Major Histocompatibility Complex (MHC)

Department of Microbiology and Immunological Diagnostics, Pomeranian Medical University
Definitions

**Histocompatibility (transplantation) antigens**
Antigens on tissues and cells that determine graft rejection transplanted between two genetically different individuals.

**Major histocompatibility (MHC) antigens**
Histocompatibility antigens that cause a very strong immune response and are most important in rejection.

**MHC complex**
Group of genes on a single chromosome encoding the MHC antigens.

**HLA (Human Leukocyte Antigens)**
MHC antigens of man (first detected on leukocytes).

MHC=HLA
Genes in the MHC were first identified as important genes in rejection of transplanted tissues.

Genes within the MHC were highly polymorphic.
Three classes of molecules encoded by the MHC genes:
- Class I
- Class II
- Class III

- **Class I MHC molecules** is found on all nucleated cells (not RBCs)

- **Class II MHC molecules** is found on APC
  - Dendritic cells, Macrophages, B cells, other cells
Human Leukocyte Antigens: build and functions
Human Leukocyte Antigens: build and functions

• **Class III MHC molecules**
  – Some complement components (e.g., C4, C2, factor B)
  – Transporter proteins (TAP1, TAP2)
  – TNF

Class III MHC molecules has **no direct role** in immune responses and **no determine** graft survival.
Three-dimensional structures of MHC molecules and the TCR have been determined by X-ray crystallography method.
Structure of Class I MHC

- Two polypeptide chains, a long α chain and a short β (β2 microglobulin)
- Four regions
  - Cytoplasmic region containing sites for phosphorylation and binding to cytoskeletal elements
  - Transmembrane region containing hydrophobic amino acids
Structure of Class I MHC

- Four regions
  - A highly conserved $\alpha_3$ domain to which CD8 binds
  - A highly polymorphic peptide binding region formed from the $\alpha_1$ and $\alpha_2$ domains
- $\beta_2$-microglobulin helps stabilize the conformation
Structure of Class I MHC
Variability map of Class I MHC α Chain
Structure of Class I MHC Ag-Binding Groove

- Groove composed of an α helix on two opposite walls and eight β-pleated sheets forming the floor
- Residues lining the groove are most polymorphic
- Groove accommodates peptides of 8-12 amino acids long

From Janeway et al., Immunobiology 6th Ed.
Structure of Class II MHC

- Two polypeptide chains, \( \alpha \) and \( \beta \), of roughly equal length
- Four regions
  - Cytoplasmic region containing sites for phosphorylation and binding to cytoskeletal elements
Structure of Class II MHC

- Four regions
  - Transmembrane region containing hydrophobic amino acids
  - A highly conserved $\alpha_2$ and a highly conserved $\beta_2$ domains to which CD4 binds
  - A highly polymorphic peptide binding region formed from the $\alpha_1$ and $\beta_1$ domains
Structure of Class II MHC
Variability map of Class II MHC β Chain
Structure of Class II MHC Ag-Binding Groove

- Groove composed of an α helix on two opposite walls and eight β-pleated sheets forming the floor
- Both the α1 and β1 domains make up the groove
- Residues lining the groove are most polymorphic

From Janeway et al., Immunobiology 6th Ed.
Structure of Class II MHC Ag-Binding Groove

- Groove is open and accommodates peptides of 13-25 amino acids long, some of them are outside of the groove

From Janeway et al., Immunobiology 6th Ed.
Important Aspects of MHC

- Although there is a high degree of polymorphism for a species, an individual has maximum of **six different class I MHC products** and only slightly more class II MHC products (taking into account only the major loci).

- Each MHC molecule has only **one** binding site. The different peptides a given MHC molecule can bind, all bind to the same site but only one at a time.
Important Aspects of MHC

• Each MHC molecule can bind many different peptides.

• MHC polymorphism is determined only in the germline. There are no recombination mechanisms for generating diversity.

• MHC molecules are membrane-bound; recognition by T cells requires cell-cell contact.
Important Aspects of MHC

• Alleles for MHC genes are **co-dominant**. Each MHC gene product is expressed on the cell surface of each nucleated cell.

