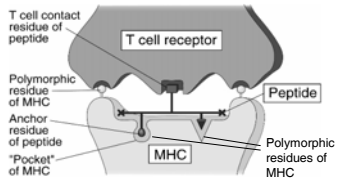
HLA Genetics

Different MHC alleles confer different functional properties on the adaptive immune system

Each different allelic MHC molecule (allotype) confers the ability to bind different peptides

Consequences for regulation of adaptive immunity:



• The large number of MHC alleles means each individual has a nearly unique set of peptide-presenting allotypic MHC molecules

• These molecules present self-peptides during thymic development of the T cell repertoire and select a nearly unique repertoire of T cell clones that differs among individuals

Consequences for Transplantation

The differences MHC between individuals means that the cells of a donor who differs from the recipient by any of the MHC alleles are recognized as non-self by the T cells of the recipient and are attacked as if they were a foreign substance

This difference is the origin of the name "major histocompatibility complex" that reflects the role for these molecules as the primary genetically determined targets for graft rejection or compatibility

Nomenclature

Genotype: the collection of genes in an individual, usually referring to a small segment of a chromosome

Alleles: the alternative forms of a gene found at the same locus in different individuals

Allotypes or allomorphs: different protein forms encoded by alleles

Haplotype: the genes (alleles) contributed by one parent, usually referring to alleles of both class I and class II loci

Gene loci exhibit linkage, a measure of their genetic distance

Linkage disequilibrium: certain alleles in a haplotype are found together significantly more (or less) frequently than expected by chance

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

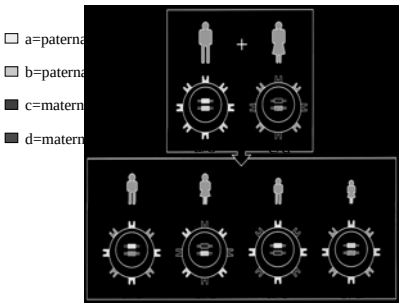
Allele	E.g. HLA-B*0801	"Specificity"	
	*0802	HLA-B8	
	…		
	*0821		
	*2701		
	*2702		
	*2703	HLA-B27	
	…		
	*2725		

But to make things "simpler", alleles are grouped in families as "specificities", e.g. HLA-B8 or HLA-B27, reflecting an old nomenclature used when human alloantibodies were used to first detect HLA "specificities"

Methods used to detect MHC alleles "HLA typing" vary in their resolution to detect specific alleles or only groups of similar alleles

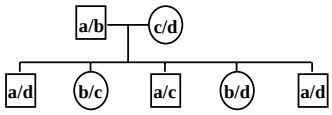
Pregnancy or transplant sera	DNA probes or primers
Complement mediated cytotoxic or immunofluorescence reactions on living lymphocytes	DNA hybridization or sequencing
Recognize broad "serologic specificities", e.g. HLA-B27	Specific alleles (or defined families of related alleles), e.g. HLA-B*2705

Codominant expression of MHC alleles



HLA genetics in transplantation: HLA haplotypes

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

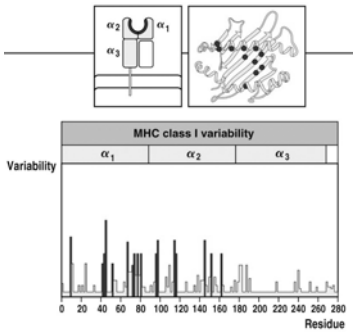


Note that in a family the parents always differ by one haplotype from the children, while children may share 0, 1 or 2 haplotypes

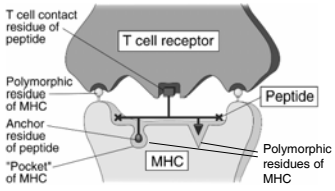
If an immune response to a peptide is determined by a particular haplotype, e.g. "a" it exhibits codominant inheritence

How does polymorphism influence binding of different peptides and what is its immunologic significance?

