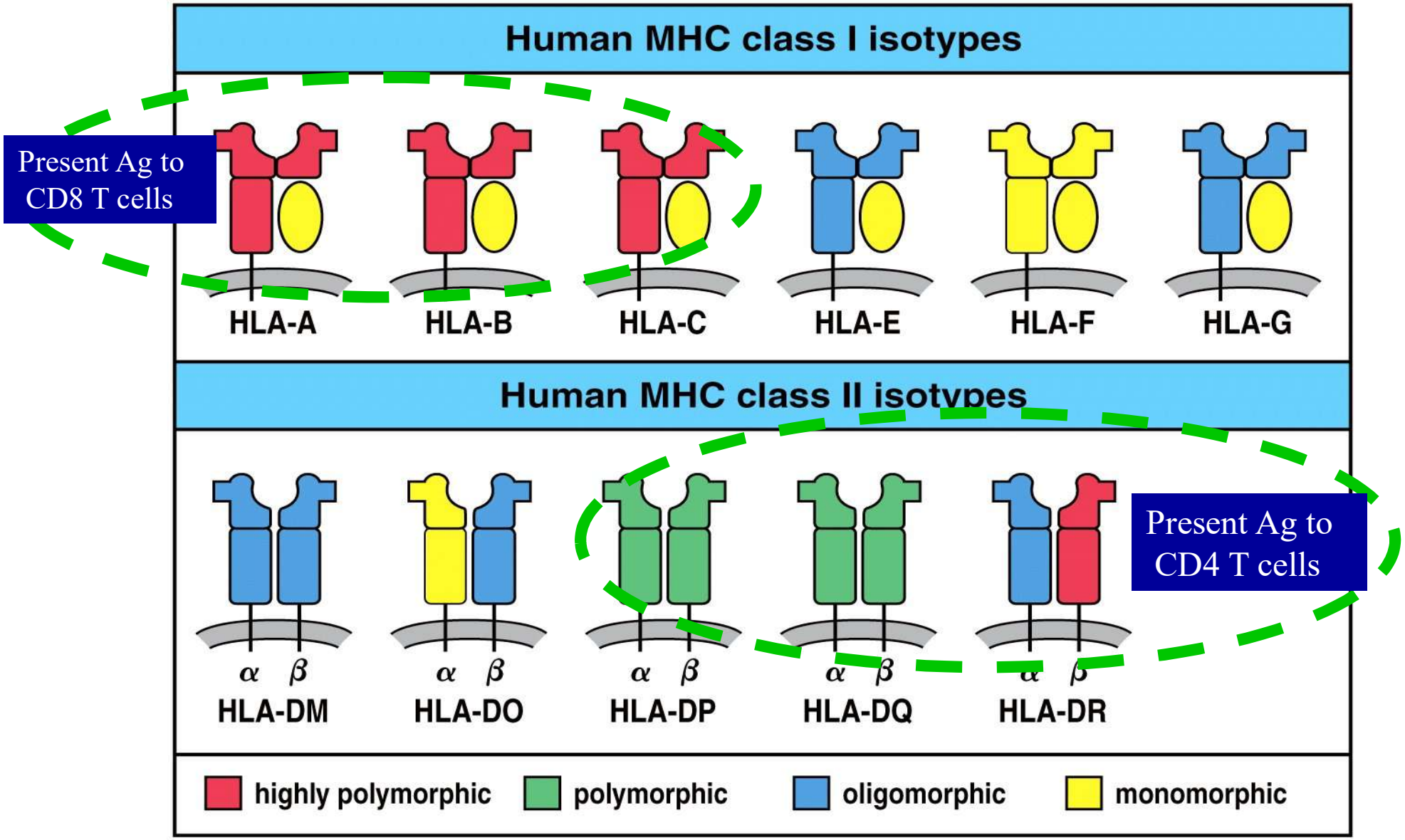


Figure 3-23 The Immune System, 2/e (© Garland Science 2005)

Different cell distribution of MHC class I and II

Tissue	MHC class I	MHC class II
Lymphoid tissues		
T cells	+++	+*
B cells	+++	+++
Macrophages	+++	++
Other antigen-presenting cells (e.g., dendritic cells)	+++	+++
Epithelial cells of the thymus	+	+++
Other nucleated cells		
Neutrophils	+++	–
Hepatocytes	+	–
Kidney	+	–
Brain	+	– †
Non-nucleated cells		
Red blood cells	–	–

Figure 3-22 The Immune System, 2/e (© Garland Science 2005)

- IFN-γ increases the expression of MHC II in APC and induces the expression in non-APC cells at sites of infection

MHC & Antigens

- **MHC Class I**
  - present *endogenously* derived peptides.
  - these can be either self or derived from viruses
  - because MHC Class I is present on all cells any cell can interact with T cells if infected by a virus
- **MHC Class II**
  - present *exogenous* antigen which has been phagocytosed and processed.eg. Bacteria
  - This is performed by professional antigen presenting cells eg macrophages

Peptide Binding by Major Histocompatibility Complex (MHC) Antigen-presenting Proteins

MHC I

- Peptides of intracellular origin
- Peptides 9-10 residues long
- Deep pockets bind peptide side-chains
- Deep pockets bind peptide N- and C-termini

MHC II

- Peptides of extracellular origin
- Peptides 15 residues or longer
- Shallow pockets bind peptide sidechains
- Peptide termini free
- H-bonds to peptide backbone