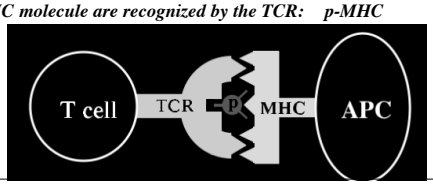
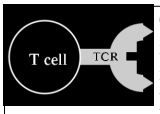
Adaptive immunity: How T cells recognize antigen

• Each clonal T cell receptor (TCR) is specific for a particular sequence of amino acids in a small peptide antigen (9-16 amino acids)

• The peptide is generated from proteins in antigen-presenting cells, where it bind to MHC (Major Histocompatibility Complex) molecules

• ***Both the amino acids of the bound peptide and the presenting MHC molecule are recognized by the TCR: p-MHC***





Generation of the repertoire of T cell clones in the adaptive immune system of each individual presents three challenges:

First

The great number of pathogen peptides

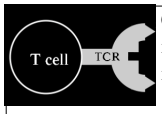
Peptides of 10 amino acids in length

20 amino acids

# of different peptides = 20^{10} =~ $\mathbf{10^{13}}$

Require **> 10^{13}** T cell clones each with different TCRs to recognize this array of peptides presented by different MHC molecules

Solution: a ***somatic recombination mechanism*** to generate the large number of structurally diverse clonal TCRs, not enough DNA in genome to encode this number of different TCR genes

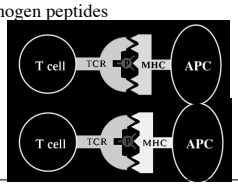


Generation of the repertoire of T cell clones in the adaptive immune system of each individual presents three challenges:

Second

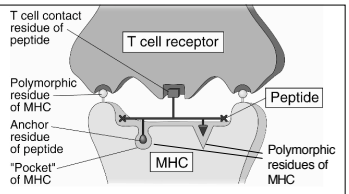
Microbial pathogens can mutate around a stereotyped defense recognition system

Solution: evolve many alternative forms of MHC molecules that bind completely different pathogen peptides



P-MHC

(HLA)

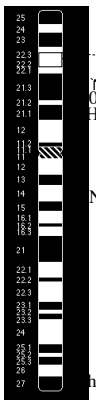


• Adaptive immune system based on differences of *MHC molecules among individuals* that confer specificity for different peptides

• The specificity of peptide binding is determined by *pockets* in the MHC molecules that only bind certain amino acid side chains

• This evolutionary strategy equips the species with a *large number of alternative MHC molecules that differ in their binding pockets*, and thus bind and present different peptides

• This results in MHC genes being extremely polymorphic



Strategy: duplicate loci and increase number of alleles

0 1 2 3 4m bp

HLA-A HLA-B HLA-C HLA-DR HLA-DP HLA-DQ

Number of alleles

451	238	789	522	34; 71	23; 124

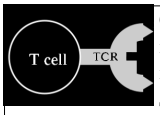
Possible Combinations of alleles

84,689,682

304,360,151,838,000,000

n 6

Human Leukocyte Antigens (HLA)



Generation of the repertoire of T cell clones in the adaptive immune system of each individual presents three challenges:

Three

The adaptive immune system must develop T cell clones that specifically bind and recognize pathogen peptides prior to encountering the pathogen

Solution: Use self-peptides as a surrogate for pathogen peptides

Problem: ***the TCR of randomly generated T cell clones could either be incapable of recognizing one's own MHC, or alternatively strongly recognize self-peptides presented in self MHC***

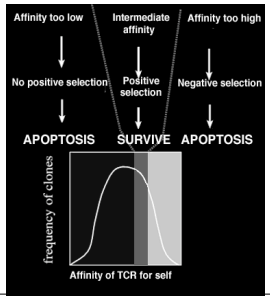
This requires a ***clonal selection process*** centered in the thymus and driven by self p-MHC to select the repertoire of clones with TCR appropriate for the self-MHC and self-peptides of each individual

Non-reactive against self ("Tolerance")

Reactive against non-self

The selection process (Thymic "education") has two stages that occur during T cell development

• **First stage** selects T cell clones that recognize self-peptide in an individual's own MHC molecules - ***positive selection***



• **Second stage *negative selection*** eliminates *overtly* self reactive clones with high affinity for self-peptide- MHC **…"central tolerance"**

(Self-peptides are used as a surrogate for foreign peptides)

The result of thymic selection is a T cell repertoire that recognizes, but does not overtly react with self

