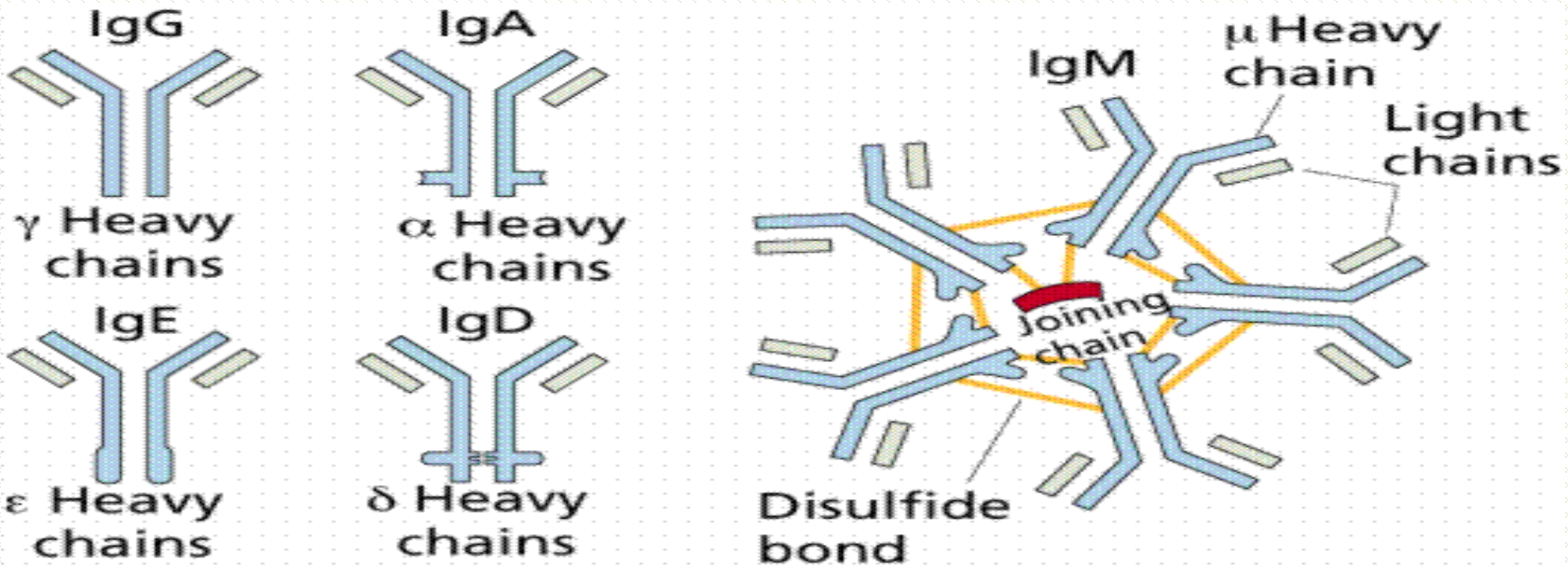
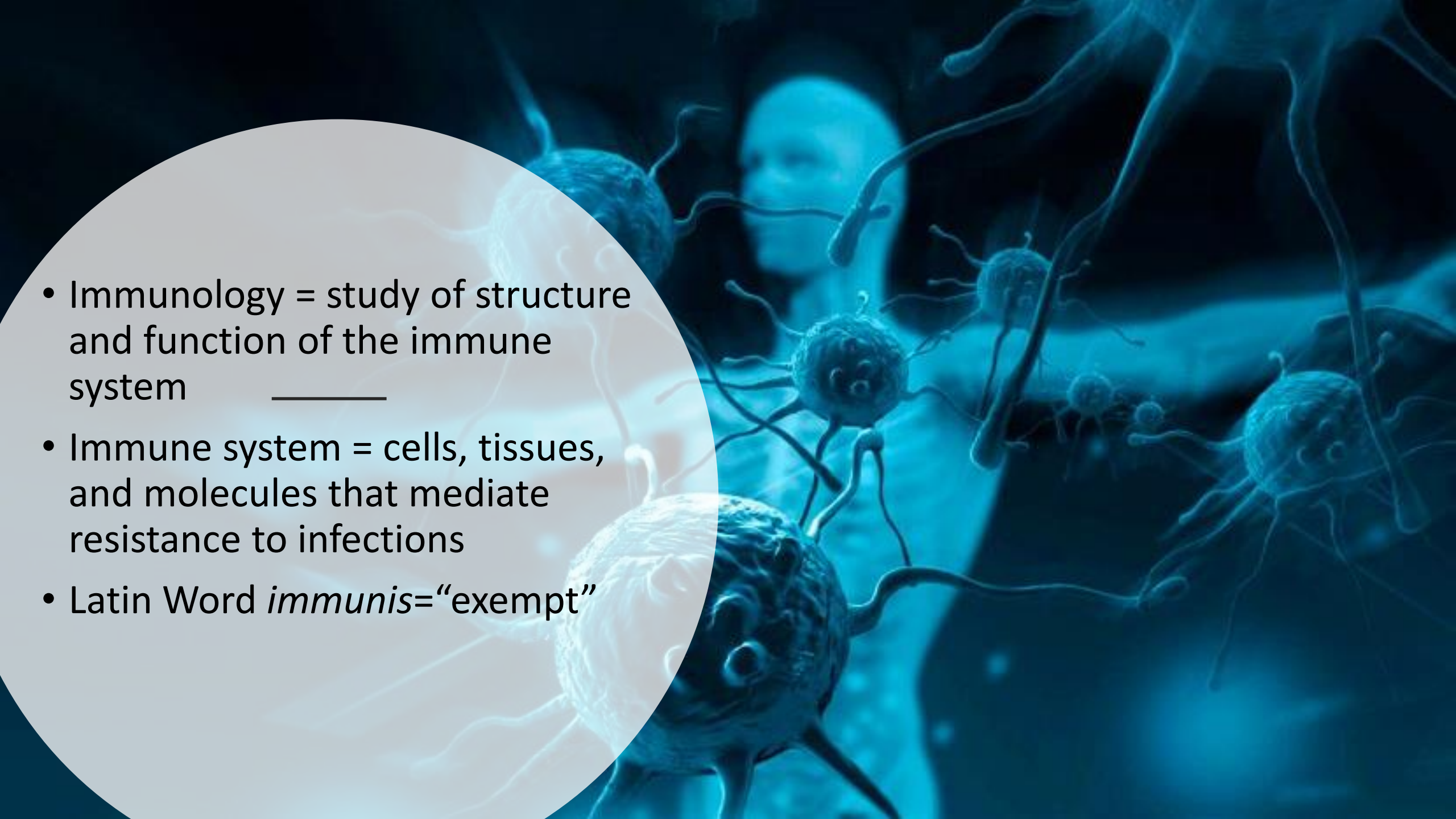
Immunoglobulins



