Antibody Molecules: Structure and Function

Lecture Objectives

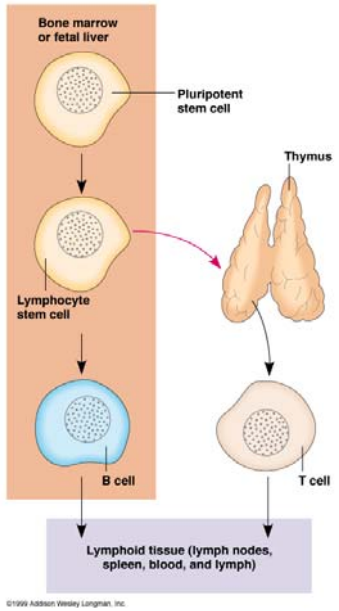
- What Antibodies are
- The structure and function of Abs
- Mechanism of Function of Abs in immunity

What are Antibodies?

Antibodies and their Characteristics

- Proteins that recognize and bind to a particular antigen with very high *specificity*.
- Made in response to exposure to the antigen.
- One virus or microbe may have several *antigenic determinant sites*, to which different antibodies may bind.
- Each antibody has at least two identical sites that bind antigen: *Antigen binding sites*.
- Valence of an antibody: Number of antigen binding sites. Most are bivalent.
- Belong to a group of serum proteins called immunoglobulins (Igs).

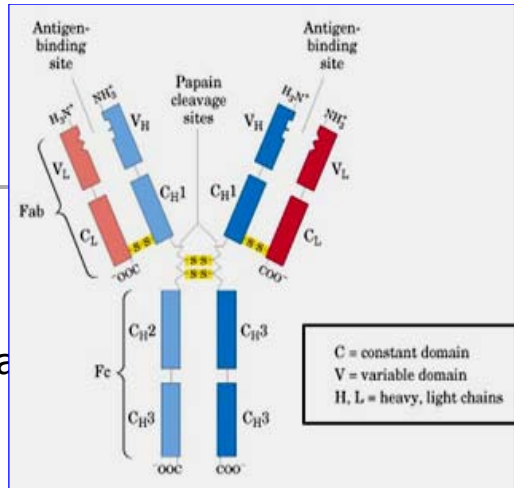
Antibodies are Produced by B Lymphocytes



STRUCTURE AND FUNCTION OF Abs

Antibodies

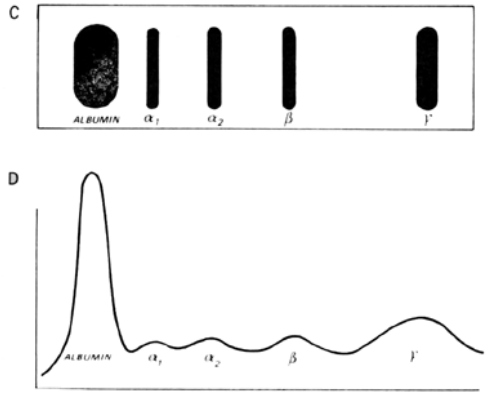
- = Immunoglobulins or γ globulins
- Structure
  - 12 domains (~110 aa
  - Heavy chains & light chains
  - F*ab* region
  - Variable region
  - F*c* region
  - Constant region



Y shaped glycoproteins

γ Globulin

- Name based on electrophoretic activity
- Abs constitute 20% of plasma proteins



Antibody Structure

- Product of B cell activation (plasma cell)
  - Made up of four polypeptides
  - Connected by disulfide bonds
  - Antigen binding fragment (Fabs)
  - Crystallizable fragment (Fc)
  - Fc and Fab joined by a hinge region
  - Various Classes based on Fc

Fab

- Variable (N-terminal of the heavy and light chains)
- Binds to the antigenic determinant
- Swiveling enables more efficiency
- Held together by disulfide bonds

Fc

- Constant (C-terminal of heavy chain)
- Binds to macrophages
- Anchors Ig to lymphocyte
- Also held together by disulfide bonds
- Responsible for class identification