• A peptide must associate with a given MHC of that individual, otherwise no immune response can occur. That is **first level** of control.
Important Aspects of MHC

• Mature T cells must have a T cell receptor that recognizes the peptide associated with MHC. This is the second level of control.

• Cytokines (especially interferon-γ) increase level of expression of MHC.
Important Aspects of MHC

• Endogenous peptides associate with class MHC I and are recognized by Tc cells
• Exogenous peptides associate with class MHC II and are recognized by Th cells.
**Presentation of antigens by HLA class I to cytotoxic lymphocytes T CD8+**

1. Endogenous antigen is fragmented in proteosome to the short peptide fragments
2. The TAP proteins transport the short peptides to endoplasmic reticulum
3. The peptide binds MHC class I molecules
4. Golgi apparatus transports the complex to the surface of nucleated cells and presents the complex to Tc lymphocytes
5. Tc lymphocytes recognize the MHC I-peptide by TCR and CD8 receptors
6. Tc lymphocytes product the granzymes and perforins which damage infected cells.
Presentation of antigens by HLA class II to helper lymphocytes T CD4+

1. Exogenous antigen after the endocytosis process is fragmented in endosome to the short peptide fragments
2. In the endosome the short peptides bind MHC class II molecules
3. Golgi apparatus transports the complex to the surface of APC and present the complex to lymphocyte Th
4. Th lymphocytes recognize the MHC II-peptide by TCR and CD4 receptors
5. Th lymphocytes product the cytokines which activate other cells such as Tc lymphocytes, phagocytic cells, B lymphocytes to damage the antigens
<table>
<thead>
<tr>
<th>Presented proteins</th>
<th>HLA class I</th>
<th>HLA class II</th>
</tr>
</thead>
<tbody>
<tr>
<td>Endogenous peptides</td>
<td>8-10 aminopeptides (viral proteins, proteins of allograft)</td>
<td>Exogenous peptides</td>
</tr>
<tr>
<td>Lenght of chain</td>
<td>30 minutes</td>
<td>&gt;12 aminopeptides</td>
</tr>
<tr>
<td>Time of reach to the surface of cellular membrane</td>
<td>2-4 minutes</td>
<td></td>
</tr>
</tbody>
</table>
Activity of lymphocytes Tc CD8+

Activity of lymphocytes Th1 CD4+

Activity of lymphocytes Th2 CD4+
HLA (human leukocyte antigens)

Chromosome 6

It is seventh chromosome which was fully described after 20, 21, 22, 7, 14, and chromosome Y. Chromosome 6 contains genetic material equal to 6% of all people's genome. It consists of 166 mln (166,880,988) nucleotide pairs, giving 1557 active genes and 633 pseudogenes. Among these genes there are 130 genes which may cause increased susceptibility to disease.

The most polymorphic genes in these chromosomes is locus HLA-B and also in the whole genome (559 alleles according to Anthony Nolan Research Institute Database; 2004; 748 alleles – 2006; 3051 alleles – 2015)
MHC – Major Histocompatibility Complex in people

β2-microglobulin is encoded in 15 chromosome

HLA class II

HLA class I

HLA – human leukocyte antigen

Numbers of specificities in individual locus (wg Anthony Nolan Research Institute Database, 2008)
**HLA class I classical molecules (HLA – class Ia)**
- HLA - A
- HLA - B
- HLA - C

**HLA class I unclassical molecules (HLA – class Ib)**
- HLA – E (trophoblast)
- HLA – F (limph.B, skin)
- HLA – G (trophoblast)
- MICA, MICB (endothelial cells)
- CD1 (encoded outside the MHC)

---

**HLA class II classical molecules**
- HLA - DR
- HLA - DQ
- HLA - DP

**HLA class II unclassical molecules**
- HLA - DM
- HLA - DO
**Numbers of described antigens and alleles**  
(according to Anthony Nolan Research Institute Database, **2004**)