HLA alleles act as Immune Response genes by determining which peptides are presented to a T cell

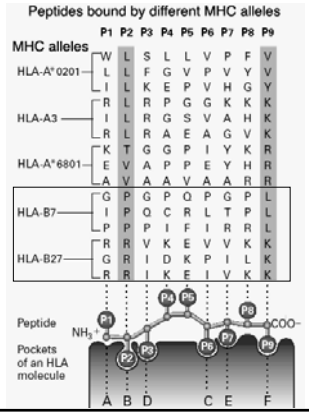


Polymorphic Amino acids that distinguish alleles of MHC class I molecules are found primarily in pockets that determine peptide binding or on the surface that interacts with the TCR



The precise size, shape and charge of each peptide binding pocket in an MHC allotype are determined by particular amino acids coded for by polymorphisms that distinguish each of the MHC class I alleles

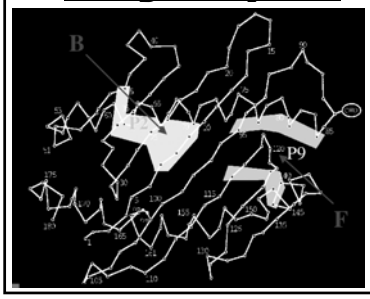
These change the type of peptide that is bound and the interaction of the MHC with the TCR



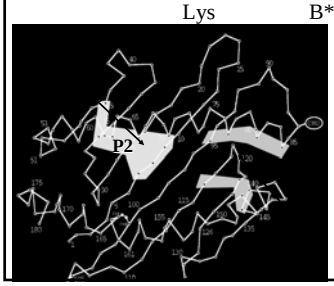
Primary Anchors P2, P9

Secondary anchors P3, P6, P7

Location of MHC class I pockets termed “B” and “F” that bind P2 and P9 amino acid side chains of the peptide



Example	B pocket position 45	Allele	Amino acid bound at P2
	Glu	B*2705	Lys/Arg
	Lys	B*4001	Asp/Glu



Amino acids of the MHC molecule that form the B and F pockets confer the main specificity for binding peptides through complementary interactions with amino acids at position P2 and P9 in the peptide

What peptides are found in MHC molecules?

•Elution of peptides from MHC molecules reveals that class I molecules typically bind 2000-10,000 different peptides per cell

•Each of these peptides has the dominant motif reflecting the relatively conserved anchor residues, e.g. for HLA-B27

Motif XRXXXXXX[KRYL]

•Most peptides are fragments of conventional cell proteins, e.g.

HRAQVIYTR 40S ribosomal protein
RRIKEIVKK Heat shock protein 89
ARLFGIRAK Breast basic conserved protein
RRFFPYYVY Proteasome subunit C5
GRWPGSSLY Lamin B receptor

•Even the most abundant peptide species accounts for only 1% of the total peptides bound, so the T cell has its work cut out

HLA alleles influence the number of peptides in a protein that can be recognized (Example HIV envelope protein), and thus the number of different T cell clones responding

Allele:	**HLA-B*27052**	**HLA-B*3501**	**HLA-B*0702**
Motif	XRXXXXXX[KRYL]	XPXXXXXXY	XPXXXXXXL

Peptides in HIV env able to bind each HLA allotype

IRGKVQKEY	KRRVVQREK	DPNPQEVVL
IRPVVSTQL	ARILAVERY	KPCVKLTPL
TRPNNNTRK	ERDRDRSIR	RPVVSTQLL
IRIQRGPGR	LRSLCLFSY	SPLSFQTHL
SRAKWNNTL	TRIVELLGR	IPRRIRQGL
LREQFGNNK	CRAIRHIPR	
FRPGGGDMR	IRQGLERIL	
WRSELYKYK		
# of peptides	**15**	**0**	**6**

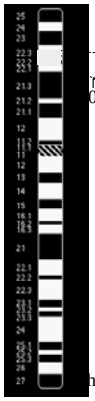
What accounts for the large number of HLA alleles?

