Histocompatibility, State of the Art testing and their Application in Transplantation

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Nothing to disclose

• Applications of HLA Testing in Solid Organ Transplants

- Types of transplants
- Sensitization, Epitopes (Eplets)
- Virtual vs. Physical Crossmatch
- Desensitization

Organ Transplantation

- Kidney
- Pancreas
- Heart
- Liver
- Intestine
- Skin
- Uterus
- Composite Tissue (Face, Limbs)
- Parathyroid
- cornea

First Organ Transplantation

- First successful Kidney transplant-In Boston, in 1959 from fraternal twins and it functioned for 20 years without immunosuppression drugs.
- First successful Liver transplant- In Denver on 7/23/1967
- First successful Heart transplant- In Cape Town, South Africa on 1/2/68
- First successful Uterus-In University of Gothenburg 2012-First baby born in 2014
- First Successful Face-In France in 2005 (survived 6 years); First Limb Transplant-In US in 1999

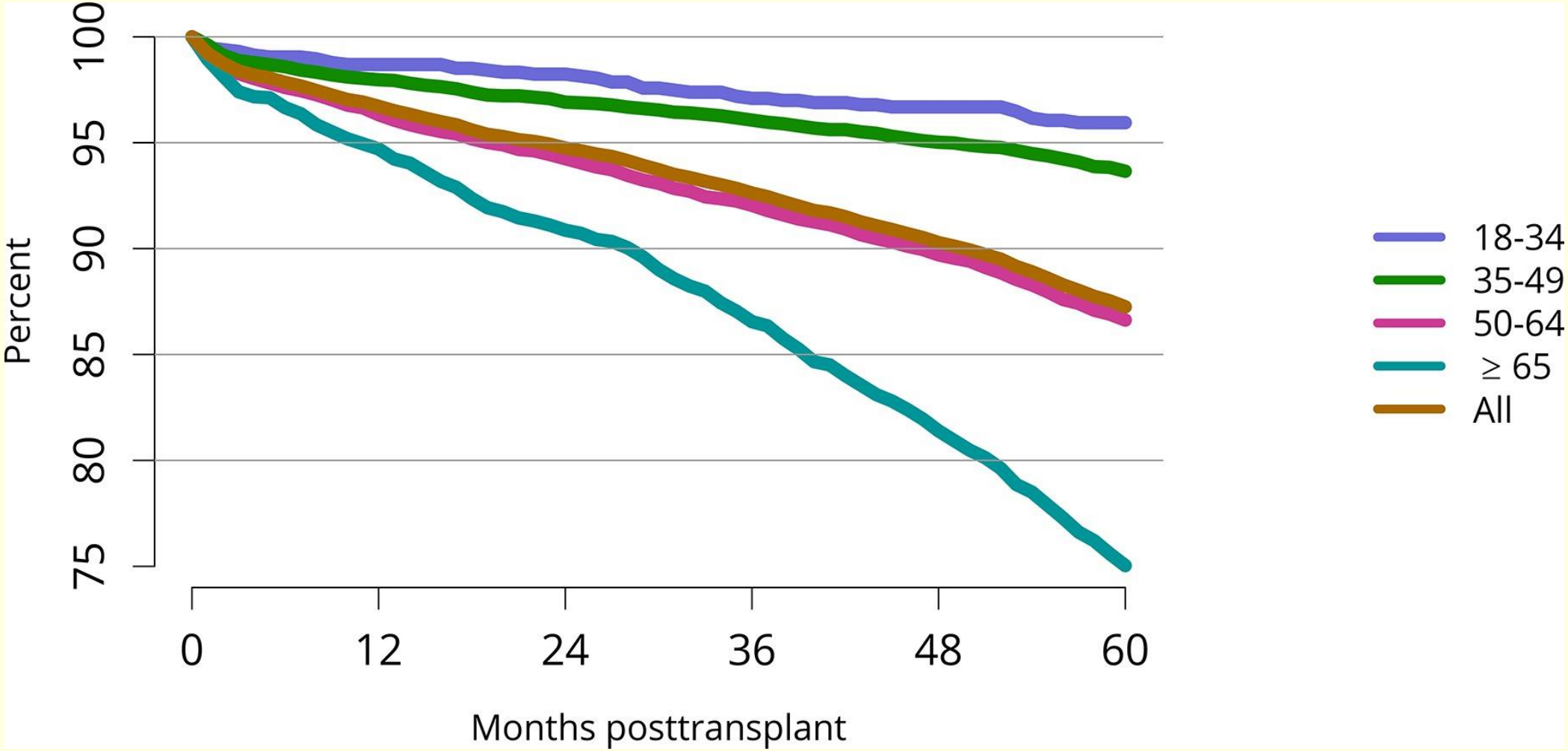
Statistics on Organ Transplantation

- There are more than 114,000+ people on the organ transplantation waiting list.
- There are 94,000+ people waiting for a Kidney, 14,000 waiting for a liver and 4,000 waiting for a heart transplant.
- Each day >90 people receive an organ transplantation, but 18 people on the waiting list die because a donor is not available.

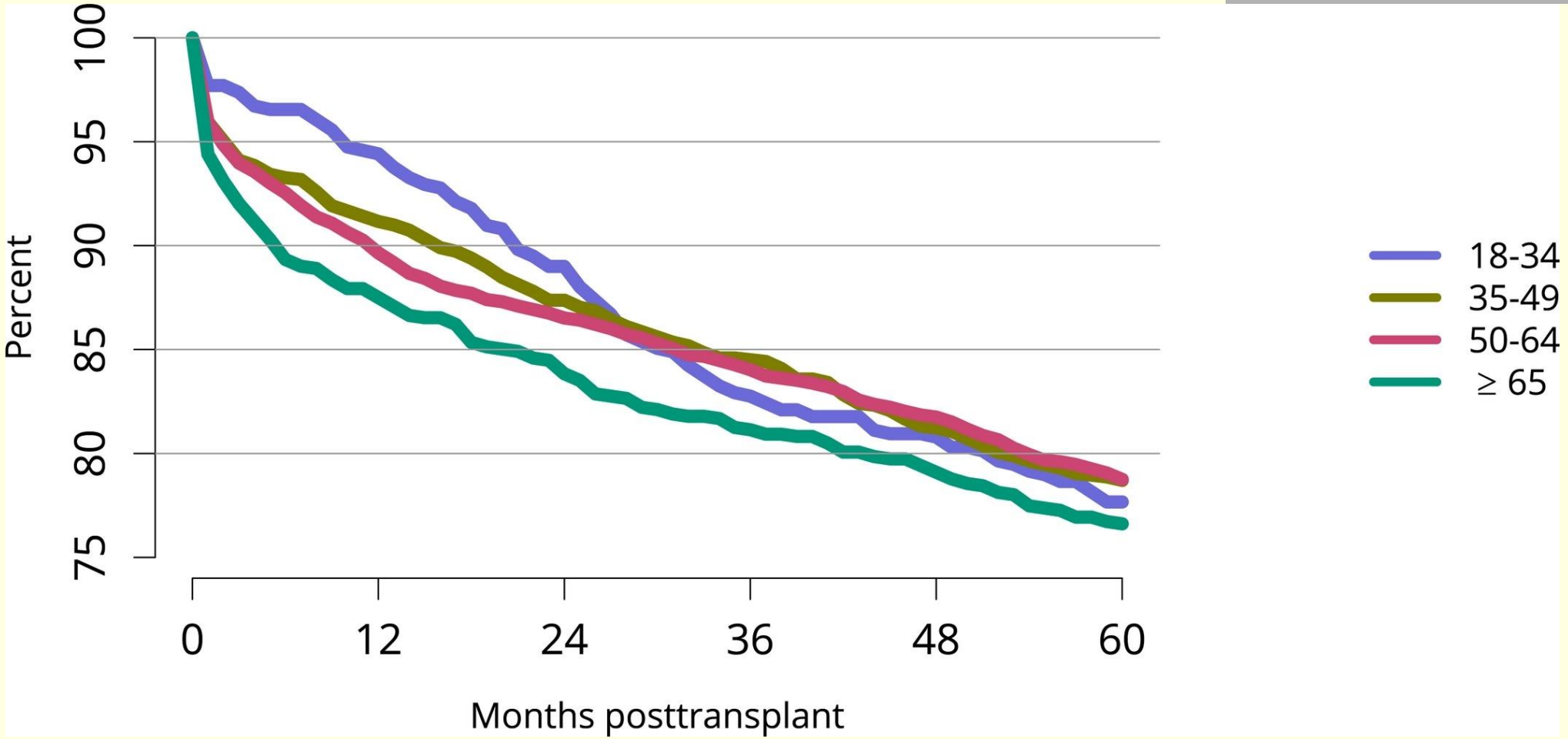
Outcome

- Some transplants have great outcome of up to 30 years, but majority are rejected earlier.
- All attempts are to have lifetime survivor.
- One of the main reason is due to sensitization to HLA antigens expressed on the cell surface of allograft.

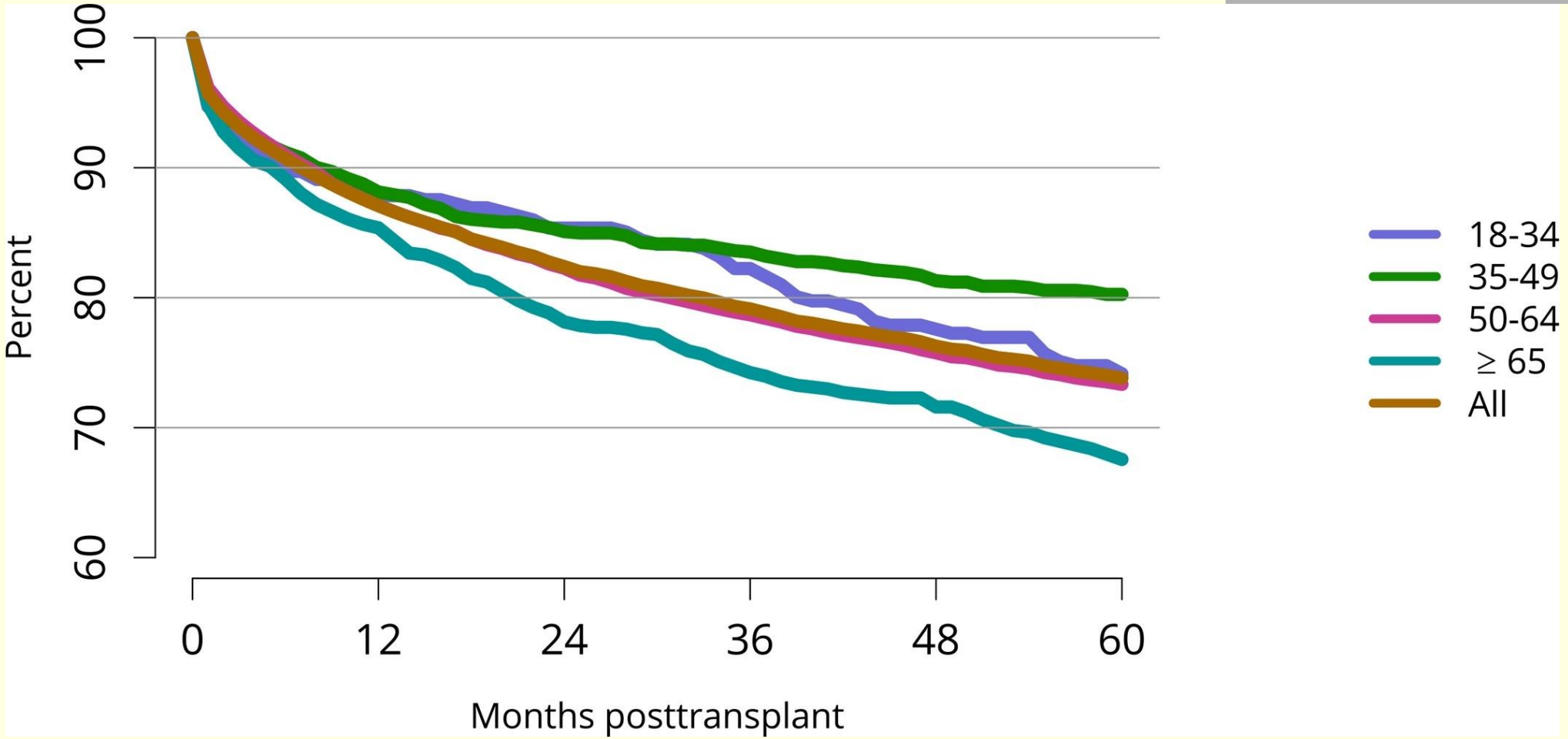
Patient survival among adult deceased donor kidney transplant recipients