Immunologic self is the set of self-peptides and self-MHC molecules that generates and is recognized by the individual's adaptive immune T cell repertoire

One of the major functions of the innate immune system natural killer (NK) cell population is to detect decreases in the expression of the MHC portion of "self" p-MHC

The set of self-MHC molecules varies from individual to individual because of MHC polymorphism

Accordingly, the total TCR repertoire selected on self peptide-self MHC is nearly unique for each individual

• Major selective advantages to the species since there is essentially no set of stereotyped recognition structures shared by different individuals in the species

• Other individuals of the same species inherit different MHC alleles and their cells and tissues are recognized as non-self and attacked as if they were pathogens…Histocompatibility

• However because the adaptive immune system is patterned on self, it sets the stage for the development of autoimmune disease

Primary immune response

The T cell clones *generated by selection on self-peptides* that recognize, but are relatively unresponsive to self (tolerance), are then used in each adaptive immune response to identify non-self peptides typically encoded by pathogens

The non-self peptides are analogously presented by self-MHC molecules and are recognized by TCR of T cell clones as "not quite-self" (altered self) when triggered by innate immune signals, resulting in T cell activation

Types of surveillance for pathogen peptides

There are fundamentally two classes of pathogens that the immune system must recognize and respond to: viruses and bacteria

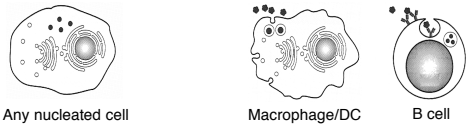
• A viral peptide on a cell's MHC molecules signifies to a T cell that it is infected and should be killed

• A bacterial peptide on a phagocytic cell that ingested a bacterium signifies to a T cell the phagocyte has ingested a foreign substance and must be helped to eliminate the pathogen by the activated T cell

Two different classes of MHC molecules direct the different immune responses to the two different pathogen types in this surveillance

Virus - or Pathogen - infected cell

Bacteria or components of an extracellular pathogen that have been phagocytized

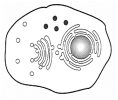
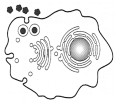


Any nucleated cell | Macrophage/DC | B cell

Peptide presented on MHC class I molecules

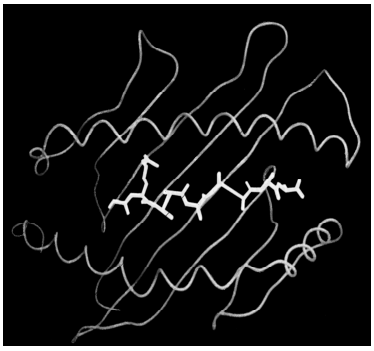
Peptide presented on MHC class II molecules

The immune system makes this distinction by loading and recognizing peptides in either class I or class II MHC

Challenge:	Cytosolic Virus or Pathogen	Ingested Bacteria or Endocytic Pathogen	Extracellular Pathogen or Toxin
			
Presenting cell:	Any cell	Macrophage/DC	B cell
Peptide degraded in:	Cytosol	Endocytic vesicles	Endocytic vesicles
Peptides bind to:	**MHC class I**	**MHC class II** (or I)	**MHC class II**
Presented to:	CD8 T cells	CD4 T cells (or CD8)	CD4 T cells
Effect on presenting cell of T cell recognition:	Death of cell presenting the viral antigen	Activation of cell to enhance pathogen killing	Provision of help to B cell for production of antibodies

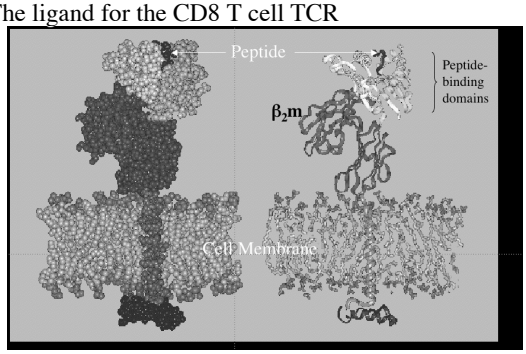
Class I and II MHC molecules

Structural features that determine peptide binding



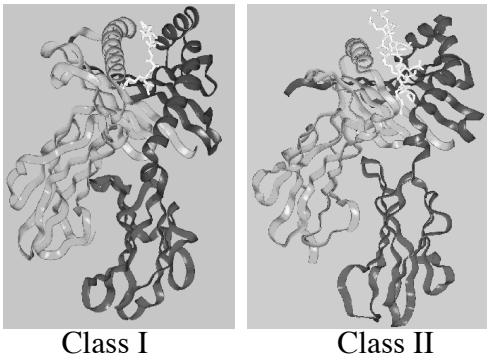
Structure of peptide-binding class I MHC domain