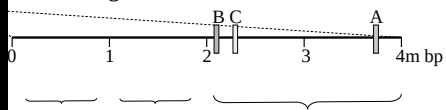
- Homogeneity in a population allows a pathogen to adapt to a molecularly stereotyped adaptive immune response=disadvantage
- Heterozygosity results in a more vigorous T cell response with more clones recognizing more peptides
- Epidemics favor individuals with rare allotypes= *frequency dependent selection*
- Selection for alleles depends on particular pathogen environment
- Small populations loose alleles by chance=*drift*
- Tribal amalgamation results in a large population with many alleles

MHC molecule expression

Assembly of class I MHC molecules

Organization of the MHC





Class II: HLA-DR, HLA-DQ, HLA-DP

Class III: C4A, C4B, C2, Bf, TNF-α

Class I: HLA-A, HLA-B, HLA-C

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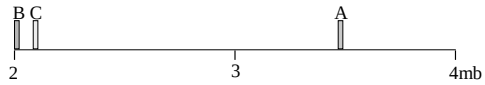
Polygenic human MHC class I (HLA-A,B,C) genes

Nomenclature and number of alleles

MHC class I loci	Specificity (Antigen)	Allele designation	# of alleles
HLA-**A** α-chain	A1, A2,…	A*0101,…	303
HLA-**B** α-chain	B7, B8,…	B*0702,…	559
HLA-**C** α-chain	Cw1, Cw2...	C*0101,...	150



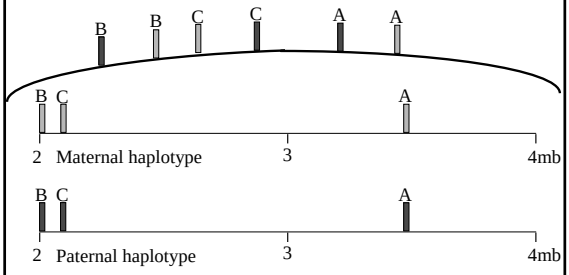
(β_2 microglobulin encoded on chromosome 15)

Maximum number of different types of MHC class I molecules (allotypes) expressed on the cell surface

	Nucleated cells	Antigen presenting cells
Class I (HLA-A)	2	2
Class I (HLA-B)	2	2
Class I (HLA-C)	2	2
Total	6	6

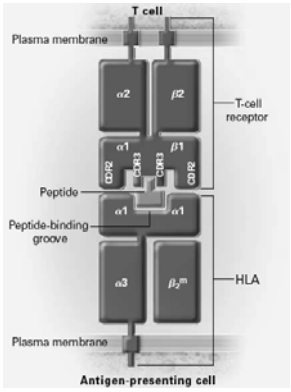
Each of these 6 MHC molecules selects its *own T cell repertoire* that only recognizes peptides presented by that particular type of MHC molecule - *MHC restriction*

Codominant expression of MHC genes yields 6 different class I molecules (allomorphs) on the cell



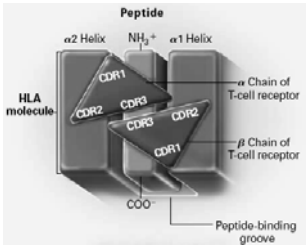
10-100,000 molecules of each type are present on most cells

What does the T cell see?



T cell receptors interact both with the MHC molecule and with the bound peptide

CDR3 regions of TCR α-chain and β-chain primarily, but variably, interact with the antigenic peptide

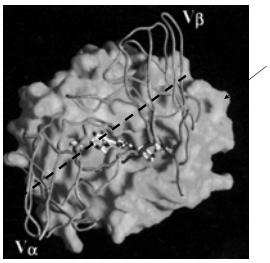


TCR-Peptide-MHC interaction

Critical for understanding

- T cell recognition of peptide
- Selection of T cell repertoire
- Regulation of adaptive immune response

T cell receptors interact both with the MHC molecule and with the bound peptide



MHC

However each TCR contacts peptide and MHC slightly differently

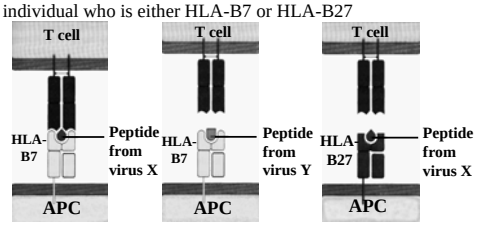
MHC I-CD8 TCR

What are the immunologic consequences of the dual specificity of TCR for peptide and MHC?