Immunoglobulin Classes

- Isotypes – based on the Fc fragment or heavy chain
- 5 Immunoglobulin (Ig) classes
  - IgG (γ chain, primary, memory, most abundant)
  - IgM (μ chain, pentavalent, complement fix)
  - IgA (α chain, local immunity, secretory IgA)
  - IgD (δ chain, receptor for Ag on B-cells)
  - IgE (ε chain, allergic reactions, react with basophils)

Immunoglobulin Classes

IgG

- Structure: Monomer
- Percentage serum antibodies: 80%
- Location: Blood, lymph, intestine
- Half-life in serum: 23 days
- Complement Fixation: Yes
- Placental Transfer: Yes
- Known Functions: Enhances phagocytosis, neutralizes toxins and viruses, protects fetus and newborn.

Immunoglobulin Classes

IgM

- Structure: Pentamer-equivalent to 10 binding sites
- Percentage serum antibodies: 5-10%
- Location: Blood, lymph, B cell surface (monomer)
- Half-life in serum: 5 days
- Complement Fixation: Yes
- Placental Transfer: No
- Known Functions: First antibodies produced during an infection. Avid binding- ∧ binding sites. Effective against microbes and agglutinating antigens.

Immunoglobulin Classes

IgA

- Structure: Dimer
- Percentage serum antibodies: 10-15%
- Location: Secretions (tears, saliva, intestine, milk), lung, genitourinary tract, blood and lymph.
- Half-life in serum: 6 days
- Complement Fixation: No
- Placental Transfer: No
- Known Functions: Localized protection of *mucosal* surfaces. Provides immunity to infant digestive tract.

Immunoglobulin Classes

IgD

- Structure: Monomer
- Percentage serum antibodies: 0.2%
- Location: B-cell surface, blood, and lymph
- Half-life in serum: 3 days
- Complement Fixation: No
- Placental Transfer: No
- Known Functions: In serum function is unknown. On B cell surface, initiate immune response.

Immunoglobulin Classes

IgE

- Structure: Monomer
- Percentage serum antibodies: 0.002%
- Location: Bound to mast cells and basophils throughout body. Blood.
- Half-life in serum: 2 days
- Complement Fixation: No
- Placental Transfer: No
- Known Functions: Allergic reactions. Possibly lysis of worms.

Immunoglobulin Sub-classes

- IgG
  - a) IgG1 - Gamma 1 (γ1) heavy chains
  - b) IgG2 - Gamma 2 (γ2) heavy chains
  - c) IgG3 - Gamma 3 (γ3) heavy chains
  - d) IgG4 - Gamma 4 (γ4) heavy chains
- IgA
  - a) IgA1 - Alpha 1 (α1) heavy chains
  - b) IgA2 - Alpha 2 (α2) heavy chains

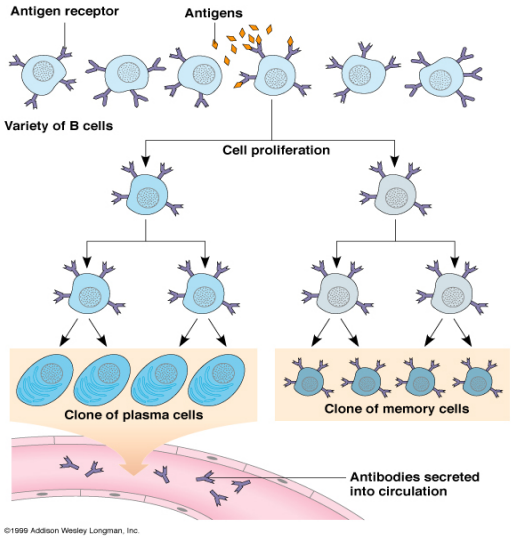
Class switching

- **Class switching** the process by which B cell undergoes V-C gene rearrangement to express a new heavy chain isotype without altering the specificity (V-region) of the antibody produced.