<table>
<thead>
<tr>
<th>Locus HLA-</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of alleles (n=1012)</td>
<td>303</td>
<td>559</td>
<td>150</td>
</tr>
<tr>
<td>Number of antigens (n=97)</td>
<td>28</td>
<td>59</td>
<td>10</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Gen HLA-</th>
<th>DRA</th>
<th>DRB1</th>
<th>DRB3</th>
<th>DRB4</th>
<th>DRB5</th>
<th>DQA1</th>
<th>DQB1</th>
<th>DPA1</th>
<th>DPB1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of alleles (n=644)</td>
<td>3</td>
<td>363</td>
<td>40</td>
<td>12</td>
<td>17</td>
<td>25</td>
<td>56</td>
<td>20</td>
<td>108</td>
</tr>
<tr>
<td>Nuber of Antigens (n=39)</td>
<td>-</td>
<td>21</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>-</td>
<td>9</td>
<td>-</td>
<td>6</td>
</tr>
</tbody>
</table>
Numbers of described antigens and alleles
(according to Anthony Nolan Research Institute Database, 2006)

<table>
<thead>
<tr>
<th>Gen HLA-</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of alleles (n=1394)</td>
<td>429</td>
<td>748</td>
<td>217</td>
</tr>
<tr>
<td>Number of antigens (n=97)</td>
<td>28</td>
<td>60</td>
<td>10</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Gen HLA-</th>
<th>DRA</th>
<th>DRB1</th>
<th>DRB3</th>
<th>DRB4</th>
<th>DRB5</th>
<th>DQA1</th>
<th>DQB1</th>
<th>DPA1</th>
<th>DPB1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of alleles (n=759)</td>
<td>3</td>
<td>511</td>
<td></td>
<td></td>
<td></td>
<td>32</td>
<td>69</td>
<td>23</td>
<td>121</td>
</tr>
<tr>
<td>Number of antigens (n=39)</td>
<td>21</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>-</td>
<td>9</td>
<td>-</td>
<td>6</td>
<td></td>
</tr>
</tbody>
</table>
Numbers of described antigens and alleles
(according to Anthony Nolan Research Institute Database, 2015)

<table>
<thead>
<tr>
<th>Gen HLA-</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of alleles (n=2028)</td>
<td>852</td>
<td>3051</td>
<td>460</td>
</tr>
<tr>
<td>Number of Antigens (n=98)</td>
<td>28</td>
<td>60</td>
<td>10</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Gen HLA-</th>
<th>DRA</th>
<th>DRB1</th>
<th>DRB3</th>
<th>DRB4</th>
<th>DRB5</th>
<th>DQA1</th>
<th>DQB1</th>
<th>DPA1</th>
<th>DPB1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of Alleles (n=914)</td>
<td>3</td>
<td>745</td>
<td>52</td>
<td>14</td>
<td>19</td>
<td>34</td>
<td>91</td>
<td>26</td>
<td>128</td>
</tr>
<tr>
<td>Number of Antigens (n=39)</td>
<td>-</td>
<td>21</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>-</td>
<td>9</td>
<td>-</td>
<td>6</td>
</tr>
</tbody>
</table>
HLA TYPING
TYPING METHODS

• **SEROLOGICAL** used to be the ‘gold’ standard. Suppressed by molecular techniques as they become more robust and time efficient. This method is mainly used for class I typing.

• **MOLECULAR** fast becoming the method of choice. On these days used in the most laboratories – for typing class I and II.
Serological method are based on LYMPHOCYTOTOXIC TEST (CDC - test)

Reaction between antigen and antibody
Antigen (blood cells), antibody (serum)

In the past serum came from:
• multigravidas
• persons who had multiple blood transfusions
• recipients who had allograft

At the presence - monoclonal antibodies
**Lymphocytotoxic test (Complement Dependent Cytotoxicity test)**

- Viable lymphocytes are incubated with HLA specific antibodies. If the specific antigen is present on the cell the antibody is bound.

- Rabbit serum as a source of complement is added, incubate. If antibody is bound to the HLA antigen on the cell surface it activates the complement which damages the cell membrane making it permeable to vital stains.