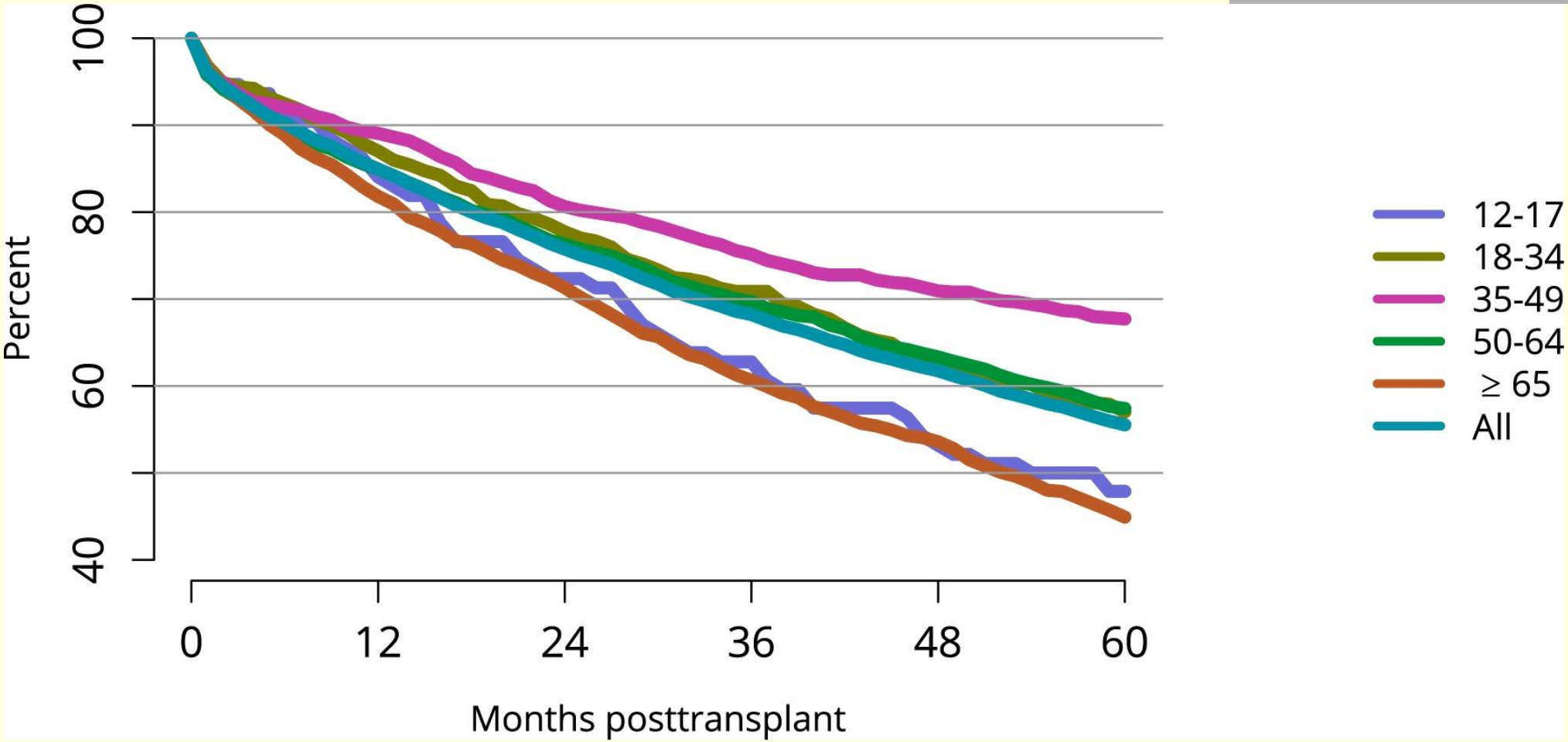
Patient survival among adult heart transplant recipients



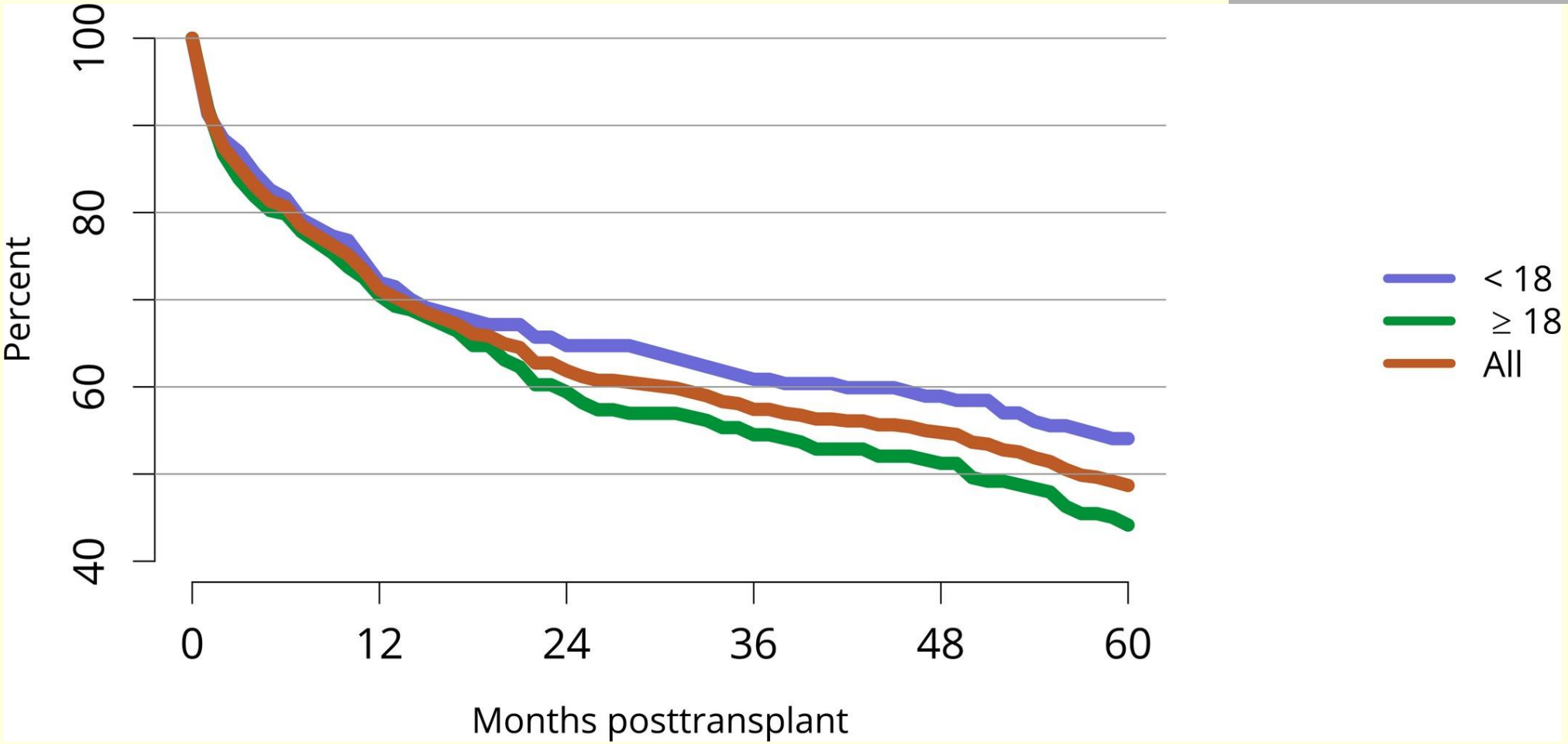
Graft survival among adult liver transplant recipients