How Do B Cells Produce Antibodies

- B cells develop from stem cells in the bone marrow of adults (liver of fetuses).
- After maturation B cells migrate to lymphoid organs (lymph node or spleen).
- Clonal Selection: When a B cell encounters an antigen it recognizes, it is stimulated and divides into many clones called plasma cells, which actively secrete antibodies.
- Each B cell produces antibodies that will recognize only one antigenic determinant.

Clonal Selection of B Cells is Caused by Antigenic Stimulation



Role of Apoptosis

- Programmed cell death ("Falling away").
- B cells that do not encounter stimulating antigen will self-destruct and send signals to phagocytes to dispose of their remains.

Clonal Selection B cells

- Clonal Selection: B cells (and T cells) that encounter stimulating antigen will proliferate into a large group of cells.
- Why don't we produce antibodies against our own antigens? We have developed *tolerance* to them.
- Clonal Deletion: B and T cells that react against *self* antigens appear to be destroyed during fetal development. Process is poorly understood.

Mechanisms of function of Abs

Definitions

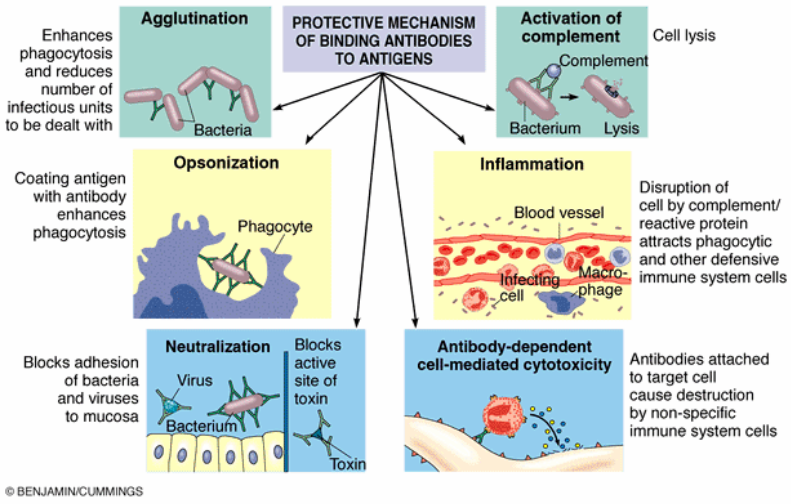
Antigen-Antibody Complex: Formed when an antibody binds to an antigen it recognizes.

Affinity: A measure of binding strength.

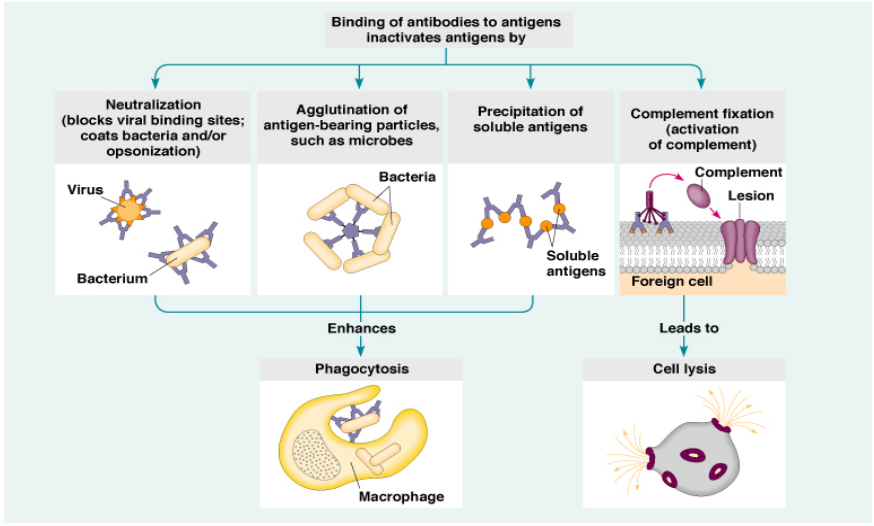
1. **Agglutination:** Antibodies cause antigens (microbes) to clump together.
   - IgM (decavalent) is more effective that IgG (bivalent).
   - Haemagglutination: Agglutination of red blood cells. Used to determine ABO blood types and to detect influenza and measles viruses.
2. **Opsonization:** Antigen (microbe) is covered with antibodies that enhances its ingestion and lysis by phagocytic cells.