Learning objectives

To understand :

- General Structure of Immunoglobulin
- Types of Immunoglobulins
- Structural variations of different immunoglobulins
- Function of different immunoglobulins
- Clinical significance of Immunoglobulins

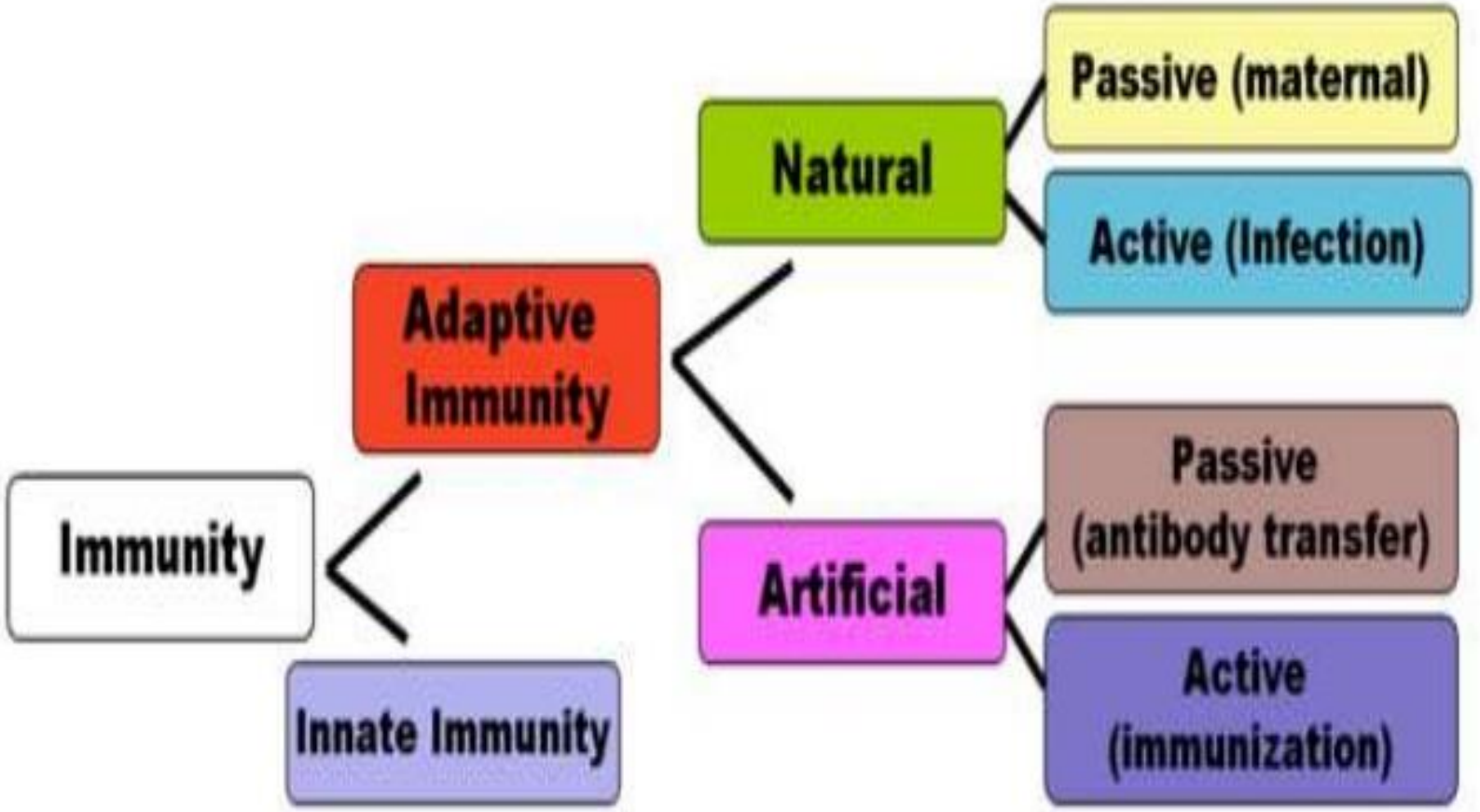


- Immunology = study of structure and function of the immune system _____
- Immune system = cells, tissues, and molecules that mediate resistance to infections
- Latin Word *immunis*=“exempt”