The ligand for the CD8 T cell TCR



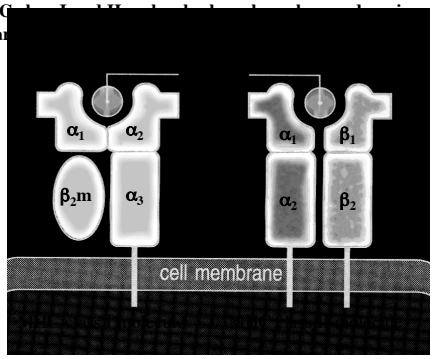
MHC Class I Domains

The overall structure of class I and class II MHC is rather similar

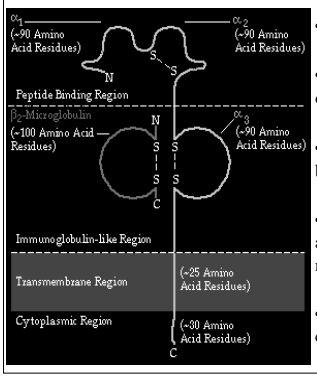


Class I | Class II

MHC … organ

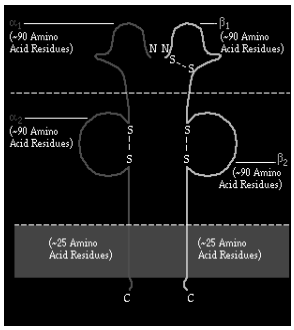


The Structure of MHC Molecules: MHC Class I



- The α chain is~ 350 AA long
- Three globular domains, α1, α2 and α3, each ~90AA
- α1 and α2 form the antigen-binding cleft
- β_2 microglobulin ~100AA, associates with the α3 domain, not MHC encoded
- ~ 70 AA transmembrane and cytoplasmic portion

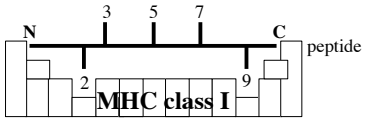
The Structure of MHC Molecules: MHC Class II



- Composed of two similar membrane spanning proteins, the α-chain and β−chain both encoded within the MHC
- Each chain is made of two globular domains, each ~90AA
- α1 and β1 domains form the antigen-binding cleft

TCR

How peptides bind



Rules for binding to MHC class I molecules

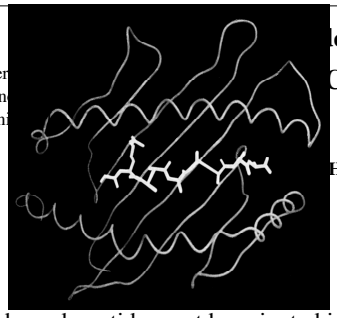
- Usually peptides are 9 amino acids in length
- Always oriented with NH2 terminus to the left
- Most often are anchored by interactions of the side chains of their 2nd (P2) and 9th (P9) amino acids to MHC pockets that confers specificity for amino acids with similar physical properties, e.g. size, charge, hydrophobicity, etc.

Cluster
tyrosin
recogni

e orientation

Class I MHC

H

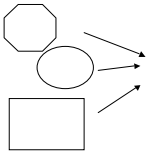


+ charged amino acids interact with negative COOH

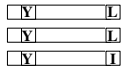
The bound peptide must be oriented in the same direction in the MHC to allow the TCR clone specific for the peptide to identify it

Rules for peptide binding to MHC class I molecules

Role of side chains



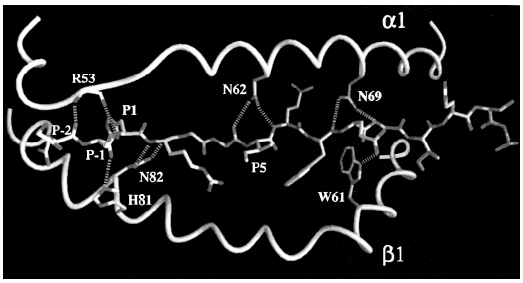
3 Different Proteins yield 3 different peptides that can bind to the same MHC molecule