3. **Neutralization:** IgG inactivates viruses by binding to their surface and neutralize toxins by blocking their active sites.
4. **Antibody-dependent cell-mediated cytotoxicity (ADCC):** Used to destroy large organisms (e.g.: worms). Target organism is coated with antibodies and bombarded with chemicals from innate immune cells.
5. **Complement Activation:** Both IgG and IgM trigger the complement system which results in cell lysis and inflammation.

Mechanism of Ab Function



Consequences of Ab Binding



Immunological Memory

Pattern of Ab Levels During Infection

Primary Response:

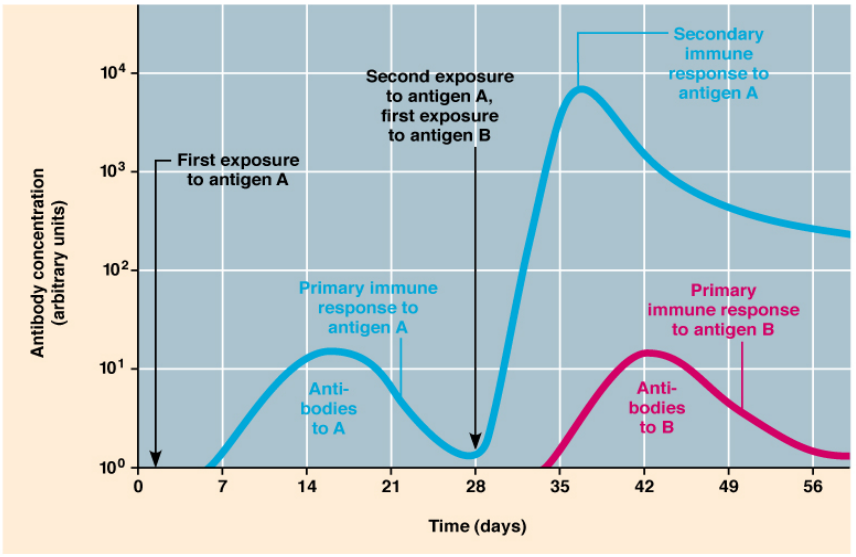
- After *initial* exposure to antigen, no antibodies are found in serum for several days.
- A gradual increase in titer, first of IgM and then of IgG is observed.
- Most B cells become plasma cells, but some B cells become long living *memory cells*.
- Gradual decline of antibodies follows.

Immunological Memory (Continued)

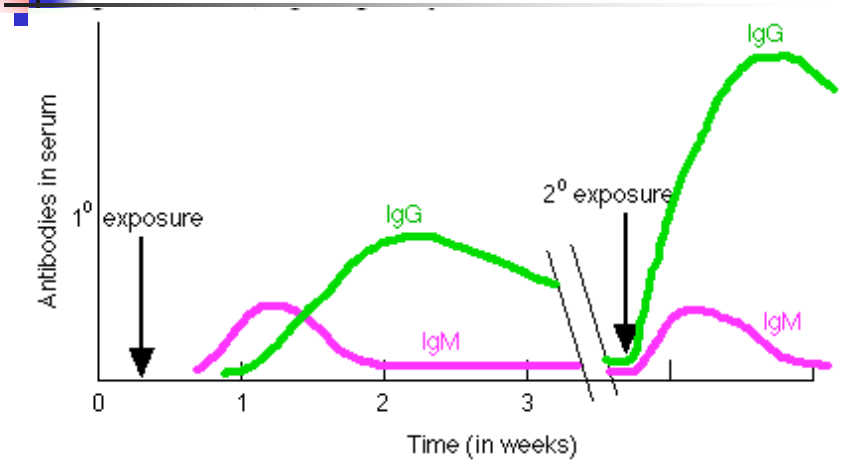
Secondary Response (Anamnestic response):

- Subsequent exposure to the same antigen displays a faster and more intense antibody response (*shorter lag phase*).
- Increased antibody response is due to the existence of memory cells, which rapidly produce plasma cells upon antigen stimulation.
- Ab Titer (Titre): The amount of antibody in the serum.