- Results are visualised by adding stain, usually a fluorochrome eg ethidium bromide although trypan blue and more often eosin also are used.

- Dead lymphocytes – with dye inside (darker), lived limphocyte – without dye (light, shining)
Lymphocytotoxic test

- Test is read by using an inverted fluorescent or light microscope.

- A mixture of T and B lymphocytes can be used for HLA class I typing. The HLA class I is mainly estimated by using this method.

- B lymphocytes are used for HLA Class II typing. In these method we must separated limphocytes B by using immunomagnetic bead separation or rosette test.
## Lymphocytotoxic test

**1. Isolation of lymphocytes** and preparing suspension containing about 2000-3000 cell/μl which vitality is more than > 80%

**2. Add** the 1 ul suspension of lymphocytes to each well on plate with monoclonal antibodies against HLA

**3. Add** the rabbit **complement**

**4. Dye** by eosine or tryptan blue

**5. Read** the test by using inverted light microscope

Antibodies bound to specific antigens HLA on the surfaces of lymphocytes and damage them by activate classical complement pathway

### Terasake plate
Lymphocytotoxic test

Advantages of this method

1. In this test we used monoclonal antibodies.
2. Easily performed, does not require expensive equipment.
3. Takes around four hours to perform
4. Doesn’t require sterile environment

Disadvantages of this method

1. We must isolate and use the living limphocytes
2. Problem with the typing HLA of cancer patients
3. False positive results as a cross reaction between HLA which have similar structure
Lymphocytotoxic test

Disadvantages

4. False negative reaction caused by:
   - contamination by erythrocytes or platelets,
   - low numbers of lymphocytes
   - change of pH in serum

5. This method is limited to the detection of specific antigens which is associated with low numbers of different monoclonal antibodies
MOLECULAR METHODS

- PCR-SSP (with Specific Sequences of Primers)
- PCR-SSO (with Specific Sequences of Oligonucleotides)
- SBT (Sequence Based Typing) and NGS (Next Generation Sequencing)
- Fluorocytometry
Nomenclature of HLA

• At the presence time we have double nomenclature: serological and molecular (confirmed and actualised by Comitett of HLA Nomenclature at WHO)
Serological typing

Genetical typing

Nomenclature of HLA

DR

DRB1*

Number of antigen

Number of allel

Serological specificity
HLA typing is used in:

1. The transplantology to the optimalisation of graft survival – typing of HLA for donor and recipient (kidney and bone marrow OBLIGATORY other organ FACULTATIVE)

2. Excluding paternity

3. Association between HLA and different autoimmune diseases
## Associate of HLA antigens with autoimmunity diseases

<table>
<thead>
<tr>
<th>Disease</th>
<th>Antigen of HLA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sclerosis multiple</td>
<td>DR2</td>
</tr>
<tr>
<td>Systemic lupus erythematosus (SLE)</td>
<td>DR2 or DR3</td>
</tr>
<tr>
<td>Myasthenia gravis</td>
<td>DR3 or B8</td>
</tr>
<tr>
<td>Rheumatoid arthritis</td>
<td>DR4</td>
</tr>
<tr>
<td>Celiac disease</td>
<td>DQ2 or DQ8</td>
</tr>
<tr>
<td>Hashimoto's thyroiditis</td>
<td>DR5</td>
</tr>
<tr>
<td>Insulin dependent diabetes mellitus.</td>
<td>Heterozygote DR3/DR4</td>
</tr>
<tr>
<td>Ankylosing spondylitis</td>
<td>B27</td>
</tr>
</tbody>
</table>
Segregation of haplotypes among family members

HAPLOTYPET
The panel of genetic determinants occurs on one chromosome; the panel of genes occurs on one chromosom and is inherited together. The child always inherits one haplotye from the mother and one haplotype from the father, so the child is 50% compatible with the parents.
Segregation of haplotypes among family members

If the parents are heterozygotic the probability of compatible between siblings is 25%

MOTHER - HETEROZYGOSITY

- A1 B8 DRB1*0301 (a)
- A2 B7 DRB1*0401 (b)

