Antibodies and the adaptive immune system
General terms to know

• Pathogen
  – Germ = disease causing agent
  – Examples: virus, bacterium, parasite, fungus, prion

• Antigen
  – Antibody generator = substances that elicit an antibody immune response
  – Examples: bacterial LPS, bacterial flagella, virus surface protein, parasite glycans, protein toxins

• Antibody
  – Immunoglobulin = a specific Y-shaped protein produced by B-cells that binds to a specific antigen
  – Examples: IgG, IgA, IgM, IgD, IgM
General terms to know
How does the human body react to an infection? --> Immune system

**Innate Immunity**
- Epithelial barriers
- Phagocytes
- Dendritic cells
- Plasma proteins
- NK cells

**Adaptive Immunity**
- Naive B cell
- Naive T cell
- Antibodies
- Effector T cells

**Short term response**
- i.e. fever, inflammation

**Developed recognition of pathogen**
- i.e. lifelong immunity
How does the human body react to a virus infection? --> Immune system

**Innate Immunity**
- Castle
- Moat
- Alligators = macrophages

**Adaptive Immunity**
- Archers
- Cannonneers
- Swordsmen

Developed from birth
Always ready, Non-learner
Non-specific

Developed from experience
Adaptor, Learner, Improver
Specific for virus/pathogen
How does the human body react to an infection? --> Immune system

**Innate Immunity**
- Epithelial barriers
- Phagocytes
- Dendritic cells
- Plasma proteins
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**Adaptive Immunity**
- Naive B cell
- Antibodies
- Naive T cell
- Effector T cells

Short term response i.e. fever, inflammation
Developed recognition of pathogen i.e. lifelong immunity
Adaptive immunity: B cells and T cells

- T and B cells develop from stem cells in bone marrow

- **Humoral immunity**
  - **B cells** mature in the bone marrow

- **Cellular immunity**
  - Due to T cells
  - **T cells** mature in the thymus
T cells:

- Recognize MHC proteins on the surface of cells that are presenting (bound to) fragment of a foreign antigen
- Activated cytotoxic T cells kill infected cell
- Activated helper T cells secrete cytokines (small signaling proteins) that help to activate T cells and B cells
Each B cell produces a specific antibody

- Gene rearrangement (VDJ or VJ recombination) of both heavy and light chain antibody genes
- Allows for $>10^{12}$ different possible antibody sequences
- Membrane-bound antibody on surface of B cells
- After B cell is stimulated, secreted form of antibody made
Each B cell produces a specific antibody by VDJ and VJ recombination

- Genetic recombination of antibody genes in early stage of B cell maturation
- Mediated by a number of recombination enzymes
- Both heavy and light chain genes undergo recombination
The role of antigen becomes critical when it interacts with and activates mature, antigenically committed T and B lymphocytes, bringing about expansion of the population of cells with a given antigenic specificity. In this process of clonal selection, an antigen binds to a particular T or B cell and stimulates it to divide repeatedly into a clone of cells with the same antigenic specificity as the original parent cell (Fig 1-10).

Clonal selection provides a framework for understanding the specificity and self/nonself recognition that is characteristic of adaptive immunity. Specificity is shown because only lymphocytes whose receptors are specific for a given epitope on an antigen will be clonally expanded and thus mobilized for an immune response. Self/nonself discrimination is accomplished by the elimination, during development, of lymphocytes bearing self-reactive receptors or by the functional suppression of these cells in adults.

Immunologic memory also is a consequence of clonal selection. During clonal selection, the number of lymphocytes specific for a given antigen is greatly amplified. Moreover, many of these lymphocytes, referred to as memory cells, appear to have a longer life span than the naive lymphocytes from which they arise. The initial encounter of a naive immunocompetent lymphocyte with an antigen induces a
What do antibodies do?