Immunity

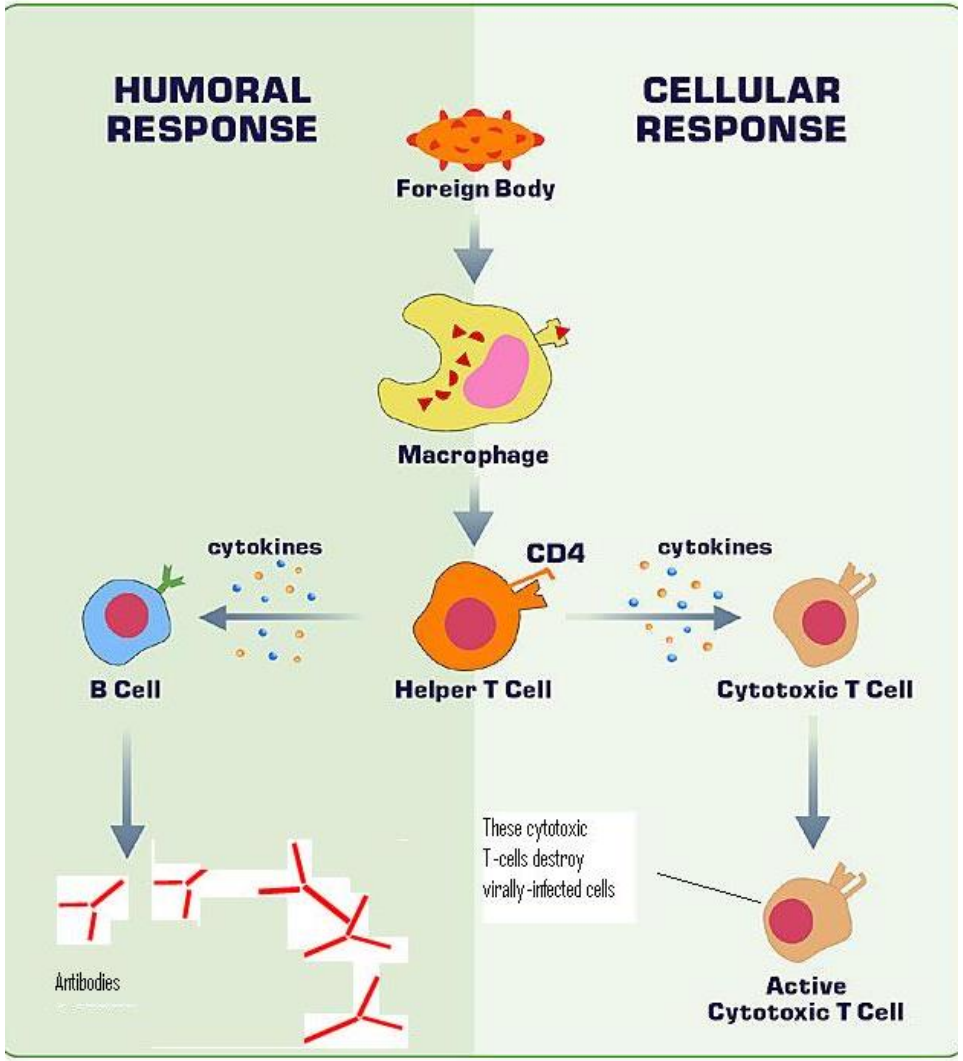
- Immunity = resistance of a host to pathogens and their toxic effects

Types of immunity



Immune response

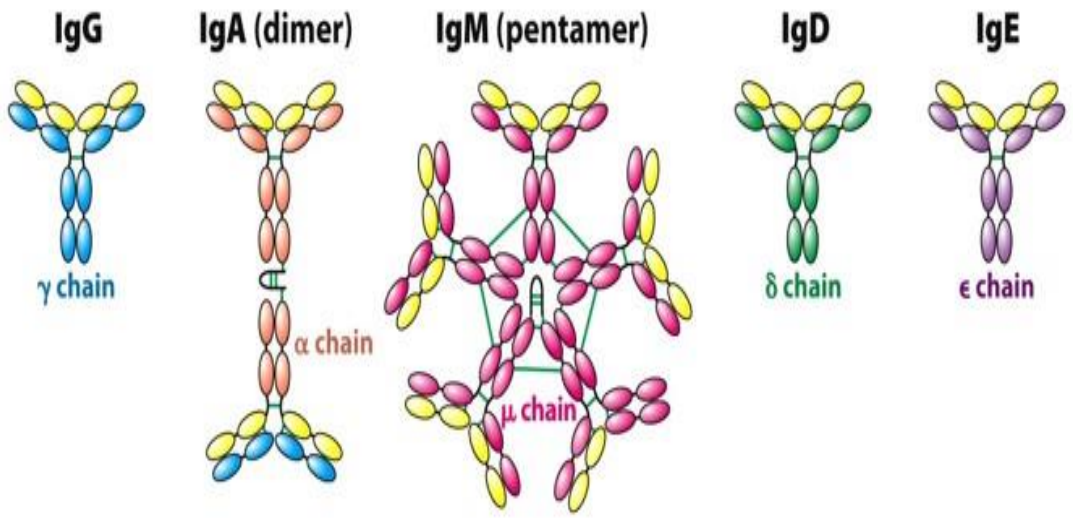
- **Immune response** = collective and coordinated response to the introduction of foreign substances in an individual mediated by the cells and molecules of the immune system



Immunoglobulins

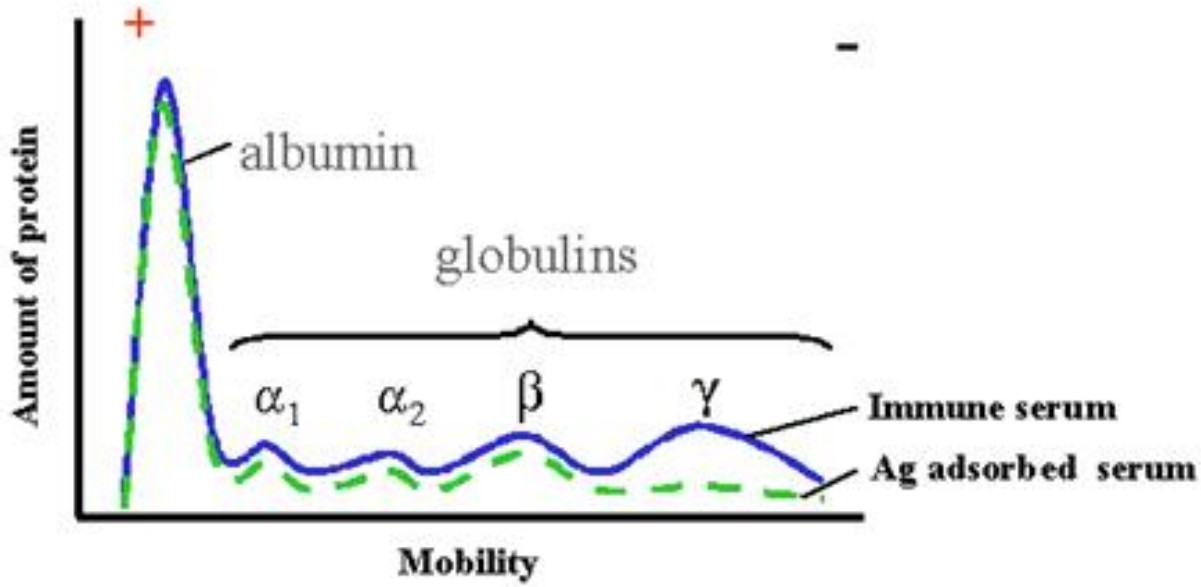
Immunoglobulins are:

- glycoprotein molecules,
- function as antibodies
- produced by plasma cells
- in response to an immunogen.