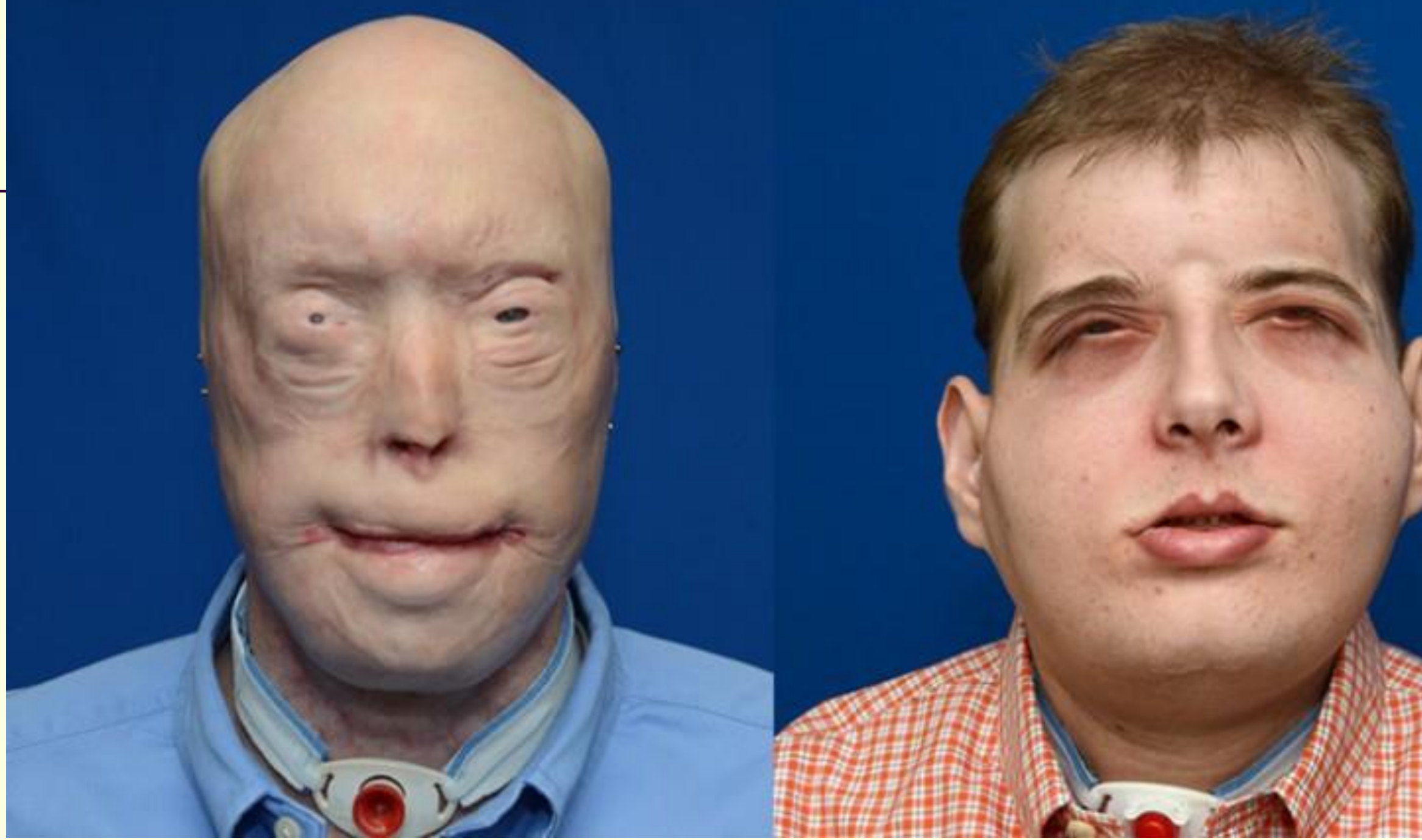


Patient survival among lung transplant recipients



Graft survival among intestine transplant recipients



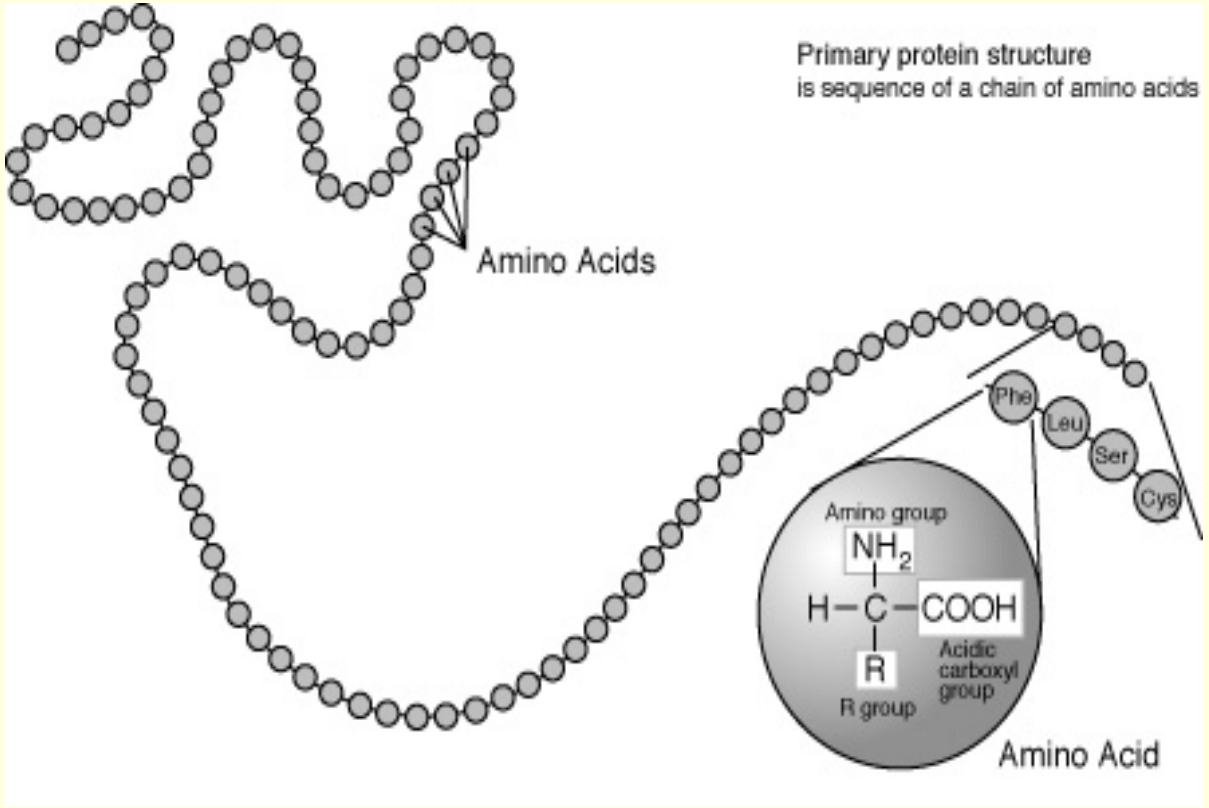


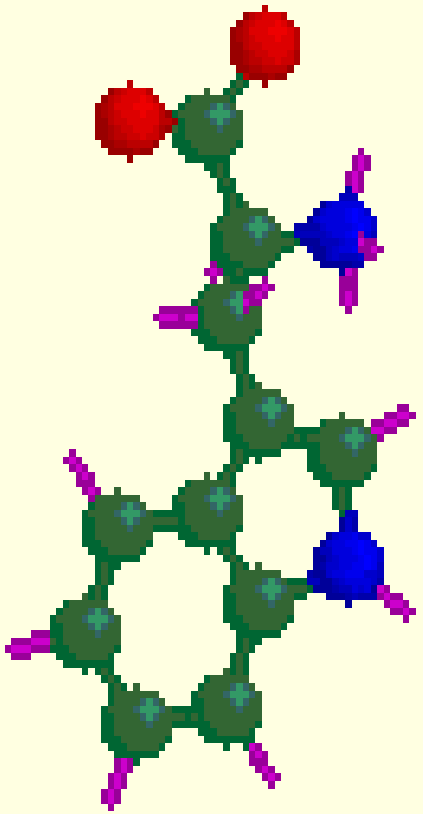
Causes of Graft Failure

	N	%
GRAFT FAILURE CAUSE		
Hyperacute Rejection	12	0.5
Acute Rejection	378	16.4
Primary Failure	160	7.0
Graft Thrombosis	169	7.4
Infection	87	3.8
Surgical Complications	6	0.3
Urological Complications	25	1.1
Recurrent Disease	97	4.2
Chronic Rejection	839	36.5
BK (Polyoma) Virus	20	0.9
Other, Specify	506	22.0
All	2299	100.0

HLA Polymorphism

- HLA system is highly polymorphic.
- Started as one non-polymorphic peptide and mutation during evolution, made them very polymorphic (survival mechanism)
- Not one antigen, but many epitopes or eplets


Primary protein structure
is sequence of a chain of amino acids
Amino Acids
Phe
Leu
Ser
Cys
Amino group
NH_2
H – C – COOH
R
Acidic carboxyl group
R group
Amino Acid



HLA class I & class II

side view

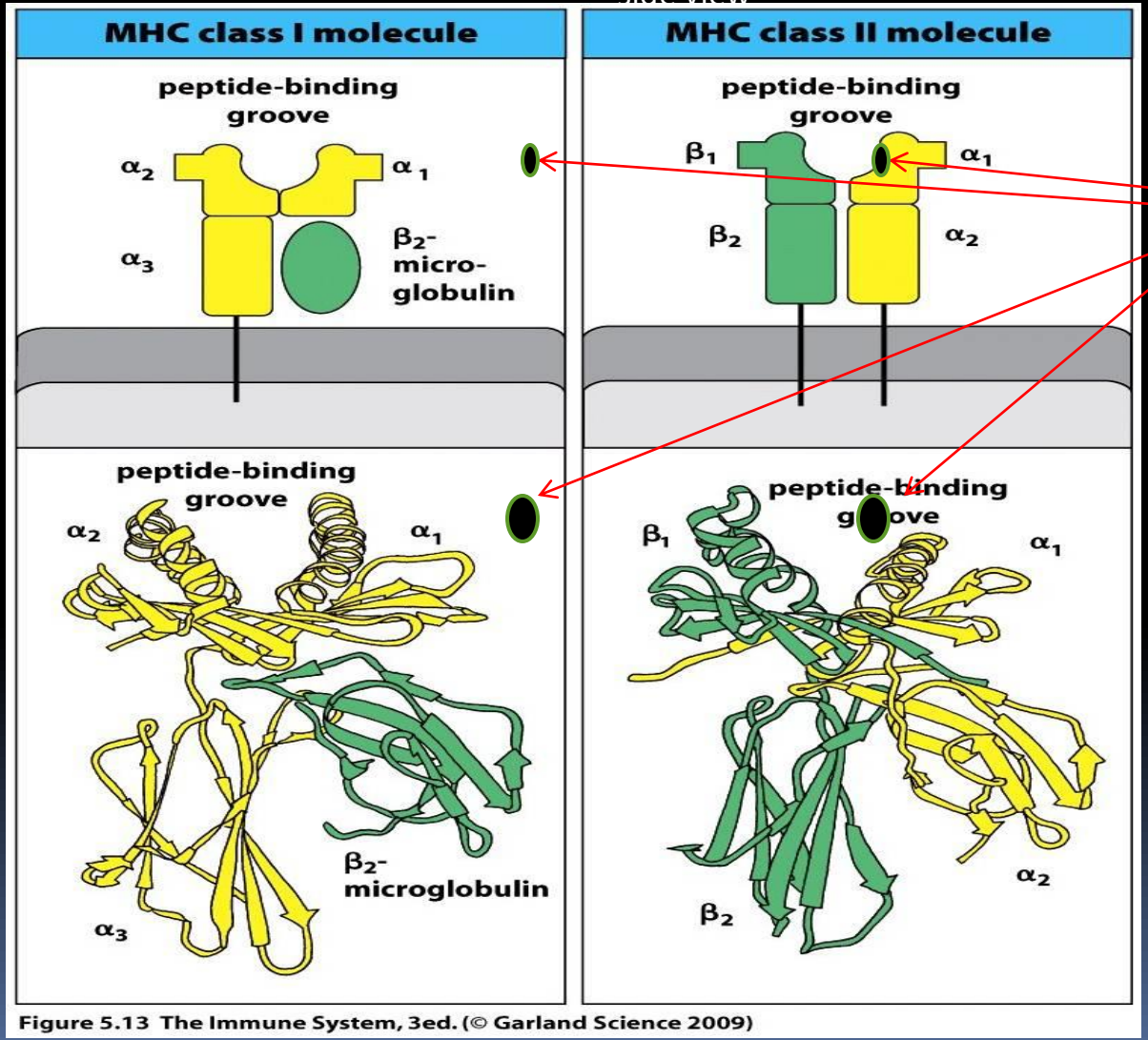
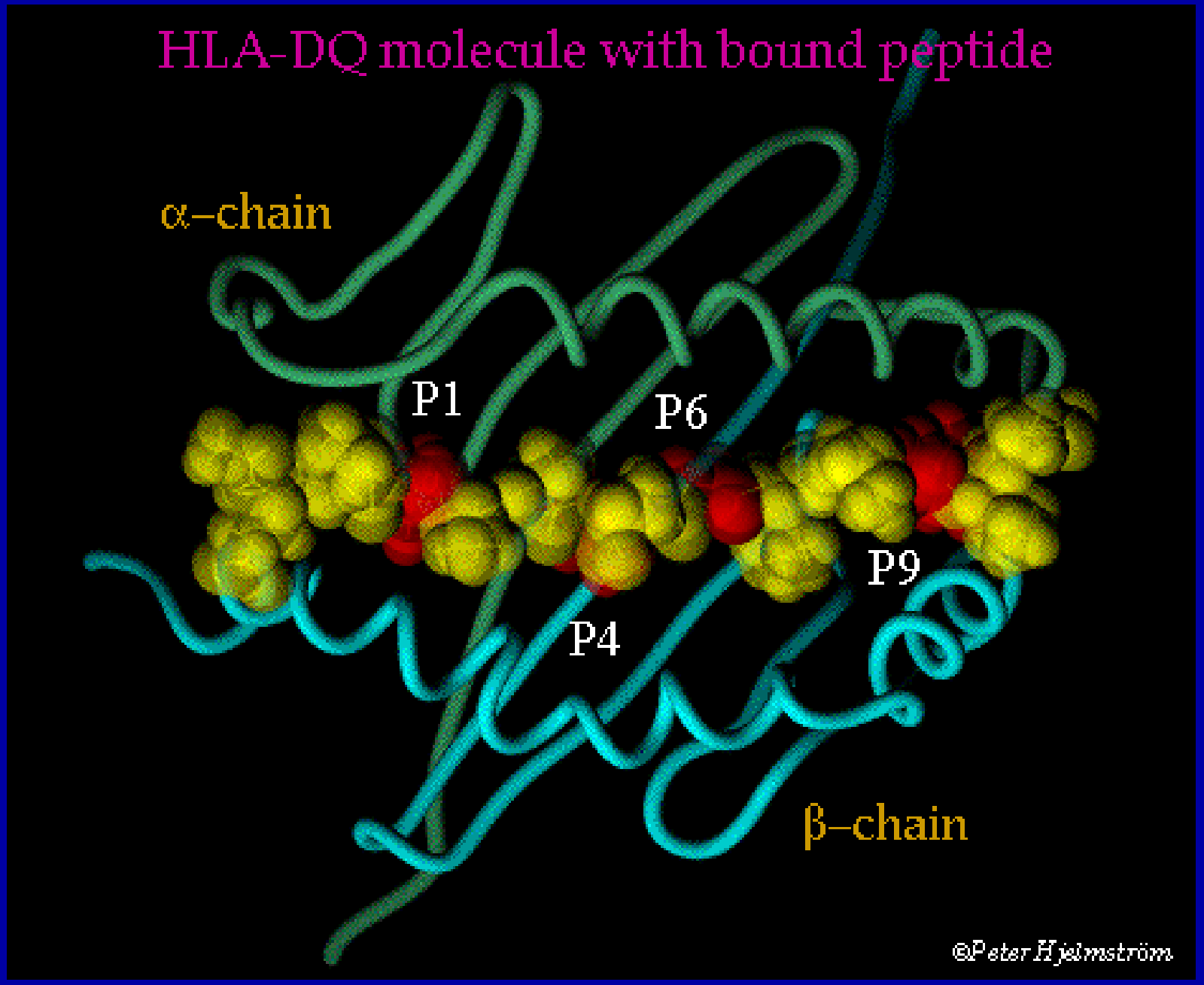


