Antigen capture and presentation to T lymphocytes

What T lymphocytes see
Innate Immunity

Immediately available or rapidly recruited

Very broad specificity
Adaptive Immunity

Rare and naïve cells require priming and expansion (i.e. a primary response takes time to develop)

- **Narrow specificity**
- **Clonally distribution**
- **Clonal selection**
Clonal Distribution & Selection

Each lymphocyte (B or T cell) expresses one receptor specificity (clonally distributed)

Each of these cells (i.e. specificities) can be silenced or promoted (clonally selected)
Control cells = control specificity
What is a good target for the adaptive immune system?

To be seen – targets must be accessible and easy to identify

To allow discrimination between self and foreign – targets must be highly variable

To avoid escape – targets must be difficult to conceal, change or remove

PROTEINS FULFILL THESE REQUIREMENTS – ACTUALLY PEPTIDES DO
The World of Peptide Antigens

Number of different peptides = \(20^N\)
where \(N =\) length of peptide

The universe of 9-mers = \(512 \times 10^9\) peptides
The human proteome \(\approx 12 \times 10^6\) peptides
i.e. plenty of discriminatory power in 9-mers
Questions

• How are source proteins captured?

• How are peptides generated?

• How are peptides displayed (presented)
Questions

• T cells of the appropriate specificity are rare - how do T cells find the antigen?

• The cellular location of a threat is important – how do T cells determine this location?

• A UNIFIED ANSWER: ANTIGEN PRESENTATION
Antigens Recognized by T Lymphocytes

T cell contact residue of peptide

Polymorphic residue of MHC

Anchor residue of peptide

Pocket of MHC

T cell receptor

MHC

Peptide

MHC RESTRICTION

Abbas et al: Basic Immunology, 4e
Capture & Display of Microbial Antigens

Immature DC

Antigen Capture

Mature DC

Antigen Presentation

Skin
Gastrointestinal tract
Respiratory tract

Microbe
Epithelium

Cell-free antigen
Dendritic cell-associated antigen

Lymphatic vessel
Connective tissue

To lymph node
To circulation and spleen

Lymph node collects antigen from epithelium and connective tissue
Blood-borne antigens are captured by antigen-presenting cells in the spleen

June 4th, 2015

Abbas et al: Basic Immunology, 4e
Immature and Mature Dendritic cells

A. Immature DC
   - Antigen Capture
   - Dendritic cell (Langerhans cell) in epidermis

B. Mature DC
   - Antigen Presentation
   - Follicle
   - Dendritic cell in lymph node

Abbas et al. Basic Immunology, 4e
Crude Recognition of Microbes

Encoded in germline; limited diversity
(pattern recognition receptors)

- Toll-like receptor
- N-formyl peptide receptor
- Mannose receptor
- Scavenger receptor
## Dendritic cells – two major classes

<table>
<thead>
<tr>
<th>Feature</th>
<th>Conventional dendritic cells</th>
<th>Plasmacytoid dendritic cells</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Surface markers</strong></td>
<td>CD11c high CD11b high</td>
<td>CD11c low CD11b negative B220 high</td>
</tr>
<tr>
<td><strong>Major location</strong></td>
<td>Tissues</td>
<td>Blood and tissue</td>
</tr>
<tr>
<td><strong>Expression of Toll-like receptors</strong></td>
<td>TLRs 4, 5, 8 high</td>
<td>TLRs 7, 9 high</td>
</tr>
<tr>
<td><strong>Major cytokines produced</strong></td>
<td>TNF, IL-6, IL-12</td>
<td>Type I interferons</td>
</tr>
<tr>
<td><strong>Postulated major functions</strong></td>
<td>Induction of T cell responses against most antigens</td>
<td>Antiviral innate immunity and induction of T cell responses against viruses</td>
</tr>
</tbody>
</table>

Capture & Presentation by DC’s

Antigen capture by dendritic cells (DCs) → Activation of dendritic cells

Antigen capture

DC in epidermis: phenotypically immature → TLR ligands, cytokines

Migration of DC

Maturation of migrating DC

Antigen presentation

T cell in epidermis: maturation

T cell

Mature dendritic cell presenting antigen to naive T cell

Lymph node
# Antigens Presenting Cells (APC)