Immunoglobulins

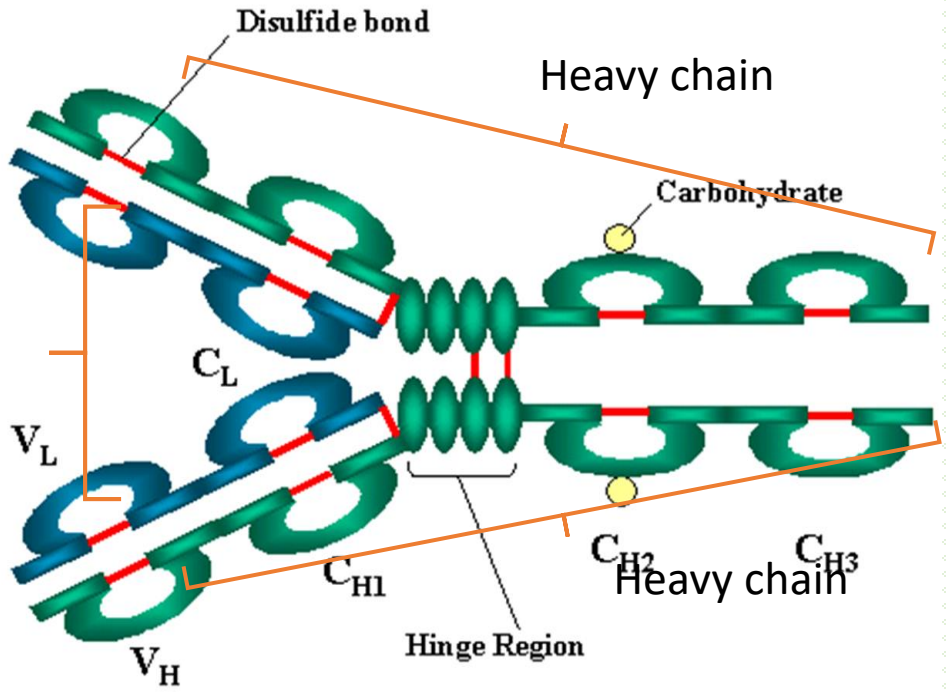
The immunoglobulins derive their name from the finding that they migrate in the region of globulins when antibody-containing serum is placed in an electrical field.