- **Agglutination**: Enhances phagocytosis and reduces the number of infectious units to be dealt with.
- **Opsonization**: Coating antigen with antibody enhances phagocytosis.
- **Neutralization**: Blocks adhesion of bacteria and viruses to mucosa.
- **Activation of complement**: Cell lysis
- **Activation of complement**: Complement
- **Inflammation**: Disruption of cell by complement/reactive protein attracts phagocytic and other defensive immune system cells.
- **Enhance killing of pathogen by cytotoxic proteins**: Macrophage

**Enhance phagocytosis and degradation of pathogen**

**Neutralize pathogen**

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Antibody Structure

Variable region
Determines specificity and affinity for antigen

Constant region
Determines antibody isotype, which can affect biological properties, functional locations,
# Antibody Isotypes

<table>
<thead>
<tr>
<th>Isotype</th>
<th>Description</th>
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<tbody>
<tr>
<td>IgA</td>
<td>Dimeric antibody found in mucosal areas (gut, lungs, genital tract, saliva, tears, breast milk) and prevents pathogen colonization.</td>
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<tr>
<td>IgD</td>
<td>Antigen receptors on B cells.</td>
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<tr>
<td>IgE</td>
<td>Binds allergens and triggers histamine release.</td>
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<td>IgG</td>
<td>Majority of antibody-based immunity against pathogens. Only antibody that can cross placenta to protect fetus.</td>
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<tr>
<td>IgM</td>
<td>Pentameric antibody with high avidity used to eliminate pathogens before there is sufficient IgG.</td>
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![Diagram of antibody structures](image)
Antibody Structure

Fc = Fragment constant
Fab = Fragment antigen binding
Fv = Fragment variable = part of Fab
One antibody has two identical antigen binding sites, each composed of six CDR loops (CDR = complementarity determining region)
Quick review – antibody structure
Immunity

• **Active Immunity**: antibodies developed as a result of antigenic stimulus

• **Passive Immunity**: antibodies transmitted passively
Active Immunity

- **Natural active immunity:** resistance to a pathogen developed during natural infection

- **Artificial active immunity:** resistance to a pathogen developed during vaccination
  - Live attenuated vaccines
  - Inactivated or subunit vaccines

Rubella virus infection (German measles)
Vaccines

- Vaccines train the adaptive immune response: long-term protective immunity against future infections
- Vaccines contain antigen(s) from a specific pathogen
- Vaccines elicit antibody production in humans
Brief History of Vaccines

- 1720, Istanbul: injection of pus from smallpox victims into skin = variolation (1 in 50 chance of dying)

- 1796, England: Edward Jenner, MD, heard claims of local dairymaids that infection with cowpox made them immune to smallpox. Dr. Jenner tested theory on 8-year old boy – it worked! Cow = vacca --> vaccination

- 1885, Louis Pasteur: dried, crushed spinal cords from dogs that had died from rabies (virus), injected into healthy dogs, did not become infected; 9-year old boy bitten by rabid dog was given injection and survived

- 1952, Jonas Salk: three strains of poliovirus (grown in cell culture) combined, inactivated with formalin (formaldehyde)
Childhood Vaccines

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<tr>
<th>Vaccines</th>
<th>Birth</th>
<th>1 mo</th>
<th>2 mos</th>
<th>4 mos</th>
<th>6 mos</th>
<th>9 mos</th>
<th>12 mos</th>
<th>15 mos</th>
<th>18 mos</th>
<th>19-23 mos</th>
<th>2-3 yrs</th>
<th>4-6 yrs</th>
<th>7-10 yrs</th>
<th>11-12 yrs</th>
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<td>RV-1 (2-dose series); RV-5 (3-dose series)</td>
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<td>Diphtheria, tetanus, &amp; acellular pertussis (DTaP: &lt;7 yrs)</td>
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<td>Tetanus, diphtheria, &amp; acellular pertussis (Tdap: ≥7 yrs)</td>
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<td>Haemophilus influenzae type b (Hib)</td>
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<td>Pneumococcal conjugate (PCV13)</td>
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<td>Pneumococcal polysaccharide (PPSV23)</td>
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<td>Inactivated poliovirus (IPV) (&lt;18 years)</td>
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<td>Annual vaccination (IV only)</td>
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**Legend:**
- Range of recommended ages for all children
- Range of recommended ages for catch-up immunization
- Range of recommended ages for certain high-risk groups
- Range of recommended ages during which catch-up is encouraged and for certain high-risk groups
- Not routinely recommended
“The easy infectious disease problems and vaccines have been solved...Now we’re left with the more difficult ones...Developing vaccines against these will require significant, new scientific insight.” -Dan Granoff, M.D., CHORI.