MHC restriction of T cell recognition

Because the TCR recognizes both peptide and MHC molecule, T cell recognition of MHC-peptide is both MHC restricted and specific for the immunizing peptide

In each of the 3 experiments the T cell is from a HLA-B7 person who recovered from infection by virus "X".
The APC target cell is either infected with virus X or Y and is from an individual who is either HLA-B7 or HLA-B27

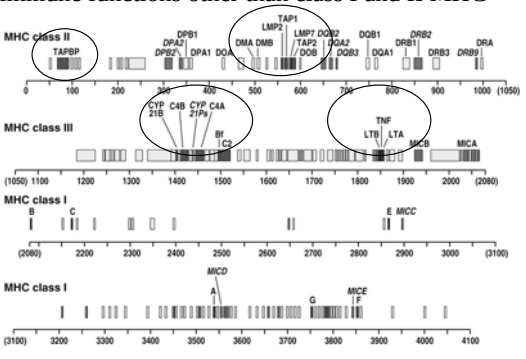


Target killed: ***Yes*** **No** **No**

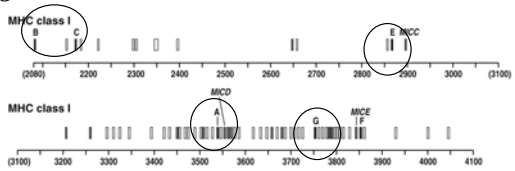
The recent sequencing of the MHC as part of the genome project has revealed a wealth of knowledge about this gene complex

Apart from the role of class I and class II MHC genes in antigen presentation, many of the other genes comprising the entire MHC appear to have been selected for antigen processing and other roles in the immune response

The MHC also contains numerous genes with immune functions other than class I and II MHC

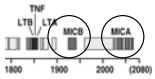


The MHC class I region contains additional class I genes other than HLA-A, B and C



MHC class IB genes

- **HLA-G** the only MHC class I expressed on trophoblast and inhibits NK cell killing of fetus
- **HLA-E** binds leader peptide of classic class I molecules, HLA-A, B and C and inhibits NK cell killing of cell by engagement of CD94 / NKG2A receptor



MHC class IB genes
MICA, MICB

- Expressed in fibroblasts and intestinal epithelium
- Upregulated by cellular injury and stress, not γ-IFN
- Engage stimulatory NK receptor NKG2D
- Physiologic means of removing damaged cells

To escape recognition, an obvious strategy for a pathogen to take is to have a gene or genes that interferes with peptide processing or presentation on MHC class I (or II) molecules

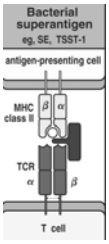
To counter this, the NK cell lineage has evolved a set of receptors that detect **"missing self"**, a decrease in the expression of class I MHC molecules

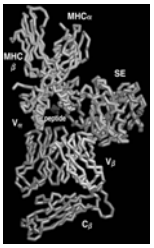
Problem: how is the absence of a molecule recognized?

- The NK cell is capable of "spontaneous" killing
- But it expresses an inhibitory receptor that binds an MHC ligand
- Normal cells are protected from spontaneous killing when they appropriately express this MHC ligand
- Loss of MHC ligand releases the NK cell from its tonic inhibition via engagement of the inhibitory receptor, resulting in its activation

Mechanisms pathogens use to subvert adaptive immune recognition

Promiscuous triggering of TCR





Staphlococcal enterotoxin or Toxic Shock Syndrome Toxin are superantigens that bind MHC II and one or a few Vβ families

The massive release of cytokines blocks an adaptive response and causes systemic toxicity

Block immunosurveillance function

- Inhibition of class I expression

  Cytomegalovirus, HIV, Herpes simplex, Adenovirus

- Inhibition of TAP transport of peptides

  Herpes simplex

- Proteasomal blockade

  Epstein Barr virus EBNA