Structural characteristics

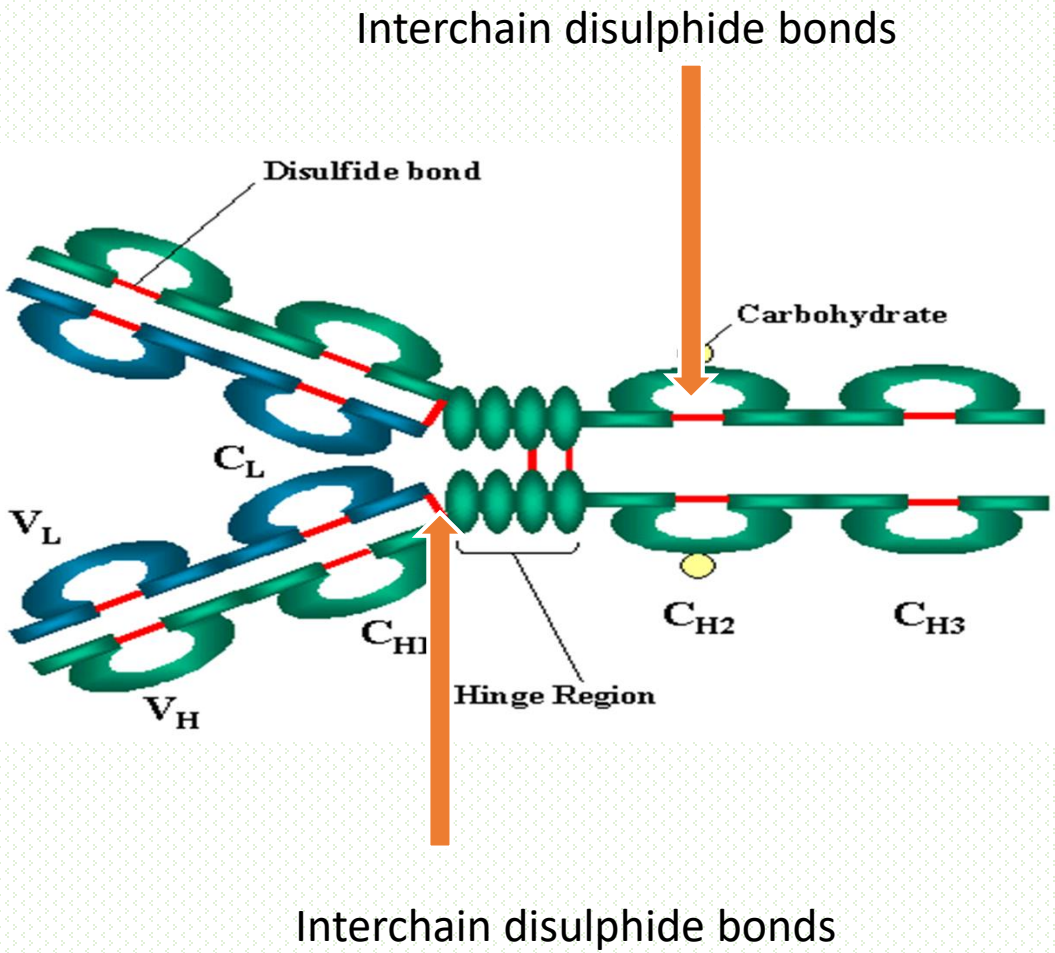
A. Heavy and Light Chains

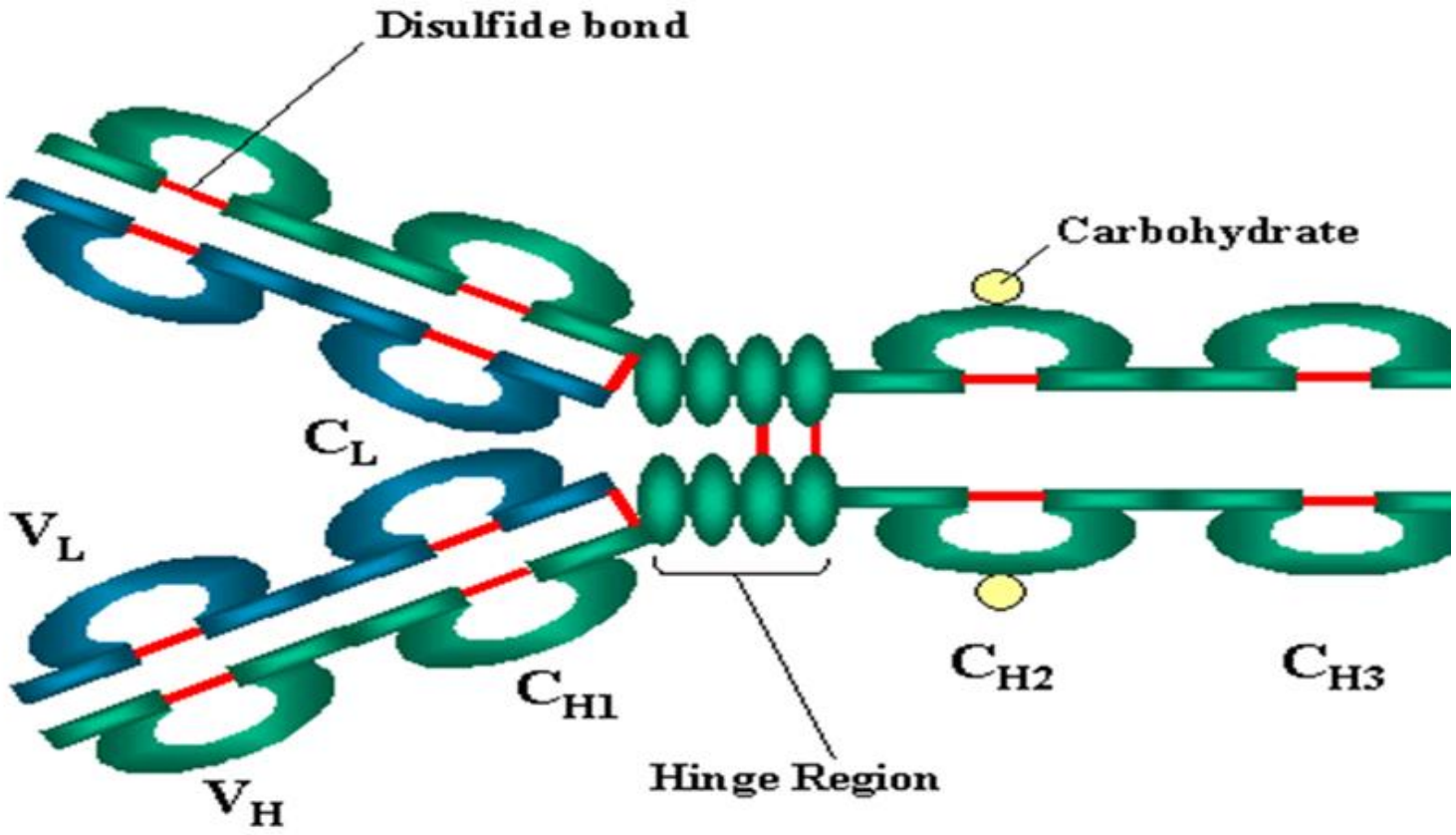
- All immunoglobulins have a four chain structure as their basic unit.
- They are composed of two identical light chains (23kD) and two identical heavy chains (50-70kD)