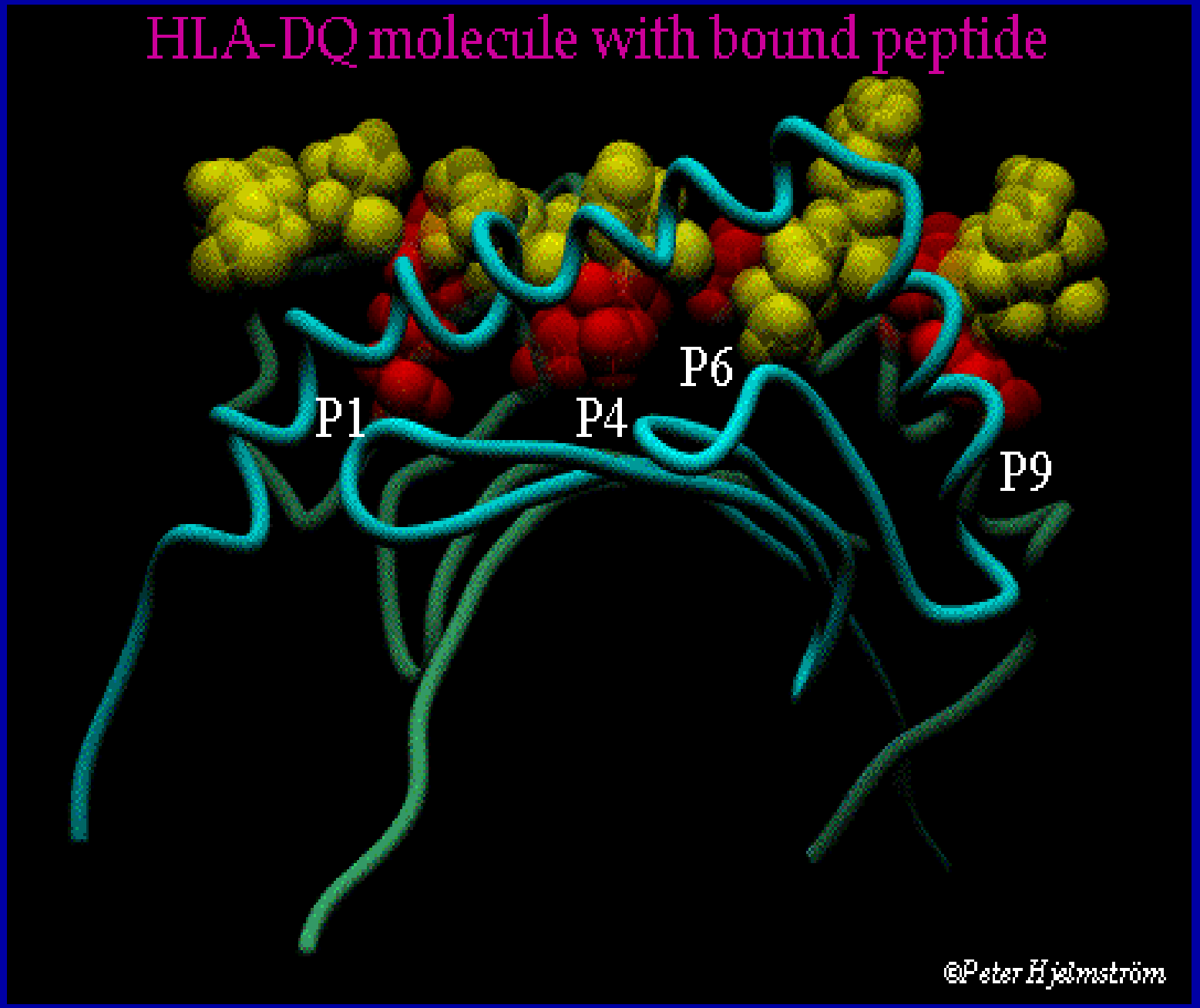
Figure 5.13 The Immune System, 3ed. (© Garland Science 2009)

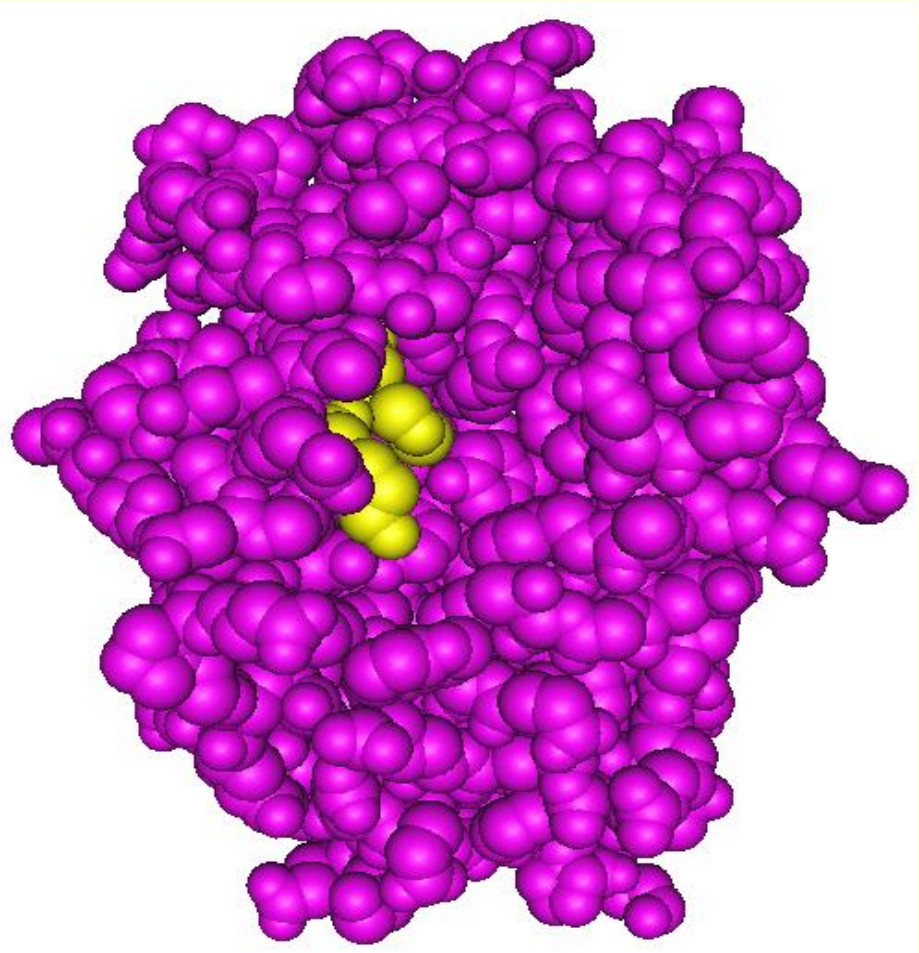
Peptide presented in the Peptide binding groove

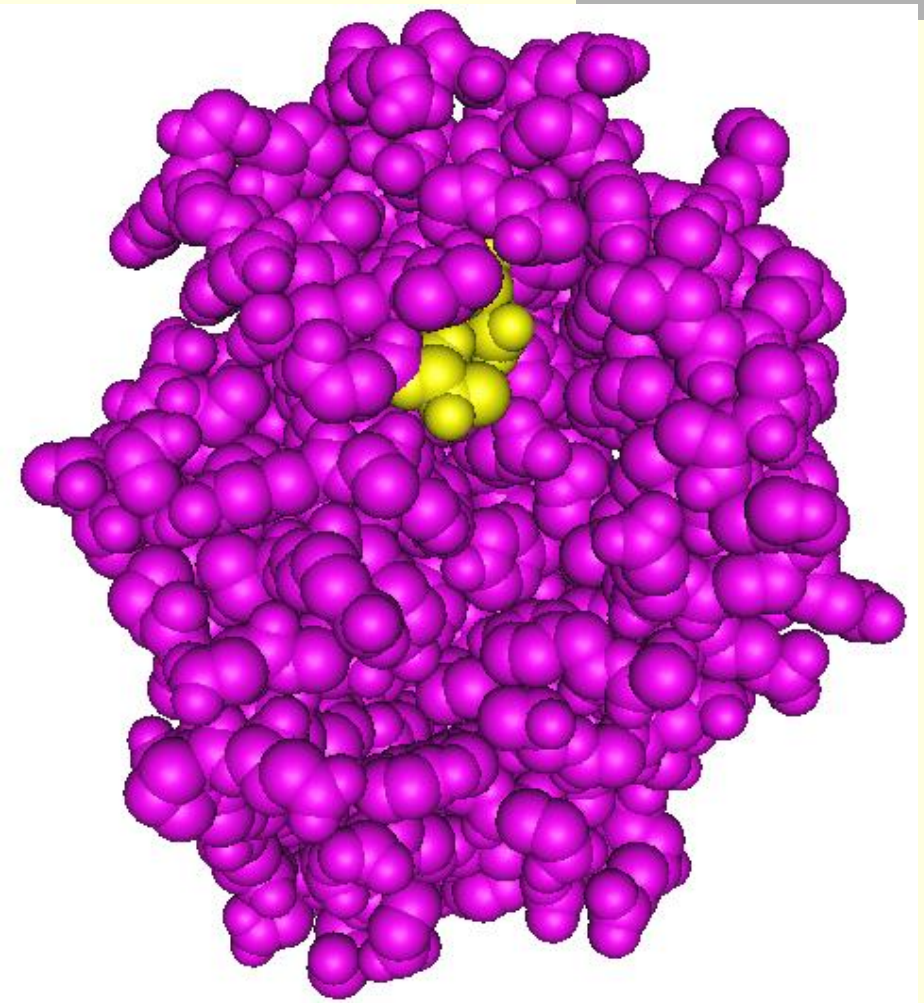
Healthy cell=self peptide
Sick cell=foreign peptide

