Lecture 6: The Humoral Immune Response
(based on lecture by Dr. Matthew Scharff, Einstein)
Questions to Consider

- How can we make antibody to every possible pathogen—i.e. Diversity
- How do we avoid making autoantibodies—i.e. Specificity
- How do we rapidly increase amount of antibody—i.e. Mobilization
- How do we switch from making IgM to IgG—i.e. Isotype Switching
- How do we increase the affinity of antibody—i.e. Affinity maturation
- How do we generate memory
Humoral Immune Response

Class Switch Recombination

IgM → IgG / IgE / IgA

Antigen-binding site

Effector arm

Somatic Hypermutation
Low affinity → high affinity

Fig 10.25 © 2001 Garland Science

Janeway and Travers, Immunobiology
Antibody Mediated Functions

B-cell activation by antigen and helper T cells

Antibody secretion by plasma cells

Neutralization
- Antibody prevents bacterial adherence

Opsonization
- Antibody promotes phagocytosis

Complement activation
- Antibody activates complement, which enhances opsonization and lyses some bacteria

Figure 9-1 Immunobiology, 6/e. (© Garland Science 2005)
IgM is Polymeric Which Increases Its Avidity
Neutralization of Viruses

B cell binds virus through viral coat protein

Peptides from internal proteins of the virus are presented to the T cell, which activates the B cell

Virus particle is internalized and degraded

Helper T cell

Activated B cell produces antibody against viral coat protein

CD154 (CD40L)

CD40

cytokines

Figure 9-3 Immunobiology, 6/e. (© Garland Science 2005)
Antibodies Neutralize Toxins

Toxin binds to cellular receptors

Endocytosis of toxin:receptor complexes

Dissociation of toxin to release active chain, which poisons cell

Antibody protects cell by blocking binding of toxin

Figure 9-24 Immunobiology, 6/e. (© Garland Science 2005)
Binding of Antigen to Surface Ig Triggers The Proliferation And Differentiation of B Cells

Antigen recognition induces expression of effector molecules by the T cell, which activates the B cell.

B cell proliferation

Differentiation to resting memory cells and antibody-secreting plasma cells

Figure 9-5 Immunobiology, 6/e. (© Garland Science 2005)
Generation of Antibodies To Polysaccharides by Conjugation to Proteins (Glycoconjugate Vaccines)

1. B cell binds bacterial polysaccharide epitope linked to tetanus toxoid protein
2. Antigen is internalized and processed
3. Peptides from protein component are presented to the T cell
4. Activated B cell produces antibody against polysaccharide antigen on the surface of the bacterium
Circulating Antibodies Reflect Somatic Mutation And Isotype Switching

![Graph showing antibody levels and affinity over time after immunization](image-url)
Ig Isotype Are Encoded by Unique Constant Regions

Figure 4-17 Immunobiology, 7ed. (© Garland Science 2008)
Ig Isotypes Have Different Functions Due to Their Unique Constant Regions

<table>
<thead>
<tr>
<th>Functional activity</th>
<th>IgM</th>
<th>IgD</th>
<th>IgG1</th>
<th>IgG2</th>
<th>IgG3</th>
<th>IgG4</th>
<th>IgA</th>
<th>IgE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neutralization</td>
<td>++</td>
<td>-</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>-</td>
</tr>
<tr>
<td>Opsonization</td>
<td>++</td>
<td>-</td>
<td>++++</td>
<td>*</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Sensitization for killing by NK cells</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Sensitization of mast cells</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+++</td>
</tr>
<tr>
<td>Activates complement system</td>
<td>+++</td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>+++</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Distribution</th>
<th>IgM</th>
<th>IgD</th>
<th>IgG1</th>
<th>IgG2</th>
<th>IgG3</th>
<th>IgG4</th>
<th>IgA</th>
<th>IgE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transport across epithelium</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+++</td>
<td>-</td>
</tr>
<tr>
<td>Transport across placenta</td>
<td>-</td>
<td>-</td>
<td>+++</td>
<td>+</td>
<td>++</td>
<td>+/-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Diffusion into extravascular sites</td>
<td>+/-</td>
<td>-</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+</td>
</tr>
<tr>
<td>Mean serum level (mg ml(^{-1}))</td>
<td>1.5</td>
<td>0.04</td>
<td>9</td>
<td>3</td>
<td>1</td>
<td>0.5</td>
<td>2.1</td>
<td>(3 \times 10^{-5})</td>
</tr>
</tbody>
</table>
IgM And IgA Are Polymeric Which Increases Their Avidity

Figure 4-23 Immunobiology, 6/e. (© Garland Science 2005)
Binding of Antigen to Surface Ig Triggers The Proliferation And Differentiation of B Cells

Antigen recognition induces expression of effector molecules by the T cell, which activates the B cell

B cell proliferation

Differentiation to resting memory cells and antibody-secreting plasma cells

Figure 9-5 Immunobiology, 6/e. (© Garland Science 2005)
Different Cytokines Secreted By T Cells Induce Switching To Different Isotypes