B. Disulfide bonds

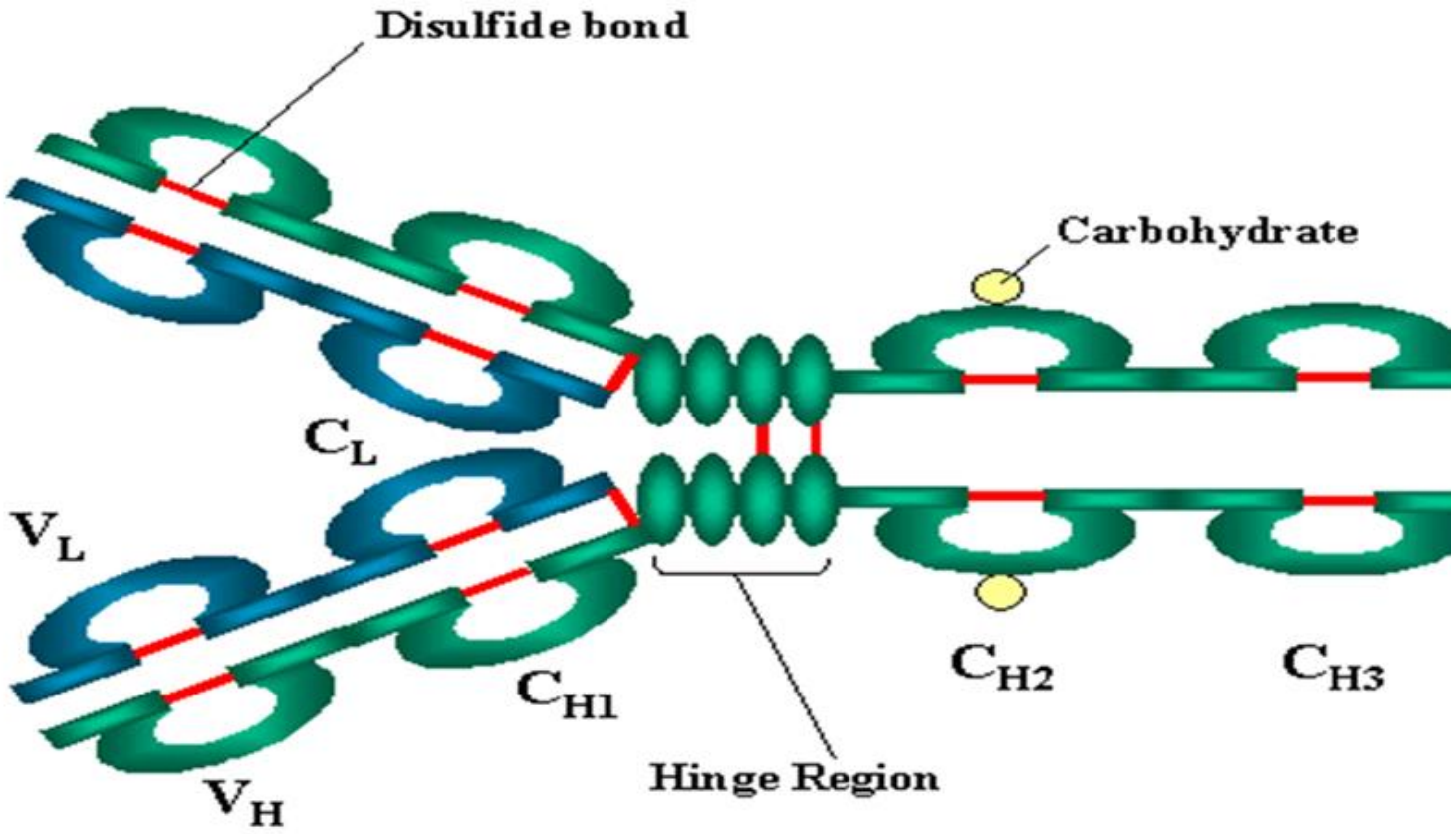
- **Inter-chain disulfide bonds -** The heavy and light chains and the two heavy chains are held together by inter-chain disulfide bonds and by non-covalent interactions.
- The number of inter-chain disulfide bonds varies among different immunoglobulin molecules.
- **Intra-chain disulfide binds -** Within each of the polypeptide chains there are also intra-chain disulfide bonds.





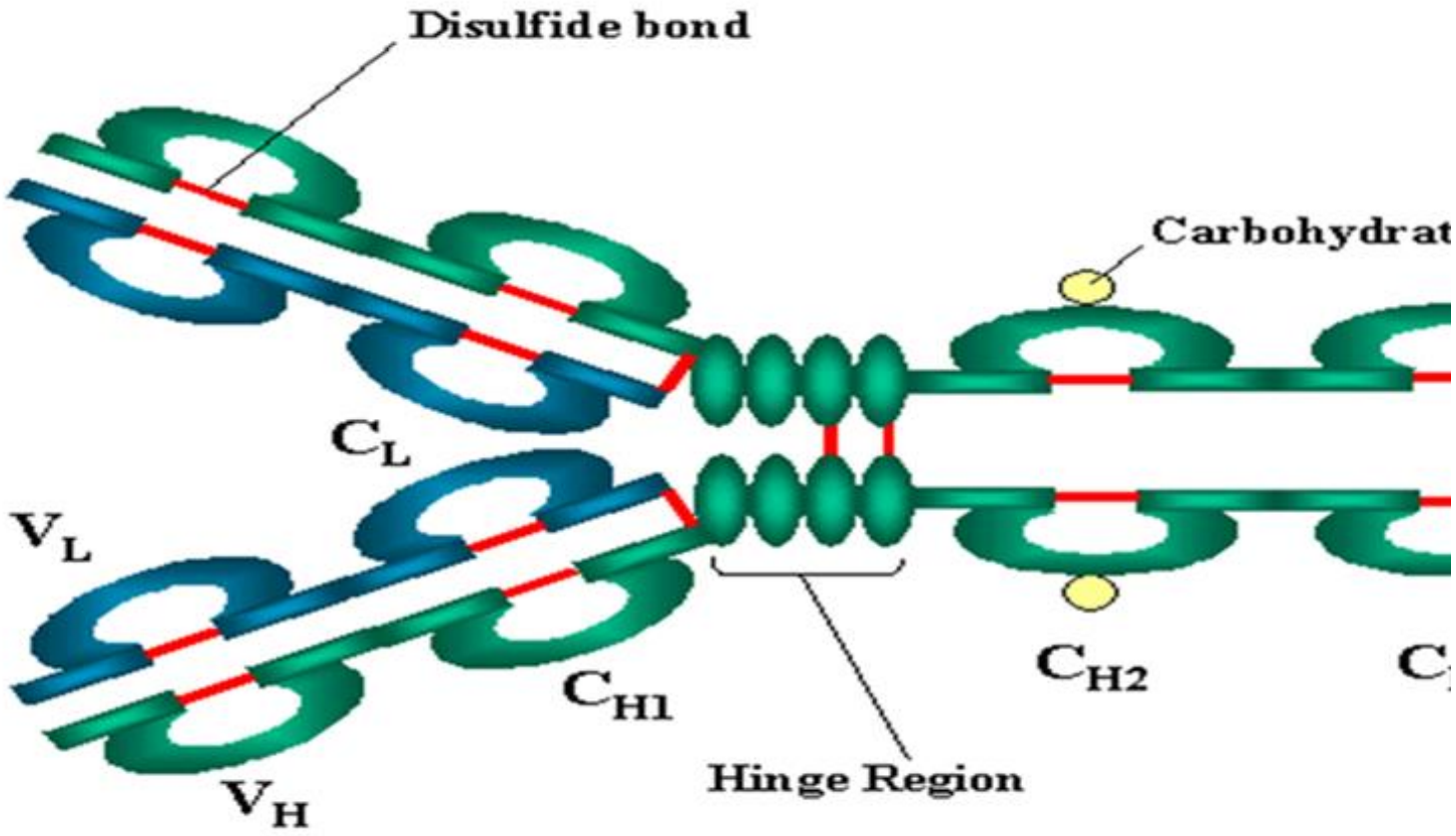
C. Variable (V) and Constant (C) Regions

- Both the heavy and light chain can be divided into two regions based on variability in the amino acid sequences. These are the
- Light Chain - V_L (110 amino acids) and C_L (110 amino acids)
- Heavy Chain - V_H (110 amino acids) and C_H (330-440 amino acids)



D. Hinge Region

- This is the region at which the arms of the antibody molecule form a Y.
- It is called the hinge region because there is some flexibility in the molecule at this point.