FATHER - HETEROZYGOSITY

- A30 B13 DRB1*1201 (c)
- A23 B5 DRB1*0402 (d)

SISTER

- (a) / (c)

BROTHER

- (a) / (d)

BROTHER

- (b) / (c)

SISTER

- (b) / (d)
Segregation of haplotypes among family members

If the one of parents is homozygotic and the second is heterozygotic the siblings are 50% compatible

<table>
<thead>
<tr>
<th>MOTHER - HETEROZYGOSITY</th>
<th>FATHER - HOMOZYGOSITY</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 B8 DRB1*0301</td>
<td>A30 B13 DRB1*1201</td>
</tr>
<tr>
<td>A2 B7 DRB1*1301</td>
<td>A30 B13 DRB1*1201</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>SISTER</th>
</tr>
</thead>
<tbody>
<tr>
<td>(a) / (c)</td>
</tr>
<tr>
<td>(a) / (c)</td>
</tr>
<tr>
<td>(b) / (d)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>BROTHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>(a) / (d)</td>
</tr>
<tr>
<td>(b) / (c)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>BROTHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>(b) / (d)</td>
</tr>
</tbody>
</table>
Crossing over

Usually we inherit the genes of HLA in chromosome of parents together but sometimes we can observe the crossing over process in the making gametes. During the process chromosomes of mother or father exchange the fragments of arms. In such cases we inherit some genes of one chromosome and genes of the other chromosome of mother or father.

<table>
<thead>
<tr>
<th>MOTHER</th>
<th>FATHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 B8 DRB1*0301</td>
<td>A30 B13 DRB1*1201</td>
</tr>
<tr>
<td>A2 B7 DRB1*0401</td>
<td>A23 B5 DRB1*0402</td>
</tr>
<tr>
<td>A30 B13 DRB1*1201</td>
<td>A30 B13 DRB1*1201</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>SISTER</th>
<th>BROTHER</th>
<th>BROTHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 B8 DRB1*0301</td>
<td>A1 B8 DRB1*0301</td>
<td>A2 B7 DRB1*0401</td>
</tr>
<tr>
<td>A30 B13 DRB1*1201</td>
<td>A23 B5 DRB1*0402</td>
<td>A30 B13 DRB1*1201</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>SISTER</th>
<th>BROTHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 B8 DRB1*0401</td>
<td>A1 B8 DRB1*0401</td>
</tr>
<tr>
<td>A30 B13 DRB1*1201</td>
<td>A30 B13 DRB1*1201</td>
</tr>
</tbody>
</table>
Types of graft (dependent on person/species)

- **Xenograft**
  Grafts between members of different species (also known as heterologous, xenogenic or heterografts)

- **Allograft**
  Grafts between **two genetically different** members of the same species (also known as allogenic or homograft)

- **Autograft**
  Graft within same individuals

- **Isograft/Syngraft**
  Grafts between **two genetically identical** members of the same species (identical twins or inbred animals)
Types of graft (dependent on location)

• **Ortothropic** – the same anatomical place – heart, lung, liver, cornea

• **Heterothropic** – different anatomical place – kidney, skin, ovary tissue
Which organs can be transplanted:
- kidneys
- bone marrow or HSC (hematopoietic stem cells)
- liver
- pancreas
- lungs
- heart
- skin
- cornea and other

At present, we can make more allografts between not related people (cadaveric organs) than family grafts.

The most frequently transplanted organs are kidneys and bone marrow.
When we want to transplant kidneys we should make the following tests in donor and recipient:

- typing the HLA
- virtual cross-match
- cross match test CXM (between donor and recipient)
- viral tests against HIV, CMV, HBV, HCV and facultatry EBV
- blood group test (compatible blood group A-A, B-B etc)
- PRA test (only for recipient)
- specificity of anti-HLA antibodies (solid phase tests) – only for recipient

The cross match (CXM) test based on lymphocytotoxic test. We incubate the isolated lymphocytes from blood of donor with the serum of recipient. We check if serum of recipient contains antibodies against the HLA of donor.