### Role of cytokines in regulating Ig isotype expression

<table>
<thead>
<tr>
<th>Cytokines</th>
<th>IgM</th>
<th>IgG3</th>
<th>IgG1</th>
<th>IgG2b</th>
<th>IgG2a</th>
<th>IgE</th>
<th>IgA</th>
</tr>
</thead>
<tbody>
<tr>
<td>IL-4</td>
<td>Inhibits</td>
<td>Inhibits</td>
<td>Induces</td>
<td></td>
<td>Inhibits</td>
<td>Induces</td>
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<tr>
<td>IL-5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Augments production</td>
</tr>
<tr>
<td>IFN-γ</td>
<td>Inhibits</td>
<td>Induces</td>
<td>Inhibits</td>
<td></td>
<td>Induces</td>
<td>Inhibits</td>
<td></td>
</tr>
<tr>
<td>TGF-β</td>
<td>Inhibits</td>
<td>Inhibits</td>
<td></td>
<td></td>
<td>Induces</td>
<td></td>
<td>Induces</td>
</tr>
</tbody>
</table>

*Figure 9-7 Immunobiology, 6/e. (© Garland Science 2005)*
Different Cytokines Signal Transcription of Different C-regions (Sterile Transcripts)
Activation-induced Cytidine Deaminase (AID)

- AID is a cytidine deaminase whose *in vitro* substrate is ssDNA
- AID may associate with RPA, RNAP II & other proteins
- Transcription is required for somatic SHM and CSR

Deficiency causes Hyper IgM type II


- AID is a cytidine deaminase whose *in vitro* substrate is ssDNA
- AID may associate with RPA, RNAP II & other proteins
- Transcription is required for somatic SHM and CSR
Activation-induced Cytidine Deaminase (AID) Mediates Class Switching

Figure 4-27 Immunobiology, 7ed. (© Garland Science 2008)
Isotype Switching Requires Recombination Between Different Switch Regions

Figure 4-21 Immunobiology, 6/e. (© Garland Science 2005)
The Poly-Ig Receptor Mediates the Transcytosis Of IgA Into Mucosal Secretions
Cells Express Multiple Fc Receptors With Unique Functions

<table>
<thead>
<tr>
<th></th>
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<th></th>
<th></th>
<th></th>
<th></th>
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</tr>
</thead>
<tbody>
<tr>
<td><strong>Structure</strong></td>
<td>α 72 kDa</td>
<td>α 40 kDa</td>
<td></td>
<td></td>
<td>α 50–70 kDa</td>
<td>β 33 kDa</td>
<td>α 55–75 kDa</td>
<td>α 70 kDa</td>
</tr>
<tr>
<td></td>
<td>γ</td>
<td>γ-like domain</td>
<td>ITIM</td>
<td>ITIM</td>
<td>γ or ζ</td>
<td>γ or ζ</td>
<td>γ 9 kDa</td>
<td></td>
</tr>
<tr>
<td><strong>Binding</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Order of affinity</strong></td>
<td>IgG1 10^8 M^{-1}</td>
<td>IgG1 2 × 10^6 M^{-1}</td>
<td>IgG1 2 × 10^6 M^{-1}</td>
<td>IgG1 2 × 10^6 M^{-1}</td>
<td>IgG1 5 × 10^5 M^{-1}</td>
<td>IgE 10^10 M^{-1}</td>
<td>IgA, IgA2 10^7 M^{-1}</td>
<td>IgA, IgM 3 × 10^9 M^{-1}</td>
</tr>
<tr>
<td><strong>Cell type</strong></td>
<td>Macrophages, Neutrophils, Eosinophils, Dendritic cells</td>
<td>Macrophages, Neutrophils, Eosinophils, Platelets, Langerhans cells</td>
<td>Macrophages, Neutrophils, Eosinophils</td>
<td>B cells, Mast cells, NK cells, Eosinophils</td>
<td>Mast cells, Eosinophils, Basophils, Macrophages, Neutrophils</td>
<td>Mast cells, Eosinophils, Basophils, Mast cells</td>
<td>Macrophages, Eosinophils, Neutrophils</td>
<td>Macrophages, B cells</td>
</tr>
<tr>
<td><strong>Effect of ligation</strong></td>
<td>Uptake, Stimulation, Activation of respiratory burst, Induction of killing</td>
<td>Uptake, Granule release (eosinophils)</td>
<td>Uptake, Inhibition of stimulation</td>
<td>No uptake, Inhibition of stimulation</td>
<td>Induction of killing (NK cells)</td>
<td>Secretion of granules</td>
<td>Uptake, Induction of killing</td>
<td>Uptake</td>
</tr>
</tbody>
</table>

Figure 8.30: Immunobiology, 7ed. (© Garland Science 2008)
Antibody Complexed To Antigen Binds to FcR’s and Activates Macrophages

Free immunoglobulin does not cross-link Fc receptors

Aggregation of immunoglobulin on bacterial surface allows cross-linking of Fc receptors

No activation of macrophage, no destruction of bacterium

Activation of macrophage, leading to phagocytosis and destruction of bacterium

Figure 9-31 Immunobiology, 6/e. (© Garland Science 2005)
Antibody Complexed To Tumor Cells Targeting Their Killing by NK Cells Through FcRs

Figure 9-34 Immunobiology, 6/e. (© Garland Science 2005)
IgE Complexed To Antigen Binds to FcR’s on Mast Cells and Mediates The Release Of Granules

Resting mast cell

Activated mast cell

Resting mast cell contains granules containing histamine and other inflammatory mediators

Multivalent antigen cross-links bound IgE antibody, causing release of granule contents

Figure 9-35 Immunobiology, 6/e. (© Garland Science 2005)
Questions to Consider

- How can we make antibody to every possible pathogen-i.e. Diversity
- How do we avoid making autoantibodies-i.e. Specificity
- How do we rapidly increase amount of antibody-i.e. Mobilization
- How do we switch from making IgM to IgG- i.e. Isotype Switching
- How do we increase the the affinity of antibody-i.e. Affinity maturation
- How do we generate memory