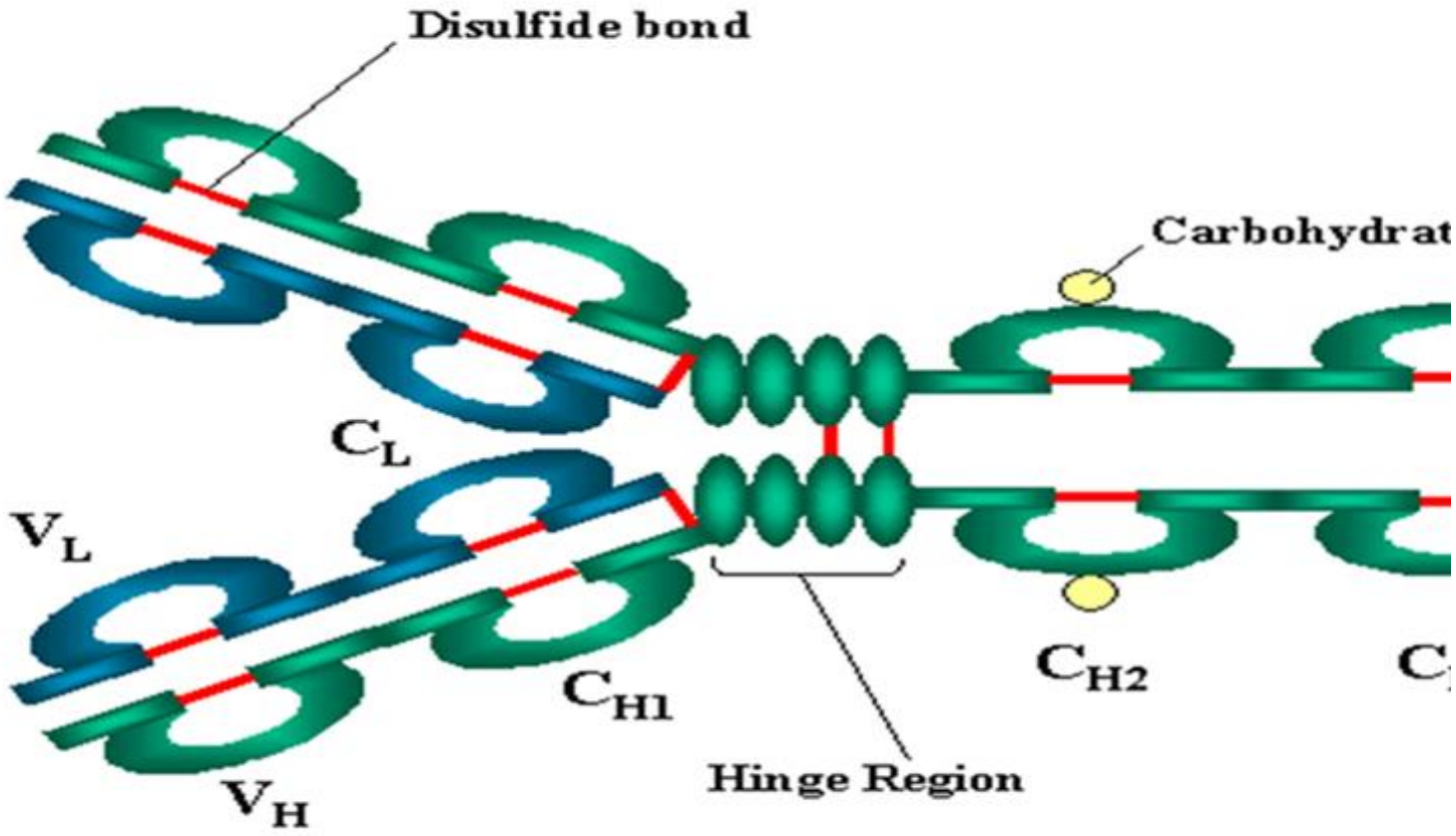


E. Domains

- Three dimensional images of the immunoglobulin molecule show that it is not a straight molecule rather, it is folded into globular regions each of which contains an intra-chain disulfide bond .
- These regions are called **domains**.