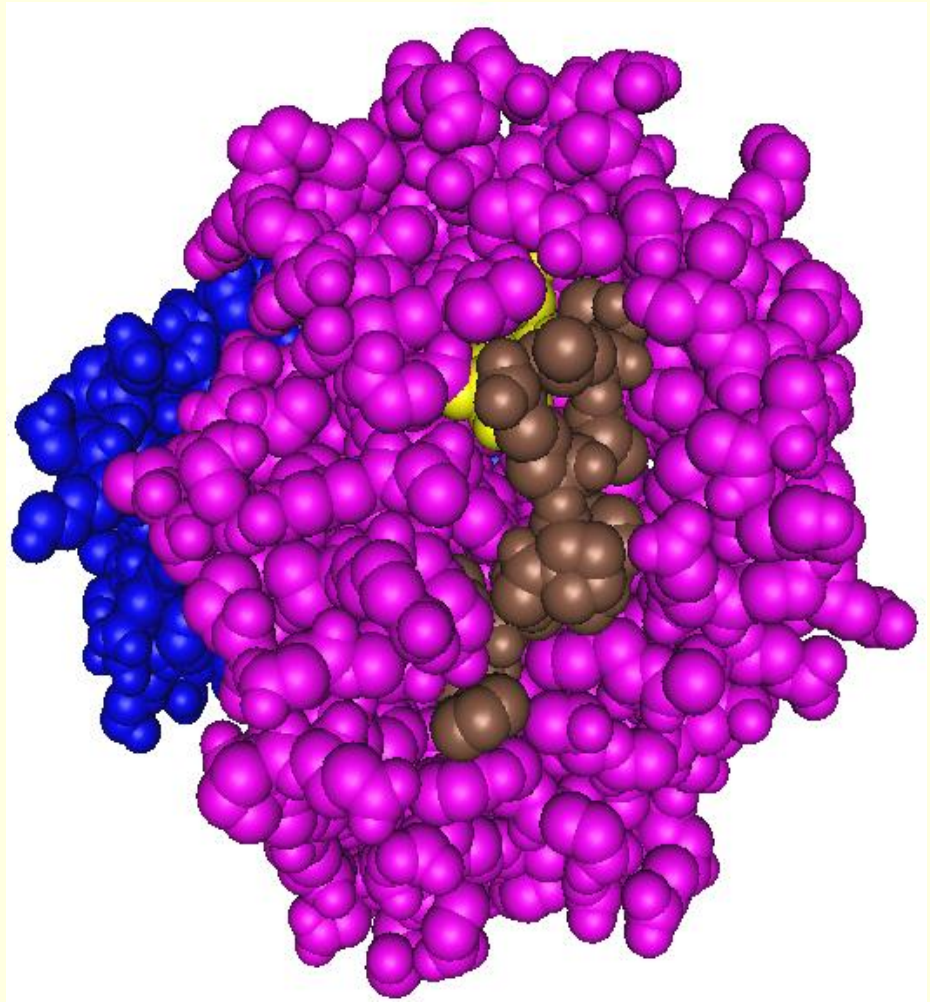

HLA-DQ molecule with bound peptide
α–chain
P1
P6
P9
P4
β–chain
©Peter Hjelmström

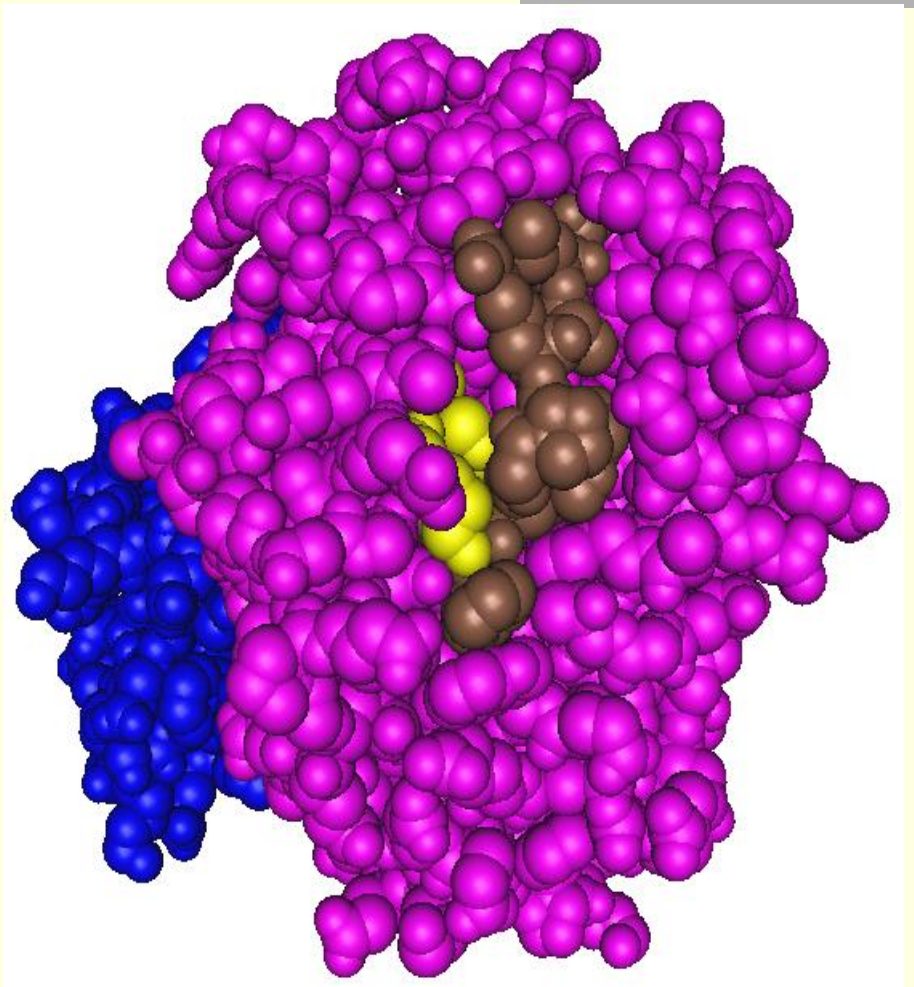
HLA-DQ molecule with bound peptide



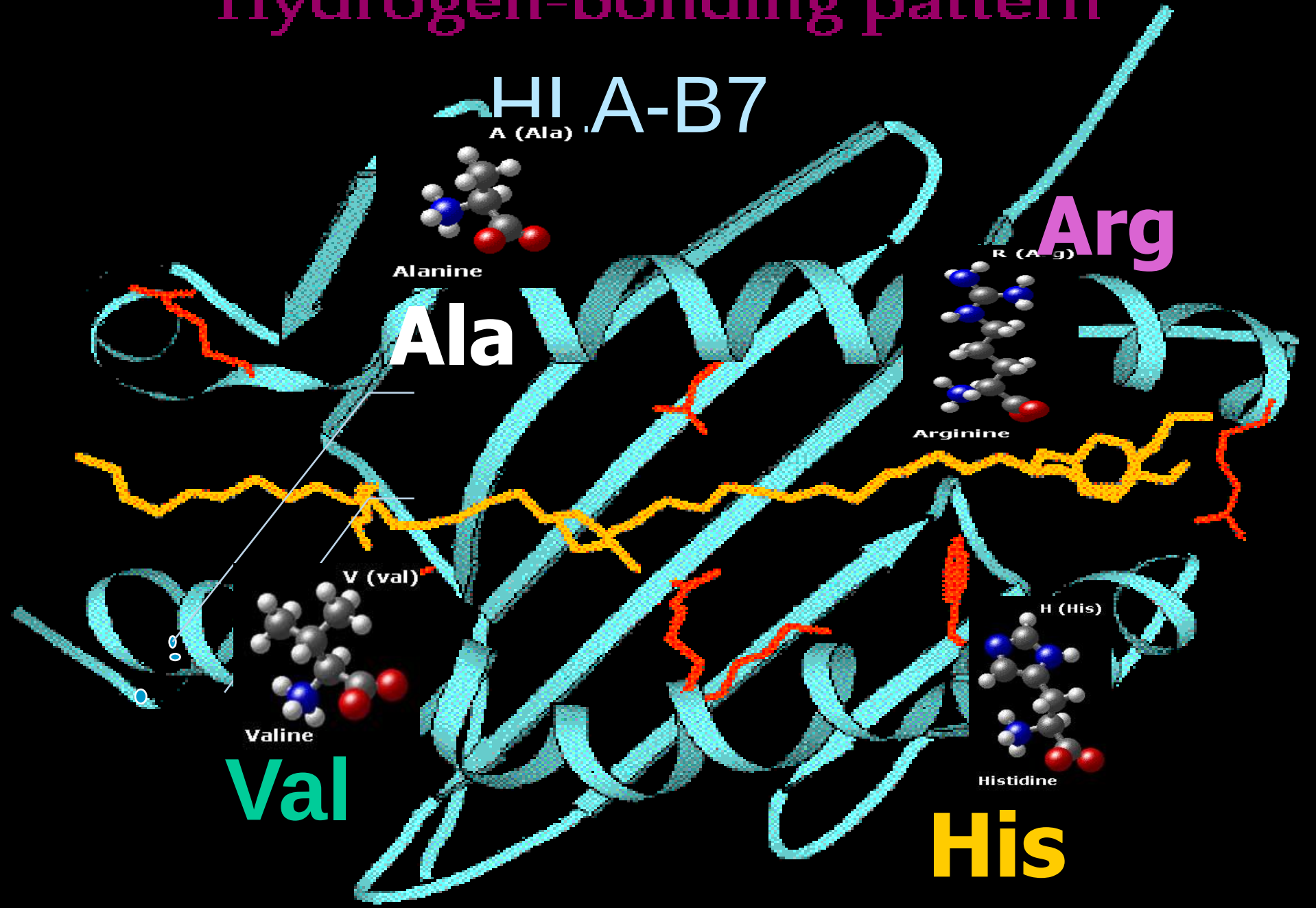


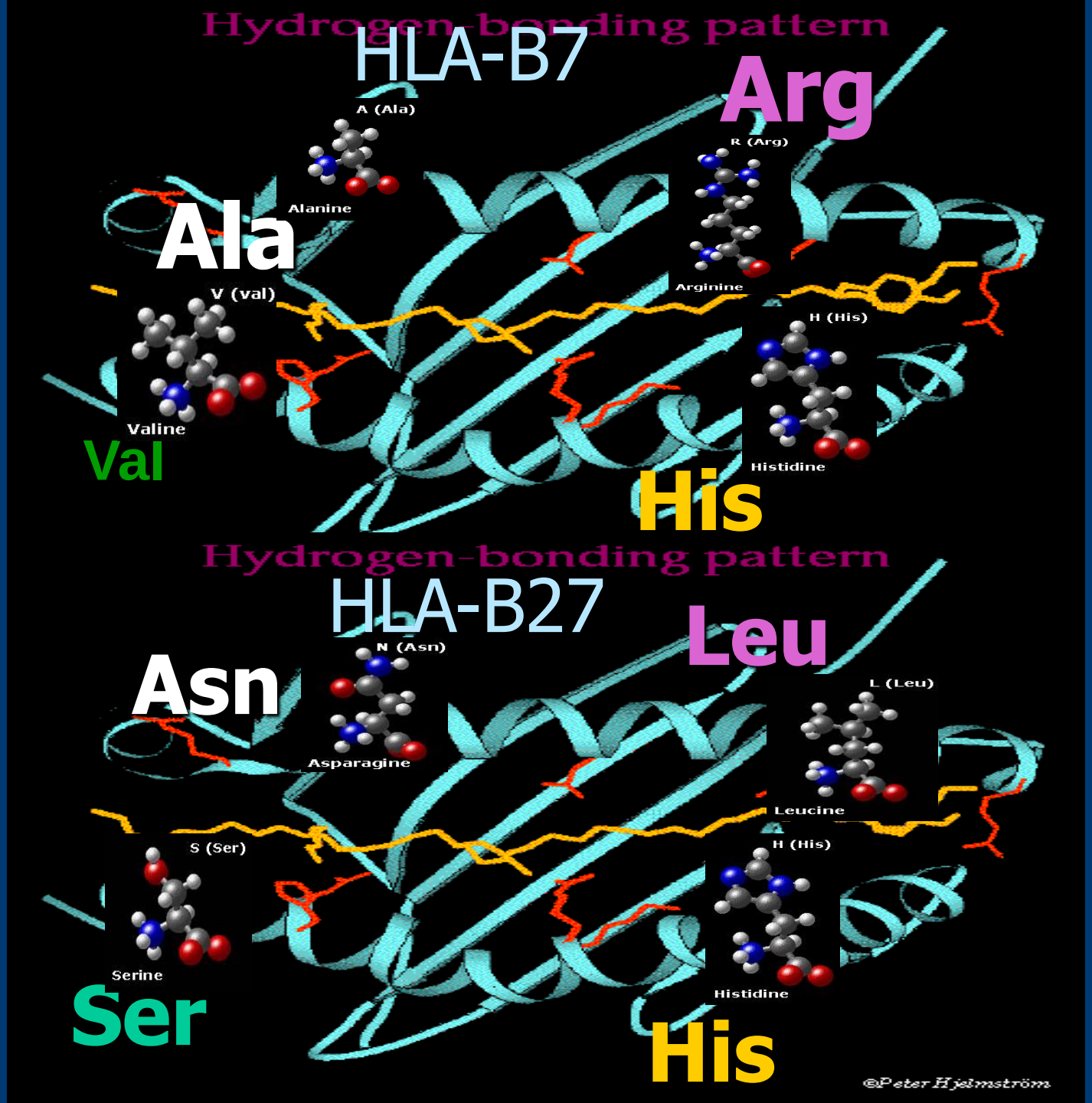


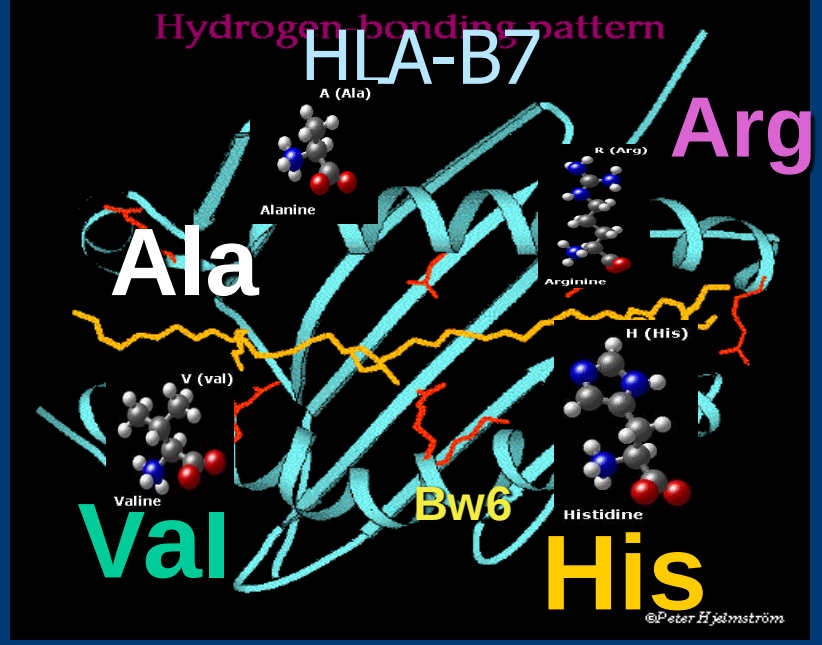


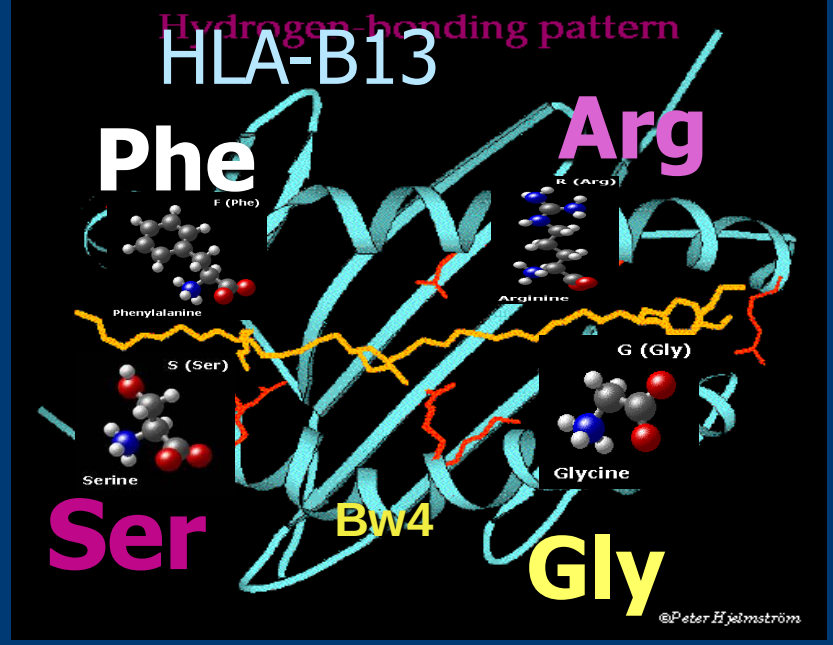


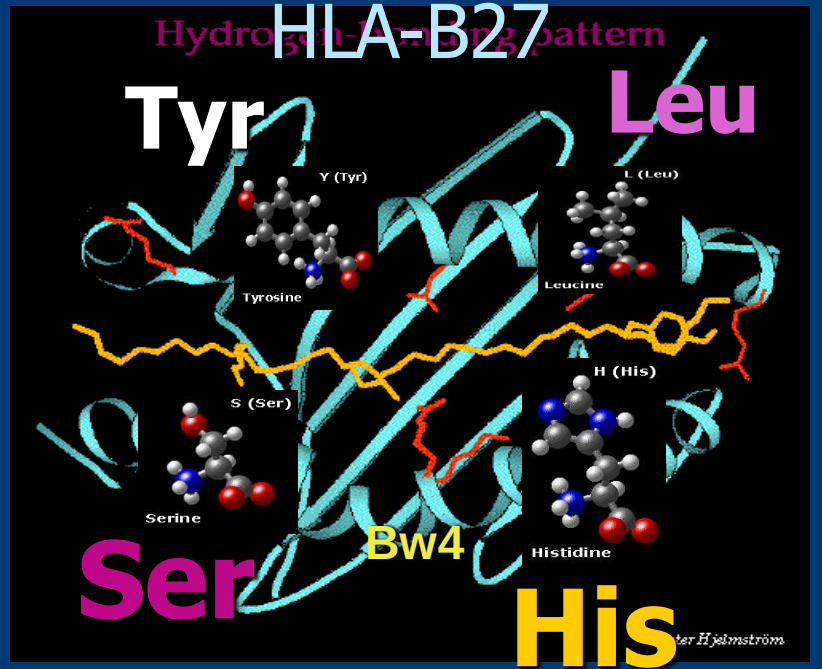
Hydrogen-bonding pattern

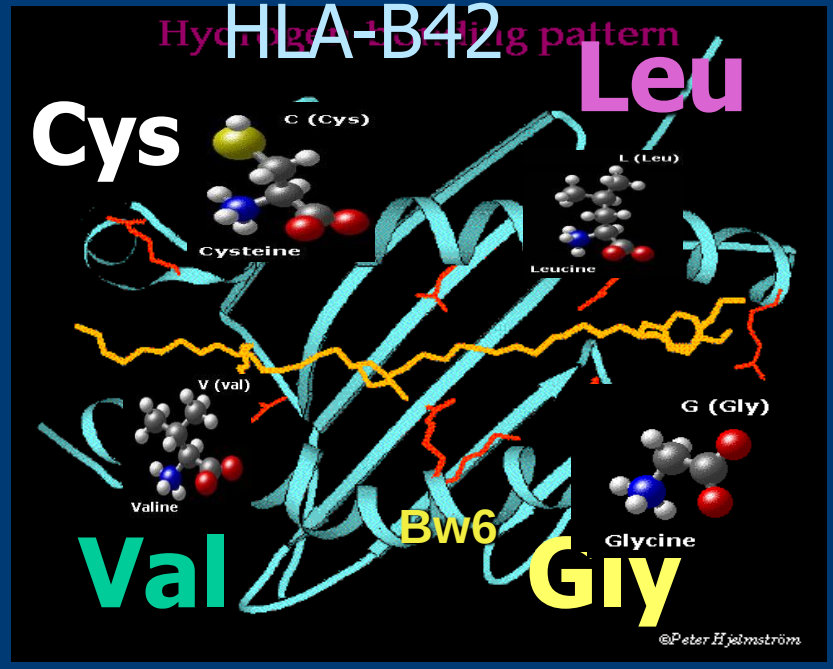



Hydrogen-bonding pattern
HLA-B7
A (Ala)
Alanine
Ala
R (Arg)
Arginine
Arg
V (val)
Valine
Val
H (His)
Histidine
His
Hydrogen-bonding pattern
HLA-B27
N (Asn)
Asparagine
Asn
L (Leu)
Leucine
Leu
S (Ser)
Serine
Ser
H (His)
Histidine
His
©Peter Hjelmström


Hydrogen bonding pattern
HLA-B7