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Expression of Class II MHC</th>
<th>Expression of Costimulators</th>
<th>Principal function</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dendritic cells</td>
<td>Constitutive; increases with maturation; increased by IFN-γ</td>
<td>Constitutive; increases with maturation; inducible by TLR ligands, IFN-γ, and T cells (CD40-CD40L interactions)</td>
<td>Initiation of T cell responses to protein antigens</td>
</tr>
<tr>
<td>Macrophages</td>
<td>Low or negative; inducible by IFN-γ</td>
<td>Low, inducible by TLR ligands, IFN-γ, and T cells (CD40-CD40L interactions)</td>
<td>Effector phase of cell-mediated immune responses</td>
</tr>
<tr>
<td>B lymphocytes</td>
<td>Constitutive; increased by IL-4</td>
<td>Induced by T cells (CD40-CD40L interactions), antigen receptor cross-linking</td>
<td>Antigen presentation to CD4+ helper T cells in humoral immune responses (cognate T cell–B cell interactions)</td>
</tr>
</tbody>
</table>

*Abbasi et al: Basic Immunology, 4e
What are MHC molecules?
MHC (HLA) gene region
MHC / HLA polymorphism

- The most polymorphic gene region known
  - About 3500 different HLA class I registered
  - About 4500 different HLA class II registered

Gene complexity at the MHC locus in man

<table>
<thead>
<tr>
<th>Class I</th>
<th>Class II</th>
</tr>
</thead>
<tbody>
<tr>
<td>gene</td>
<td>alleles</td>
</tr>
<tr>
<td>HLA-A</td>
<td>1,519</td>
</tr>
<tr>
<td>HLA-B</td>
<td>2,069</td>
</tr>
<tr>
<td>HLA-C</td>
<td>1,016</td>
</tr>
<tr>
<td>HLA-E</td>
<td>10</td>
</tr>
<tr>
<td>HLA-F</td>
<td>22</td>
</tr>
<tr>
<td>HLA-G</td>
<td>46</td>
</tr>
</tbody>
</table>

Data from the European Bioinformatics Institute (EBI) server ([http://www.ebi.ac.uk/imgt/hla/stats.html](http://www.ebi.ac.uk/imgt/hla/stats.html))
Structure of MHC / HLA molecules

Class I

Class II

June 4th, 2015
## Features of MHC genes and molecules

<table>
<thead>
<tr>
<th>Feature</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Co-dominant expression:</td>
<td>Both parental alleles of each MHC gene are expressed</td>
</tr>
<tr>
<td></td>
<td>Increases number of different MHC molecules that can present peptides to T cells</td>
</tr>
</tbody>
</table>

- **T cells**
- **MHC molecules**
- **Parental chromosomes**
Features of MHC genes and molecules

Polymorphic genes:
Many different alleles are present in the population

Ensures that different individuals are able to present and respond to different microbial peptides
Features of MHC genes and molecules

MHC-expressing cell types:
- **Class II:** Dendritic cells, macrophages, B cells
  - CD4⁺ helper T lymphocytes interact with dendritic cells, macrophages, B lymphocytes
- **Class I:** All nucleated cells
  - CD8⁺ CTLs can kill any virus-infected cell
Binding of Peptides to MHC

MHC class I closed
Peptide short

MHC class II open
Peptide longer
# Peptide interaction with MHC

<table>
<thead>
<tr>
<th>Feature</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Broad specificity</td>
<td>Many different peptides can bind to the same MHC molecule</td>
</tr>
<tr>
<td>Each MHC molecule displays one peptide at a time</td>
<td>Each T cell responds to a single peptide bound to an MHC molecule</td>
</tr>
<tr>
<td>MHC molecules bind only peptides</td>
<td>MHC-restricted T cells respond only to protein antigens, and not to other chemicals</td>
</tr>
</tbody>
</table>

- **Proteins**
- **Lipids**
- **Carbohydrates**
- **Nucleic acids**
Peptide interaction with MHC

Peptides are acquired during intracellular assembly.

Class I and class II MHC molecules display peptides from different cellular compartments.

Peptide in endocytic vesicle

\[ \alpha + \beta + \text{Ii} \rightarrow \alpha + \beta + \text{Class II MHC} \]

\[ \beta_2\text{-microglobulin} + \alpha + \text{Cytosolic peptide, transported into ER} \rightarrow \alpha + \text{Class I MHC} \]}
Peptide interaction with MHC

Stable surface expression of MHC molecule requires bound peptide

Only peptide-loaded MHC molecules are expressed on the cell surface for recognition by T cells

Very slow off-rate

MHC molecule displays bound peptide for long enough to be located by T cell

β2-microglobulin + α + Peptide + Days → MHC molecule with bound peptide

Empty MHC molecule
Peptide interaction with MHC

MHC samples intracellular peptides.