Antibody Response After Exposure to Antigen



Fractionated Ab after re-exposure



Relationship Between Cell-Mediated and Humoral Immunity

Antibody Production

T cell - Dependent Antigens:

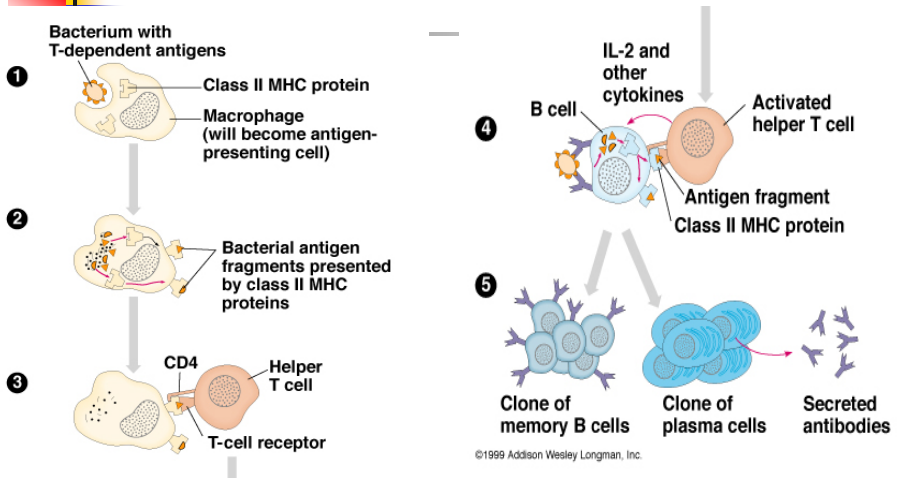
- Ab production requires assistance from T helper cells.
- A macrophage ingests antigen and presents it to T_H cell.
- T_H cell stimulates B cells specific for antigen to become plasma cells.
- Antigens are mainly proteins on viruses, bacteria, foreign red blood cells, and hapten-carrier molecules.

Antibody Production

T-Independent Antigens:

- Antibody production does not require assistance from T cells.
- Antigens are mainly polysaccharides or lipopolysaccharides with repeating subunits (bacterial capsules).
- Weaker immune response than for T-dependent antigens.

Humoral Response to T Dependent Antigens



On the other hand …

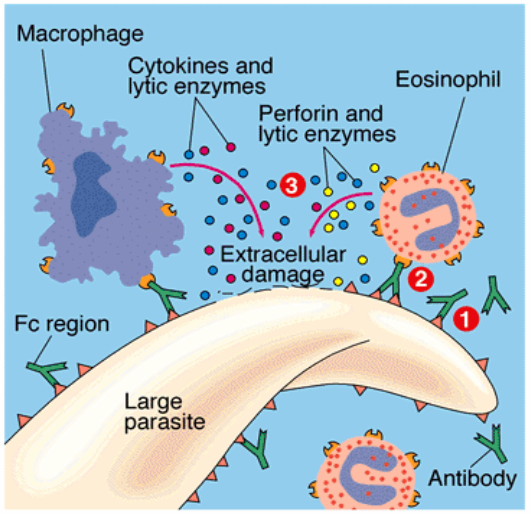
Cell-mediated immunity

- Direct involvement of T cells
- Produce and react to cytokines
- Activated simultaneously with B cell activation
- Subsets of T cells possess unique CD receptors (CD4+, CD8+)

Antibody Dependent Cell Mediated Cytotoxicity (ADCC)

- Target cell is covered with antibodies, leaving Fc portion sticking outwards.
- Natural killer and other innate immune cells that have receptors for Fc region are stimulated to kill targeted cells.
- Target organism is lysed by substances secreted by attacking cells.
- Used to destroy large organisms that cannot be phagocytosed.

Destruction of Large Parasites by ADCC



Overview of Immune Response