A (Ala)
Alanine
Ala
R (Arg)
Arginine
Arg
H (His)
V (val)
Valine
Val
Bw6
Histidine
His
©Peter Hjelmström


Hydrogen bonding pattern
HLA-B13
Phe
F (Phe)
Phenylalanine
R (Arg)
Arginine
Arg
S (Ser)
Serine
Ser
Bw4
G (Gly)
Glycine
Gly
©Peter Hjelmström


Hydrogen bonding pattern
HLA-B27
Tyr
Y (Tyr)
Tyrosine
Leu
L (Leu)
Leucine
H (His)
S (Ser)
Serine
Ser
Bw4
Histidine
His


Hydrogen bonding pattern
HLA-B42
Cys
C (Cys)
Cysteine
Leu
L (Leu)
Leucine
V (val)
Valine
Val
Bw6
G (Gly)
Glycine
Gly
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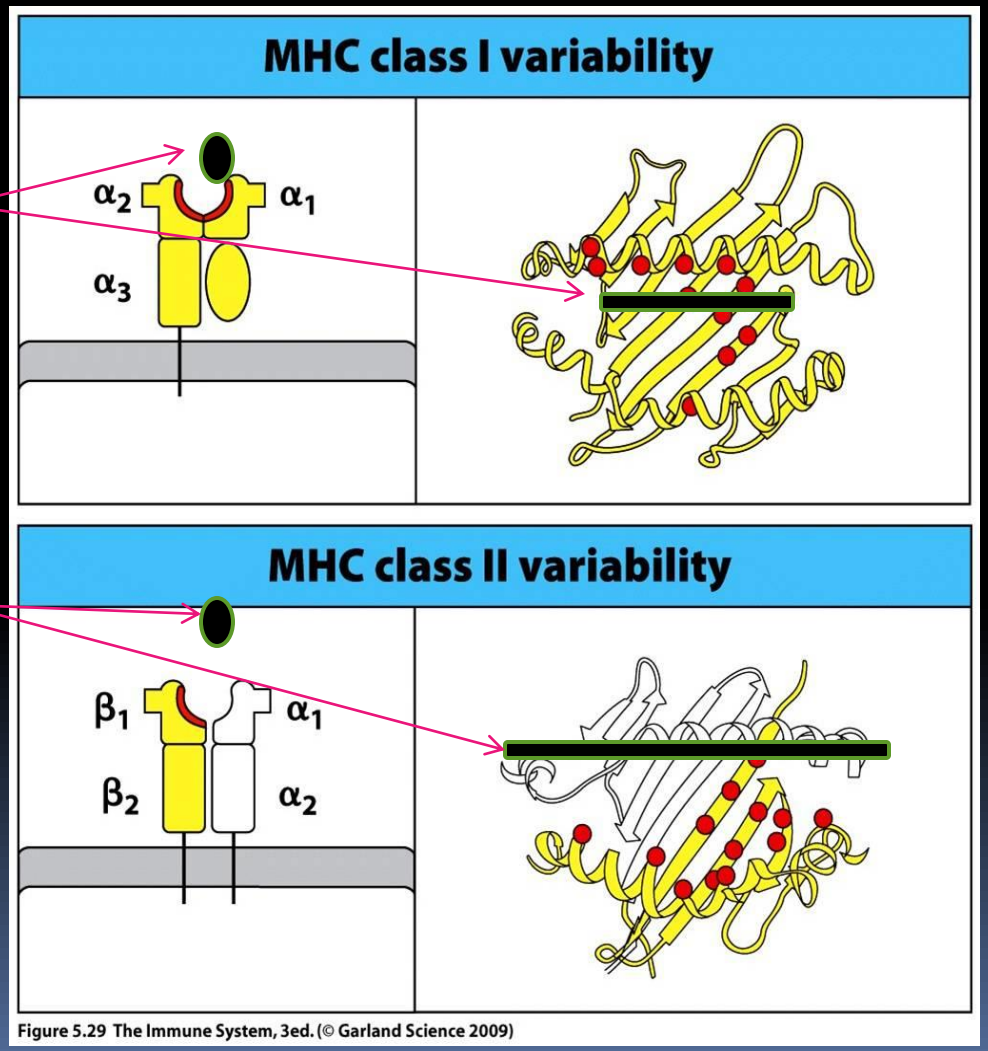
HLA Hypervariable Regions

side view

view from top, looking down

Peptide



Peptide

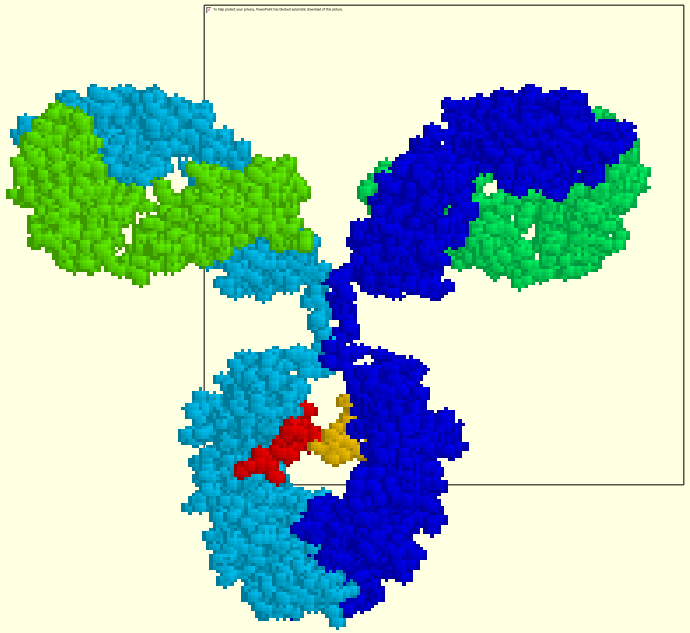
Figure 5.29 The Immune System, 3ed. (© Garland Science 2009)