They do NOT discriminate between self and non-self
MHC class II mediated antigen processing

- Uptake of extracellular proteins into vesicular compartments of APC
- Processing of internalized proteins in endosomal/lyosomal vesicles
- Biosynthesis and transport of class II MHC molecules to endosomes
- Association of processed peptides with class II MHC molecules in vesicles
- Expression of peptide-MHC complexes on cell surface
MHC class I mediated antigen processing
# Two Antigen Processing Pathways: one for each class of MHC

<table>
<thead>
<tr>
<th>Feature</th>
<th>Class II MHC Pathway</th>
<th>Class I MHC pathway</th>
</tr>
</thead>
<tbody>
<tr>
<td>Composition of stable peptide-MHC complex</td>
<td>Polymorphic $\alpha$ and $\beta$ chains of MHC, peptide</td>
<td>Polymorphic $\alpha$ chain of MHC, $\beta 2$-microglobulin, peptide</td>
</tr>
<tr>
<td></td>
<td><img src="peptide_diagram.png" alt="Peptide Diagram" /></td>
<td><img src="peptide_diagram.png" alt="Peptide Diagram" /></td>
</tr>
<tr>
<td>Cells that express MHC</td>
<td>Dendritic cells, mononuclear phagocytes, B lymphocytes; endothelial cells, thymic epithelium</td>
<td>All nucleated cells</td>
</tr>
<tr>
<td>Responsive T cells</td>
<td>CD4$^+$ T cells</td>
<td>CD8$^+$ T cells</td>
</tr>
</tbody>
</table>
Two Antigen Processing Pathways: one for each class of MHC

<table>
<thead>
<tr>
<th>Feature</th>
<th>Class II MHC Pathway</th>
<th>Class I MHC Pathway</th>
</tr>
</thead>
<tbody>
<tr>
<td>Source of protein antigens</td>
<td>Endosomal/lysosomal proteins (mostly internalized from extracellular environment)</td>
<td>Cytosolic proteins (mostly synthesized in the cell; may enter cytosol from phagosomes)</td>
</tr>
<tr>
<td>Enzymes responsible for peptide generation</td>
<td>Endosomal and lysosomal proteases (e.g., cathepsins)</td>
<td>Cytoplasmic proteasome</td>
</tr>
<tr>
<td>Site of peptide loading of MHC</td>
<td>Specialized vesicles</td>
<td>Endoplasmic reticulum</td>
</tr>
<tr>
<td>Molecules involved in transport of peptides</td>
<td>Invariant chain, DM</td>
<td>TAP</td>
</tr>
<tr>
<td>and loading of MHC molecules</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Detecting the cellular antigen location

**Antigen uptake or synthesis**

**Class II MHC–associated presentation of extracellular antigen to helper T cells**

- Macrophage
- Extracellular antigen
- Antigen in endosome
- CD4+ helper T lymphocyte
- Cytokines
- Macrophage activation: destruction of phagocytosed antigen
- B cell antibody secretion: antibody binding to antigen

**Class I MHC–associated presentation of cytosolic antigen to cytotoxic T lymphocytes**

- Cytosolic antigen
- CD8+ cytotoxic T lymphocyte
- Killing of antigen-expressing target cell

**Antigen presentation**

**T cell effector functions**
Cross-presentation

Infected cells and viral antigens picked up by host APCs

Viral antigen

Dendritic cell

Virus-specific CD8+ T cell

Costimulator

Antigen capture

Cross-presentation

T cell response

Abbas et al: Basic Immunology, 4e
T cell recognition

• MHC molecules sample peptides from the cellular protein metabolism, and T cells recognize peptide/MHC complexes in a cell-cell interaction

• Priming requires presentation AND co-stimulation
T cell recognition

- MHC’s do NOT discriminate between self and non-self – T cells do

- T cells do NOT discriminate between peptides of intra or extra-cellular protein origin – MHC pathways do
HLA polymorphism and immune specificity

Epitope Universe

Self

Foreign
To be, or not to be - encrypted
HLA polymorphism individualizes T cell responses
HLA polymorphism mismatch causes allo-responses

Epitope Universe

Self
Allo-recognition in bone-marrow transplantation

Donor TcR

Donor APC

Auto tolerance
Allo-recognition in bone-marrow transplantation

Donor TcR

Perfectly matched APC

Auto tolerance
Allo-recognition in bone-marrow transplantation

Donor TcR

Perfectly matched APC

Auto tolerance

Donor TcR

HLA unmatched patient APC

Major Histo-incompatibility
Allo-recognition in bone-marrow transplantation

Donor TcR

Perfectly matched APC
Auto tolerance

Donor TcR

HLA unmatched patient APC
Major Histo-incompatibility

Donor TcR

HLA matched patient APC
Minor Histo-incompatibility

October 4th, 2012
2012 Annual GSI meeting, Sandbjerg
Slide 45
Altered self-repertoire = equivalent of allo-response
B cell recognition

• Do NOT require MHC mediated antigen processing and presentation
• Use FDC for antigen display
• Recognizes targets of many kinds / intact structures
• May use a soluble receptor
• Recognize targets in the extracellular space