Y=Tyr
L=Leu
I=Ile

A MHC Class I molecule selects homologous peptides derived from different proteins that have P2 and P9 side chains composed of homologous amino acids, e.g. tyrosine and leucine or isoleucine

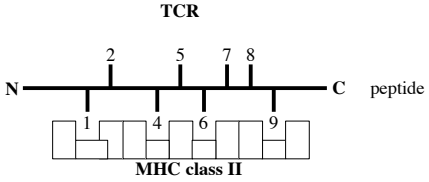
MHC class II molecule binding a peptide



Class II MHC molecules are only constitutively expressed on "professional" antigen presenting cells: DC, macrophages and B cells

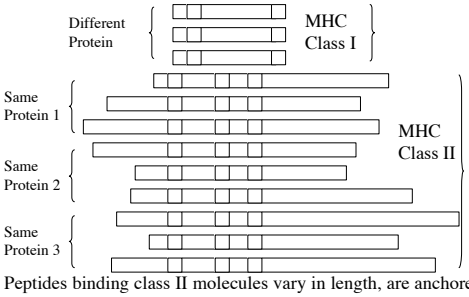
How peptides bind

Rules for binding to MHC class II molecules



- Side chains in the middle of the peptide tether it to pockets via multiple hydrogen bonds, van der Waals and electrostatic forces
- The peptide ends are free and the peptide length is variable
- Interactions with the peptide backbone orient the peptide as in class I molecules

Different rules for peptide binding to class II MHC molecules

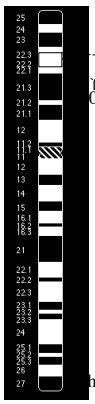


Peptides binding class II molecules vary in length, are anchored in the middle, but are also always oriented with NH2 termini to the left

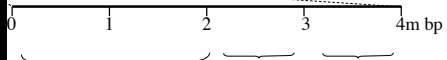
Genetic polymorphisms of MHC genes

HLA Genetics

Organization of the MHC



Two classes of peptide presenting MHC molecules are encoded by the HLA ABC and D genes



Class I	Class III	Class II
HLA-A	C4A	HLA-DR
HLA-B	C4B	HLA-DQ
HLA-C	C2	HLA-DP
	Bf	
	TNF-α	

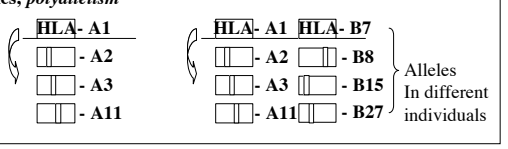
n 6

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*



MHC polymorphism is all about survival, it is an evolutionary response to the structural diversity and mutation potential of microorganisms

No practical biologic limit on the number of alleles for the species

Frequency-dependant selection- The individual with the rarest allele has the best chance to survive an infection

Duplication of a locus incurs a risk

- Each duplication results in a new set of antigen-presenting structures
- Each MHC type selects its own allele-specific TCR clonal repertoire capable of recognizing additional pathogen peptides
- However, each duplication increases the size of immune self and mandates more negative clonal selection across all repertoires during repertoire formation, reducing the size of the repertoire for each allele

Practical maximum is ~ three loci each for class I and class II

HLA-DR	HLA-A
HLA-DQ	HLA-B
HLA-DP	HLA-C

(Remember both maternal and paternal alleles are expressed)

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 in different individuals

Allotypes or allomorphs: the 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

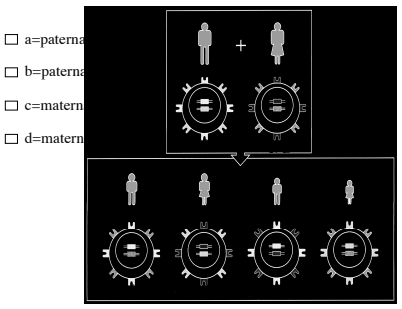
Linkage disequilibrium: certain alleles in a haplotype are found together significantly more (or less) frequently than expected by chance

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

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

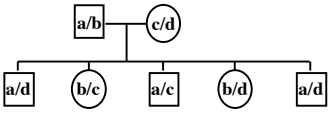
But to make things "simpler", alleles can be grouped in families, e.g. HLA-B*27
"specificity", is an old nomenclature used when human alloantibodies were used to first detect HLA serologic "specificities" or "antigens"

Codominant expression of MHC alleles



HLA genetics in transplantation

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

In situations where a transplant is required, the family is first typed to find 2 haplotype matches, then unrelated individuals are studied