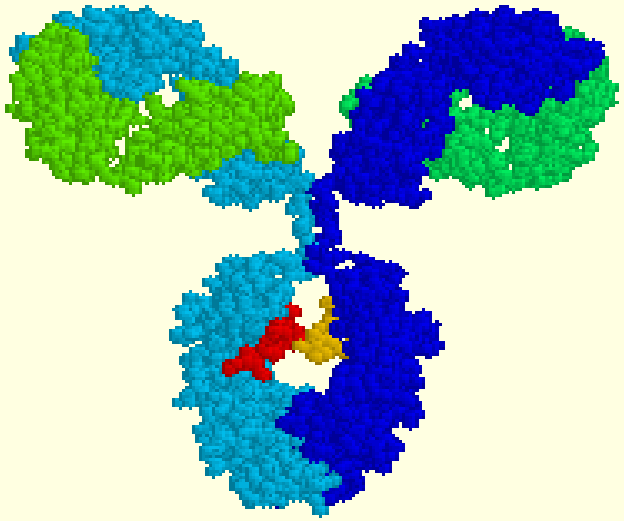
Red indicates hypervariable regions of HLA

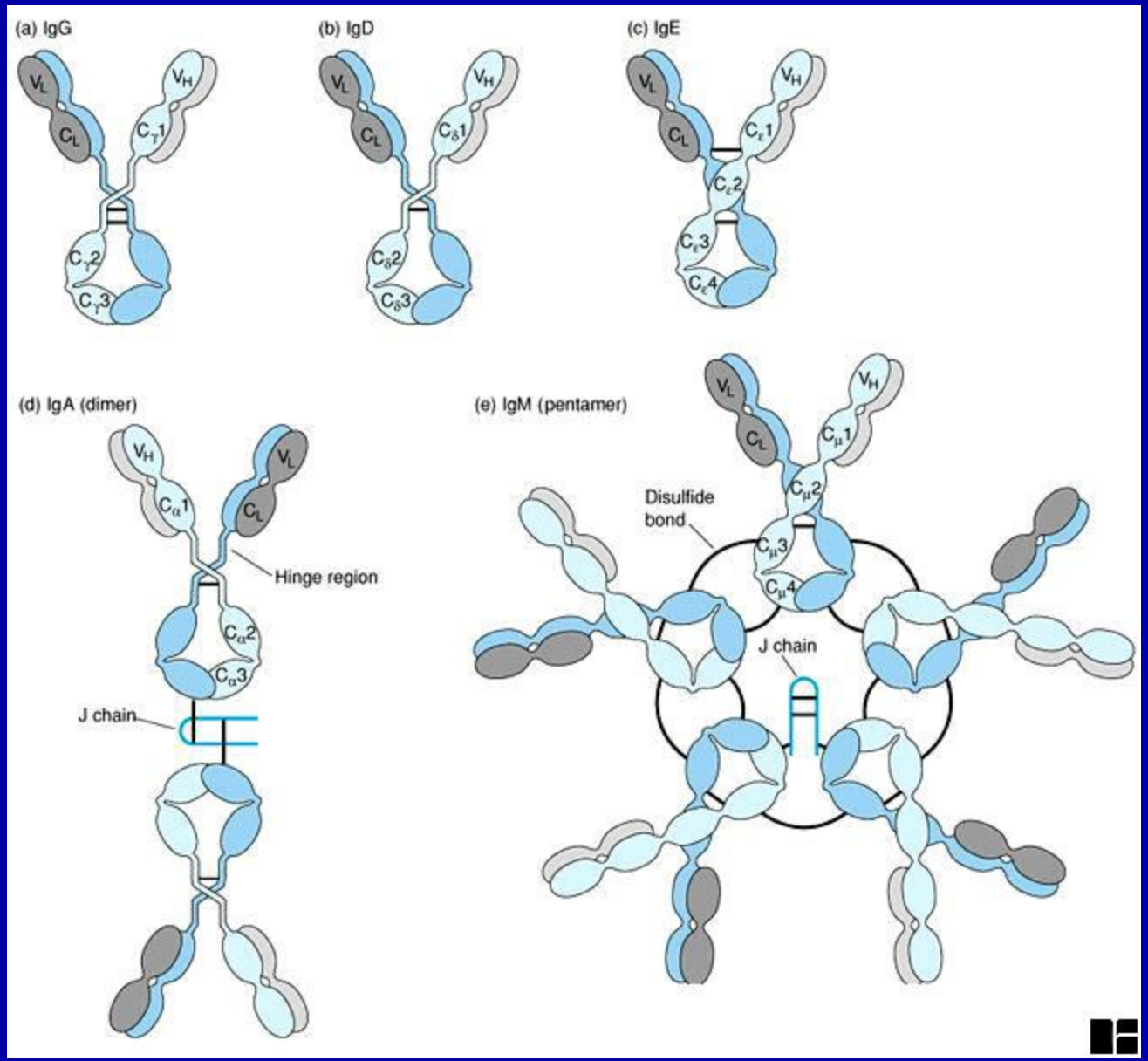
Hypervariable regions are responsible for most of the HLA alleles.

Alleles are variants of an antigen.

Immunoglobulin (IgG)






(a) IgG
(b) IgD
(c) IgE
(d) IgA (dimer)
(e) IgM (pentamer)
Hinge region
J chain
Disulfide bond
J chain

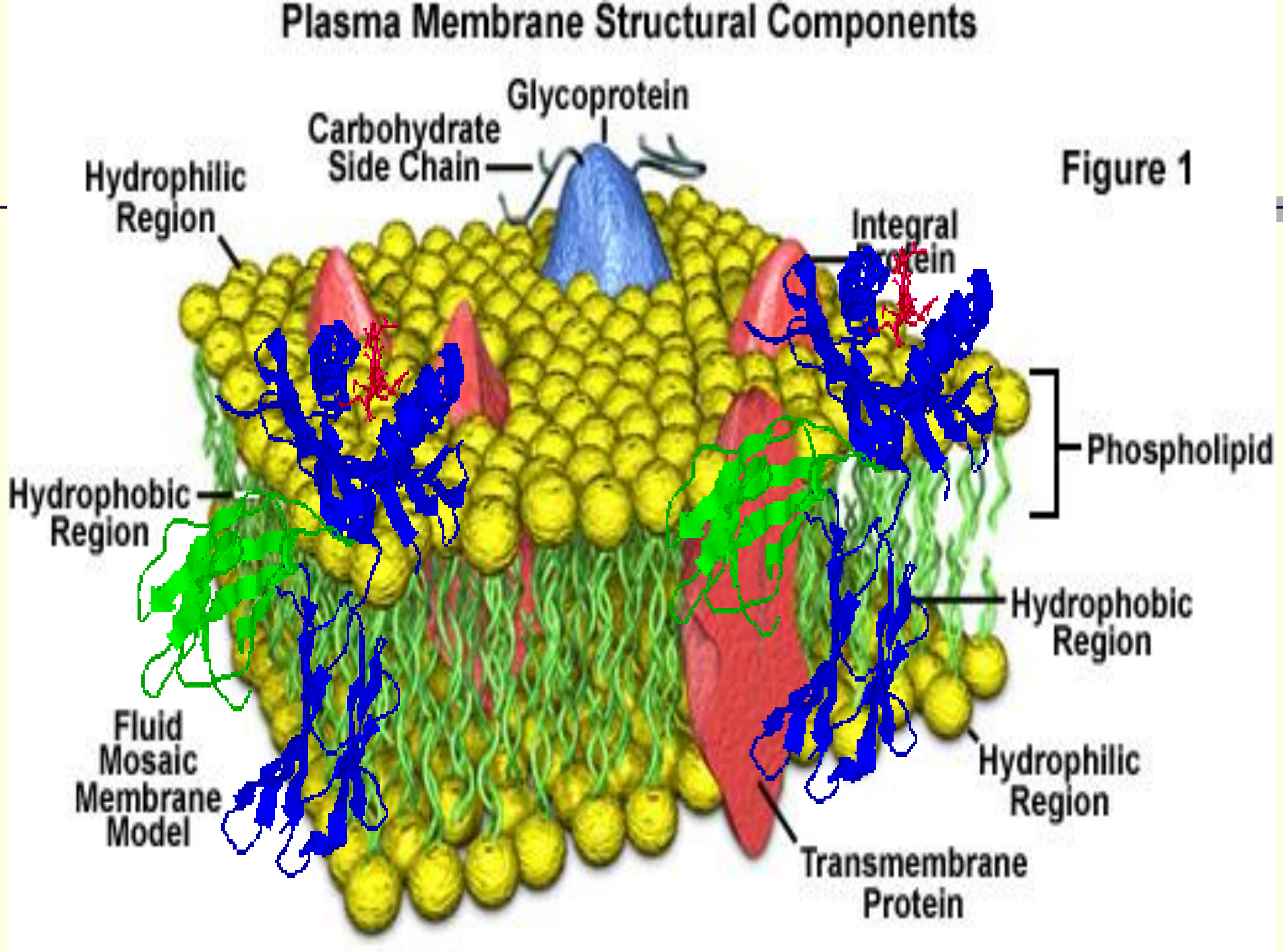
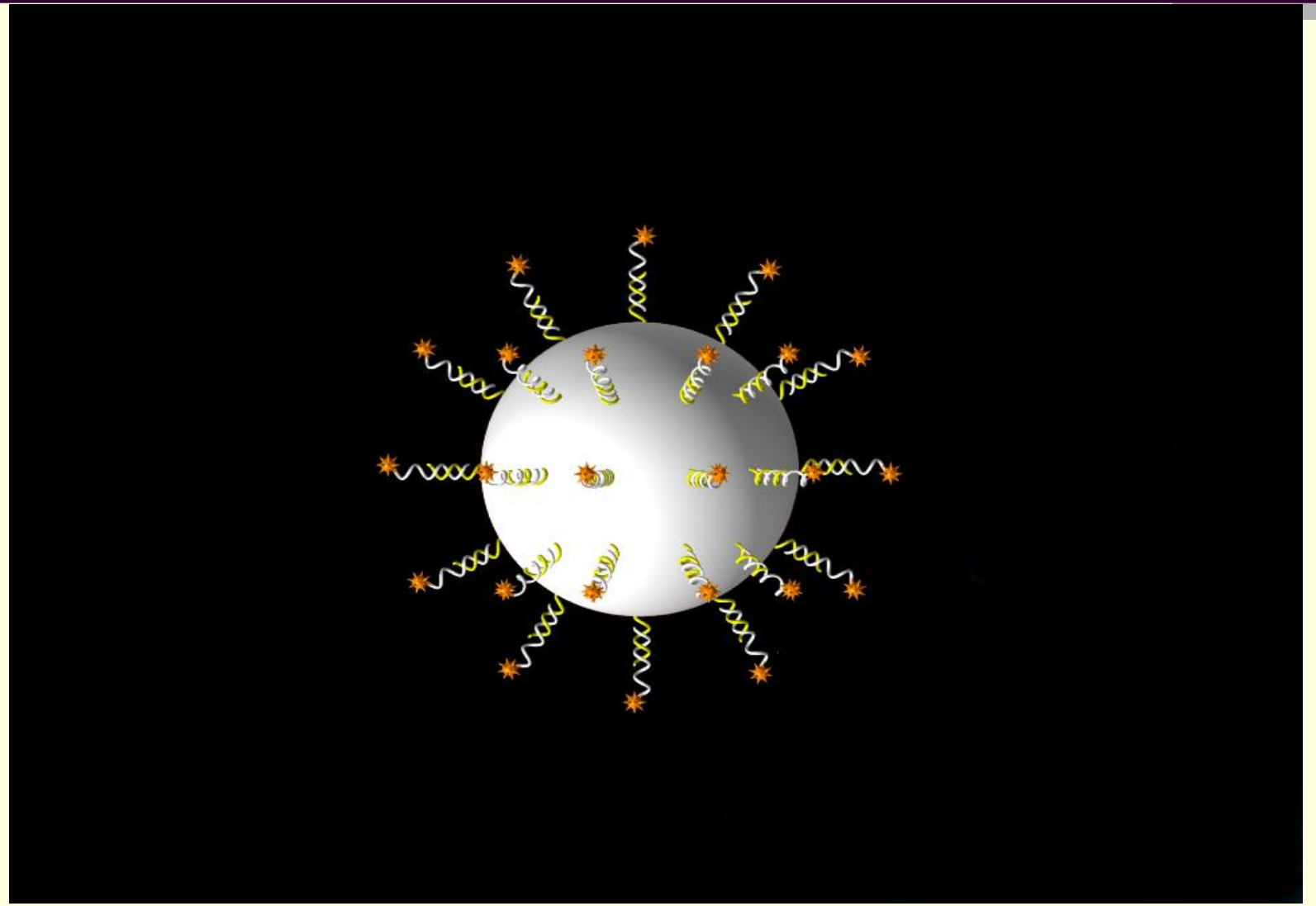