When the result of the test is positive (we can observe killed lymphocytes) - recipient is disqualified (hyperacute rejection)
PRA test – Panel Reactive Antibodies

The PRA based on lymphocytotoxic test.

Aim - estimation level of recipient alloimunization (%). We can’t estimate specificity of antibodies (anti-HLA = before transplantation, DSA = after transplantation)

Serum of recipient + panel of 30-50 different solutions of isolated lymphocytes from random chosen non related people (high polymorphism of HLA - amount > 30 different unrelated persons provide occurrence the most of HLA from our population)

30 different reaction (15 positive + 15 negative = 50%PRA)

>80% PRA – high immunized recipient
PRA test – Panel Reactive Antibodies

- test must be repeated every **3 months during all period** waiting for transplantation

recipients can have episodes of immunization:
- blood transfusion
- pregnancy
- transplantation
- viral infection (*theory of heterologous immunity* – antibodies against HLA can occur after viral infection due to cross reaction between similar epitopes on viruses and MHC of recipient)
A-HLA and DSA

**Before transplantation**
- Antibodies present in the recipient **against all** HLA molecules
- PRA - % of a-HLA without specificity
- e.g. 50% but anti-HLA-A2, anti-HLA-B8 ????
- Solid Phase Tests allow for determining of specificity

**After transplantation**
- **Donor Specific Antibodies**
- Responsible for organ rejection
- PRA reaction useless
- specificity determining of DSA - very important for next transplantation
FLUOROCYTOMETRY as the solid phase method for determining of anti-HLA antibodies

- Polystyrene microbeads coated by single HLA antigens
- Serum of recipient
- Secondary antibodies labeled with fluorochrome
- Special instrument (Luminex – fluorocytometr) divides microbeads and measures MFI (medium fluorescence intensity) on each of them
- ↑ MFI = ↑ level of antibodies
Virtual cross-match (vCXM)

- Recipient with **DSA > 5.000 MFI** (medium fluorescence intensity) – vCXM positive
- Recipient with **DSA < 5.000 MFI** (medium fluorescence intensity) – vCXM negative (in next stage biological cross-match must be performed)
- Recipient **without DSA** – vCXM negative (in next stage biological cross-match must be performed)
DONORs HLA: A1, 2  B7, 18  DR 4,11

Recipient 1: anti-HLA antibodies against A3 and B8  
vCXM negative - passes to biological CXM

Recipient 2: DSA antibodies against A2 (1.580 MFI)  
vCXM negative - passes to biological CXM

Recipient 3: DSA antibodies against A2 (7.200 MFI) and DR4 (13.257 MFI)  
vCXM positive - doesn’t pass to biological CXM
Selection HLA antigens between donor and recipient in kidney allograft

The recipient must have minimum 50% compatibility of HLA with the donor

The identical antigens in locus A, B, DR in recipient and donor are: A1, B8, DRB1*0407
The different antigens which are absent in recipient: A23, B13, DRB1*0413
Selection HLA antigens between donor and recipient in bone marrow allograft

The recipient must have 100% compatibility of HLA with the donor

HLA antigens in locus A, B, C, DR and DQ in recipient and donor are identical
Principles of qualification of recipients for kidney and bone marrow transplants

1. **Compatibility** in blood groups between the donor and recipient

2. The result of *virtual cross-match* must be *negative*

3. The result of *cross match* test (biological) must be *negative*

4. The result of viral test of donor must be negative unless the recipient has positive result also

5. Compatibility in HLA antigens between the donor and recipient
   - in bone marrow compatibility must be 100% - all antigens of HLA in locus **A, B, C, DR and DQ** must be identical
   - in kidneys compatibility must be 50% - part of antigens of HLA in locus **A, B, DR** must be identical
Allocation points (SELECTING THE RECIPIENT)

- For HLA compatibility
- HLA –A - 2 points for each absent incompatibility
- HLA –B - 5 points for each absent incompatibility
- HLA –DR - 10 points for each absent incompatibility
- Regional Centre of Qualifications – 4 points
- Dialysis time – 1 point for each year of dialysis
- PRA – 50-79% - 7 points