Spoiler alert: two upcoming classes on engineering protein vaccine antigens
Passive Immunity

- **Natural passive immunity**: maternal antibodies in colostrum/milk transferred from mother to baby
  - First ~6 months of baby’s life
  - Immunization of mothers improves passive immunity in infants

- **Artificial passive immunity**: administration of therapeutic antibodies
  - Antiserum
  - Gamma globulin (purified IgG)
  - Recombinant antibodies or antibody fragments
  - Antibodies last weeks-months
Therapeutic Antibodies

• Polyclonal: collection of many different antibodies from serum

• Monoclonal: single isolated antibody
Artificial passive immunity: ZMapp

• Mixture of three antibodies that neutralize Ebola virus
  (Neutralize = block virus from infecting cell)

• Originally isolated as mouse monoclonal antibodies

• Engineered to make chimeric human-mouse antibodies; recombinant antibodies produced in tobacco plants
Artificial passive immunity: RhoGAM

- Human blood can be Rh positive or Rh negative

- Rh factor = Rhesus factor = protein on surface of red blood cells (membrane transport protein with unknown physiological role)

- European decent: 15% Rh negative, 85% Rh positive
- African, native american, asian decent: 1% Rh negative, 99% Rh positive

- When Rh+ father and Rh- mother become pregnant, possibility of developing hemolytic disease of the newborn (HDN)
Artificial passive immunity: RhoGAM

1. Rh+ father.
2. Rh- mother carrying her first Rh+ fetus. Rh antigens from the developing fetus can enter the mother’s blood during delivery.
3. In response to the fetal Rh antigens, the mother will produce anti-Rh antibodies.
4. If the woman becomes pregnant with another Rh+ fetus, her anti-Rh antibodies will cross the placenta and damage fetal red blood cells.

• When Rh+ father and Rh- mother become pregnant, baby may have Rh+ blood

• Mother’s exposure to small amounts of baby’s Rh+ blood (usually during birth) causes Rh- mother to develop antibodies against Rh factor of baby’s blood

• If 2nd and subsequent pregnancies are with Rh+ babies, hemolytic disease of the newborn (HDN) occurs: mothers anti-Rh antibodies attack fetus blood: severe anemia, liver and heart damage: used to be responsible for death of 1 in 2,200 babies
Artificial passive immunity: RhoGAM

- RhoGAM = anti-Rh-factor antibodies derived from human donor blood plasma
- Antibodies bind Rh+ fetus blood cells and prevent mother from developing antibodies to Rh-factor
- Decreased death of 1 in 2,200 babies down to 1 in 22,000
Self vs. non-self immunity

When antibodies recognize self:

– BAD: autoimmune diseases
– GOOD: anticancer antibodies
Thursday’s guest lecture:
Dr. David Alexander

• How good antibodies are found/identified:
  – Mice
  – Humans
  – Phage/yeast display

• How recombinant antibodies are made

• Recombinant antibody fragments

• Testing antibodies
Scott Dylla, StemCentRx
"Discovery & Development of an Antibody-Drug Conjugate that Effectively Targets Triple-Negative Breast and Ovarian Tumor-Initiating Cells to Result in Sustained Tumor Regressions"

Thursday, May 21, 2015, 12:00 PM to 1:00 PM
Biomed 200
Hosted by Camilla Forsberg
https://www.soe.ucsc.edu/events/event/3841