Plasma Membrane Structural Components
Glycoprotein
Carbohydrate Side Chain
Hydrophilic Region
Integral Protein
Phospholipid
Hydrophobic Region
Hydrophobic Region
Fluid Mosaic Membrane Model
Hydrophilic Region
Transmembrane Protein

Figure 1

Structure of Microsphere Beads



Problems

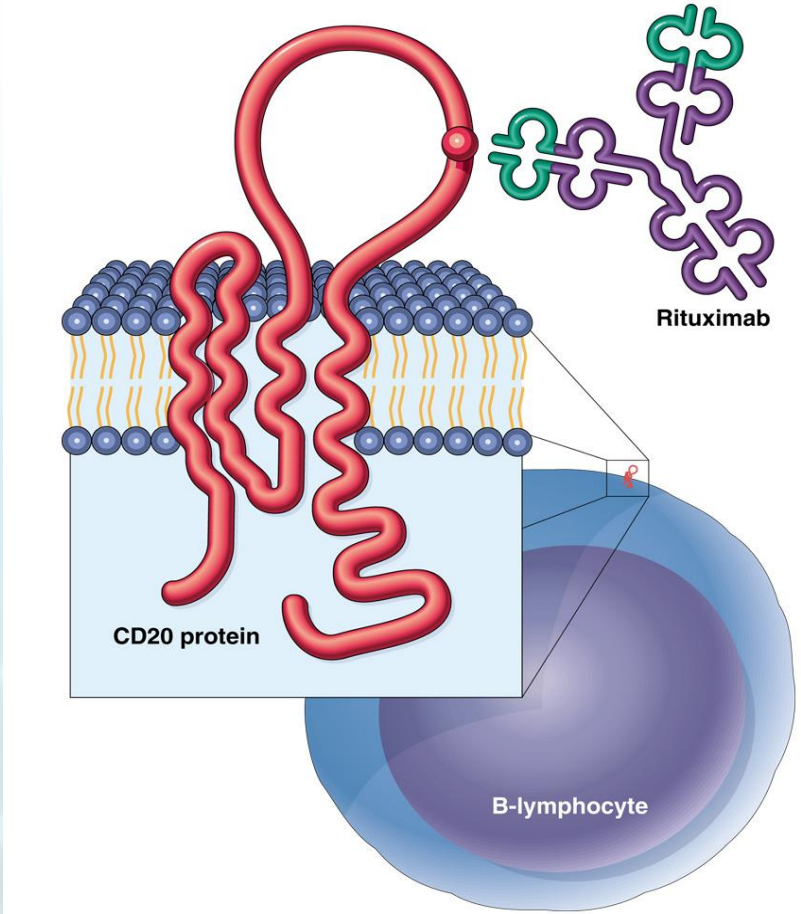
- Prozone (EDTA, Heat inactivation, FBS)
- MFI Values (?Clinically significant)
- Virtual Crossmatch
- Epitope or Eplet Antibodies (Software, Organ and platelets)

Basic Concepts in Desensitization

- Removal of existing antibodies
  - Plasmapheresis
  - Immunoadsorption
- Inhibition of residual antibody and complement cascade
  - Intravenous Immunoglobulin (IVIg)
  - Eculizumab (C5 inhibitor)
- Depletion of antibody producing cells
  - Naïve and memory B cells: rituximab
  - Plasma cells: bortezomib
- Suppression of the T cell response
  - Induction agents
  - Triple immunosuppression with CNI, MMF, steroids

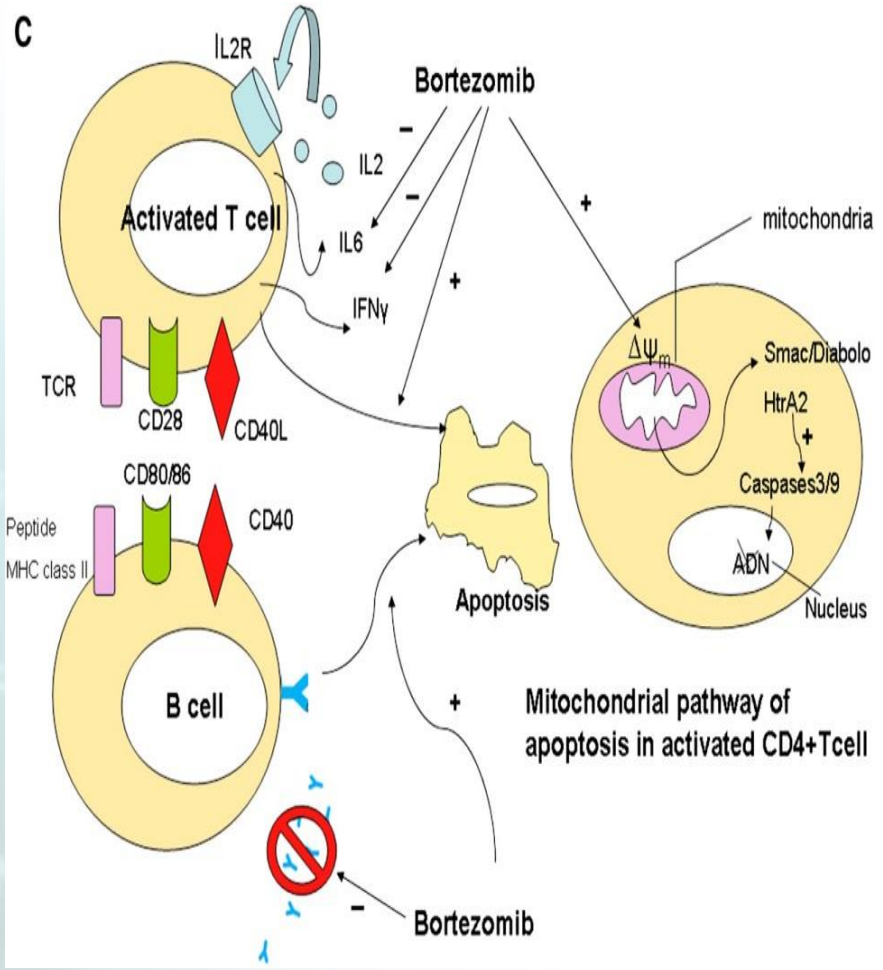
Rituximab

- Genetically engineered monoclonal murine/human antibody
  - Anti-CD20
- FDA approved for treatment of lymphoma
- Used for desensitization and AMR



Bortezomib (Velcade)

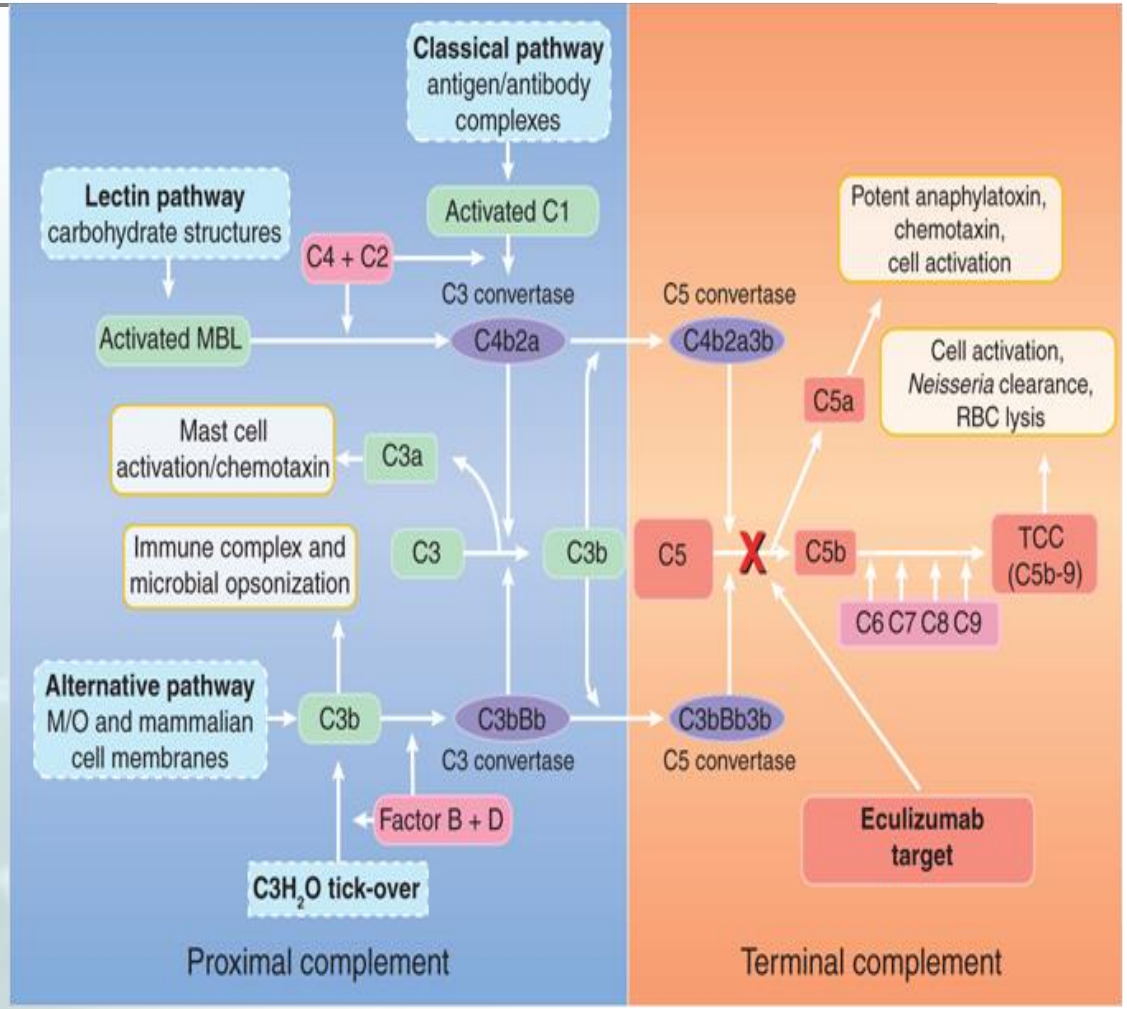
- Proteasome inhibitor
- Directly targets antibody production by plasma cells
- FDA approved for multiple myeloma
- Primarily used for AMR
- Side effects include thrombocytopenia and disabling neuropathy



Richardson et al. NEJM 2003; 348:2609

Eculizumab (Soliris)

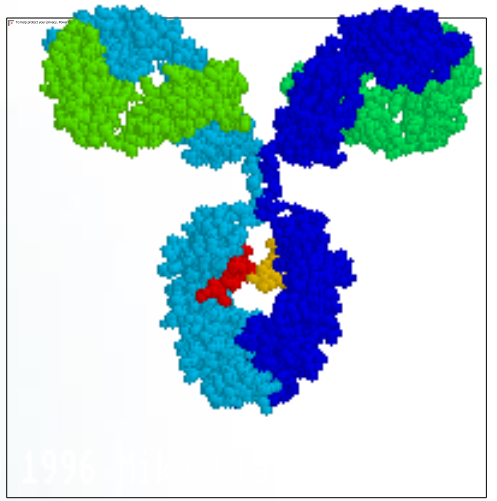
- Genetically humanized monoclonal antibody
  - Anti-C5
- Blocks the activation of terminal complement
- FDA approved for treatment of PNH
  - Primarily used for AMR
- Increased risk of infections with encapsulated bacteria



Hillmen et al. NEJM 2006; 355:1233

IdeS Monoclonal antibodies

- Cleaves where Fab ends
- Removes circulating IgG
- Antibodies rebound within 7-10 days



Summary

Transplantation is State of the Art, because improvement is never ending