- Obligatory transplantation (without counting allocation points, only vCXM and biological CXM negative)
- High immunized patients PRA >80%
- Urgent patients – without vascular access
Differences between old and new one

ALOCATION POINTS

Recipient heterozygosity HLA – A*01, *02; B*07, *08 DRB1*01, *11
Donor heterozygosity HLA – A*01, *03; B*07, *44 DRB1*01, *04
NO DIFFERENCE

Recipient heterozygosity HLA – A*01, *02; B*07, *08 DRB1*01, *11
Donor homozygosity HLA – A*01, *01; B*07, *07 DRB1*01, *01

old scoring – one compatible antigen in each locus (2+5+10 =17pt)

new scoring for each absent incompatibility – donor delivers no incompatible antigens
(2+2+ 5+5+ 10+10 = 34pt)
**GvHD disease** is caused by donor’s lymphocytes transplanted with bone marrow or HSC. These lymphocytes attack the tissues of recipient. Most common cause of death after bone marrow transplantation.

**Acute GvH** – reaction occur before 100 day after HSCT. Donor’s lymphocytes recognize foreign antigens presented by APC of recipient. The activity helper lymphocytes produce cytokines: Th1 cytokines, which stimulate Tc lymphocytes and NK cells, while Th2 cytokines stimulate B lymphocytes.

**Chronic GvH** – after 100 day after transplantation. Donor’s lymphocytes which matured from transplanted HSC recognize antigens of recipient as foreign antigens and induce response against them.
Early acute GvH reaction with widespread, almost confluent hyperpigmented lichenoid papules and toxic epidermal necrosis-like appearance on knee.

Disease process are also covered: liver (hiperbilirubinemia) and intestines (diarrhea)

Late chronic GvH reaction with hyperpigmented sclerotic plaques on the back

Acute GvH reaction with vivid palmar erythema
## Different patterns of graft rejection

<table>
<thead>
<tr>
<th>Type of rejection</th>
<th>Time taken</th>
<th>Cause</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperacute</td>
<td>Minutes-hours</td>
<td>Preformed anti-donor antibodies or incompatibility in blood groups</td>
</tr>
<tr>
<td>Acute accelerated</td>
<td>24h</td>
<td>Reactivation of sensitized T cells or very low concentration of anti-donor antibodies not detected in CXM</td>
</tr>
<tr>
<td>Acute</td>
<td>Days – weeks</td>
<td>Primary activation of T cells</td>
</tr>
<tr>
<td></td>
<td>&lt;1 year</td>
<td>Causes unclear: antibodies, immune complexes, slow cellular reactions, recurrence of disease or non immunological causes (immunosupresive drugs, cold ischemia time, age of donor/recipient).</td>
</tr>
<tr>
<td>Chronic</td>
<td>&gt;1 year</td>
<td></td>
</tr>
</tbody>
</table>
- **Hyperacute rejection**
  This occurs in instances when the recipient has preformed high titer antibodies. A graft may show signs of rejection within minutes to hours due to immediate reaction of antibodies and complement.

- **Acute accelerated rejection**
  Transplantation of a second graft with the same incompatible antigens as in the first one, results in a rapid (24h) rejection. It is due to presence of T-lymphocytes sensitized during the first graft rejection. Accelerated rejection is mediated by immediate production of lymphokines, activation of monocytes and macrophages, and induction of Tc.

- **Acute rejection**
  The normal reaction that follows the first grafting of a foreign transplant takes 1 - 3 weeks. This is known as acute rejection and is mediated by T lymphocytes sensitized to class I and class II antigens of the allograft, elicitation of lymphokines and activation of monocytes and macrophages.

- **Chronic rejection – most common cause of graft rejection**
  Some grafts may survive for months or even years, but suddenly exhibit symptoms of rejection. This is referred to as chronic rejection, the mechanism of which is not entirely clear. The hypotheses are that this may be due infection, causes which led to failure of the first organ, loss of tolerance induced by the graft, etc.