1. Light Chain Domains - V_L and C_L

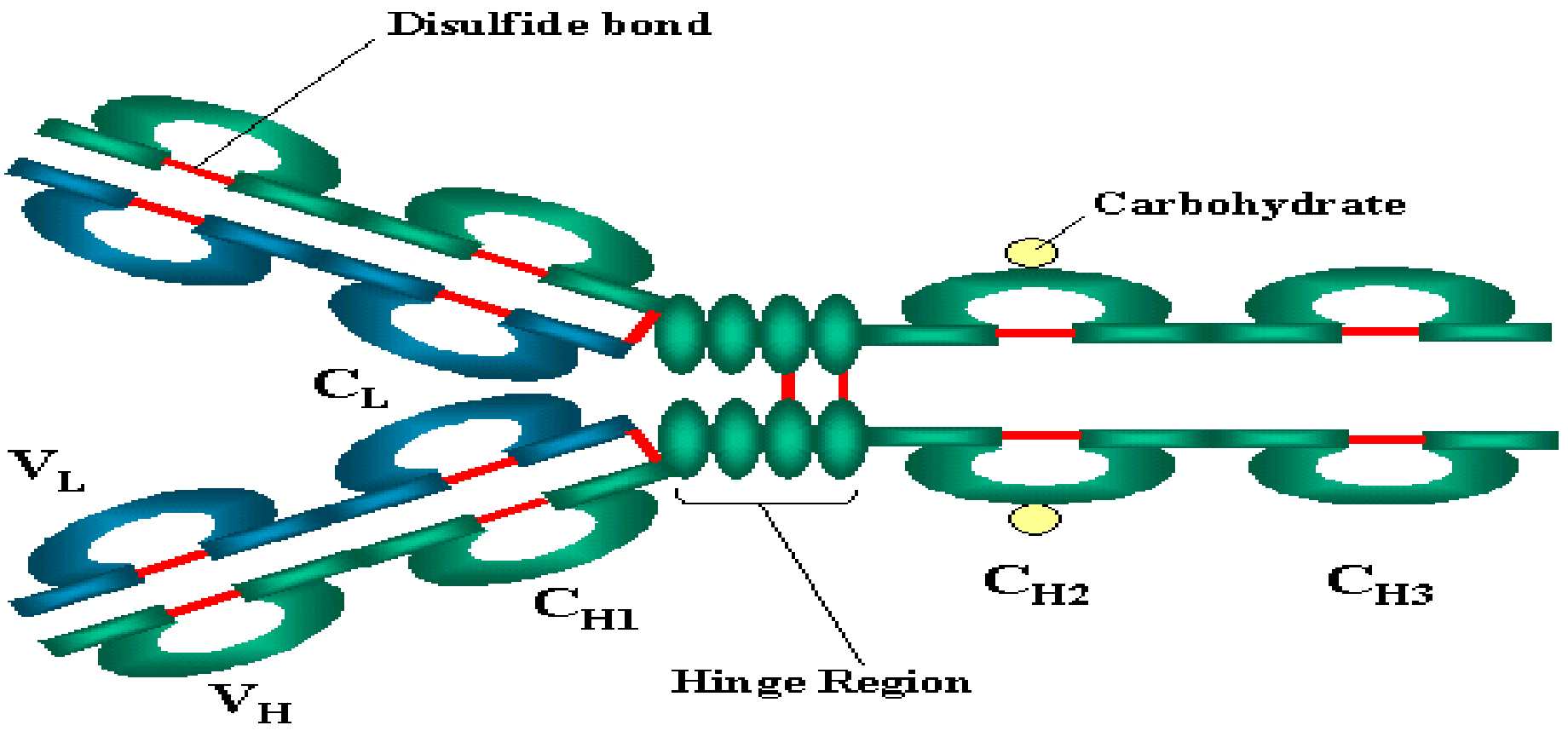
2. Heavy Chain Domains - V_H, C_{H1},CH_2C_{H3} (or C_{H4})



F. Oligosaccharides

- Carbohydrates are attached to the C_{H2} domain in most immunoglobulins.
- However, in some cases carbohydrates may also be attached at other locations.

Basic structure of immunoglobulins



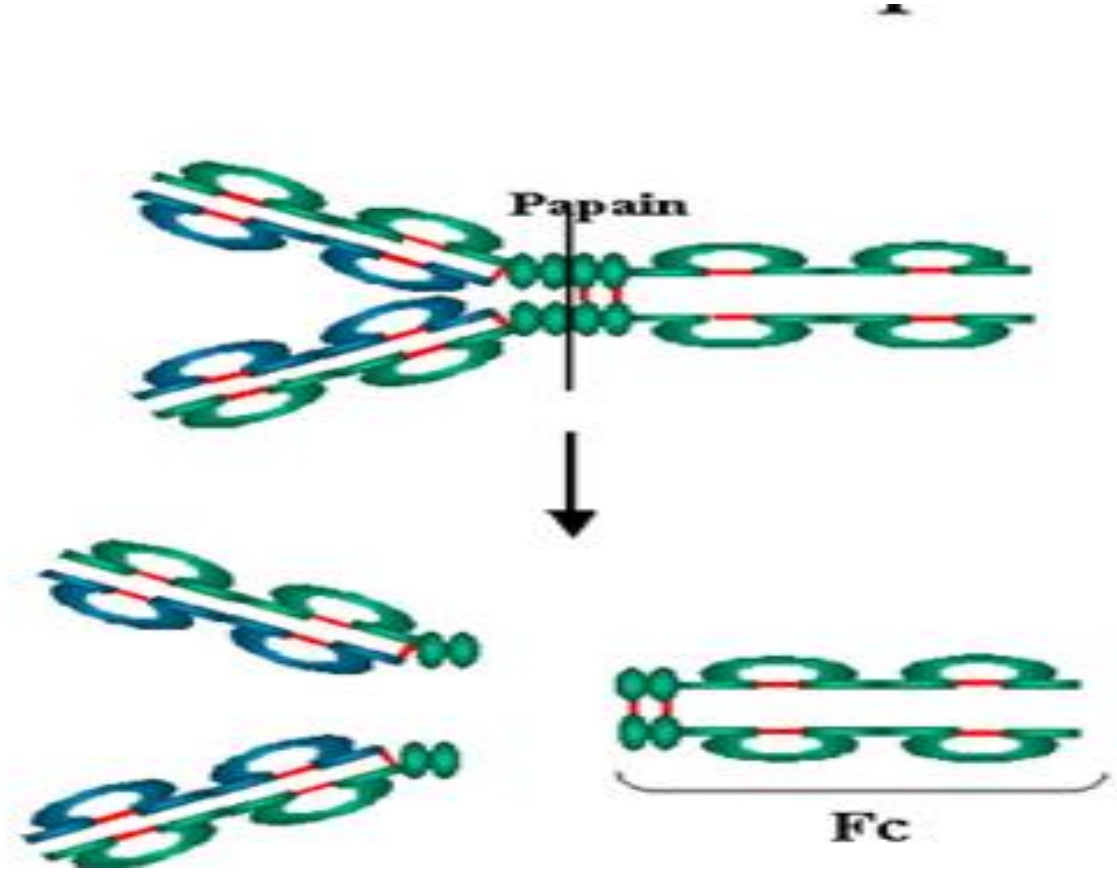
Immunoglobulin fragments: structure/function relationships

Immunoglobulin fragments produced by proteolytic digestion –

A. Fab

Digestion with papain breaks the immunoglobulin molecule in the hinge region before the H-H inter-chain disulfide bond.

This results in the formation of two identical fragments that contain the light chain and the V_H and C_{H1} domains of the heavy chain.

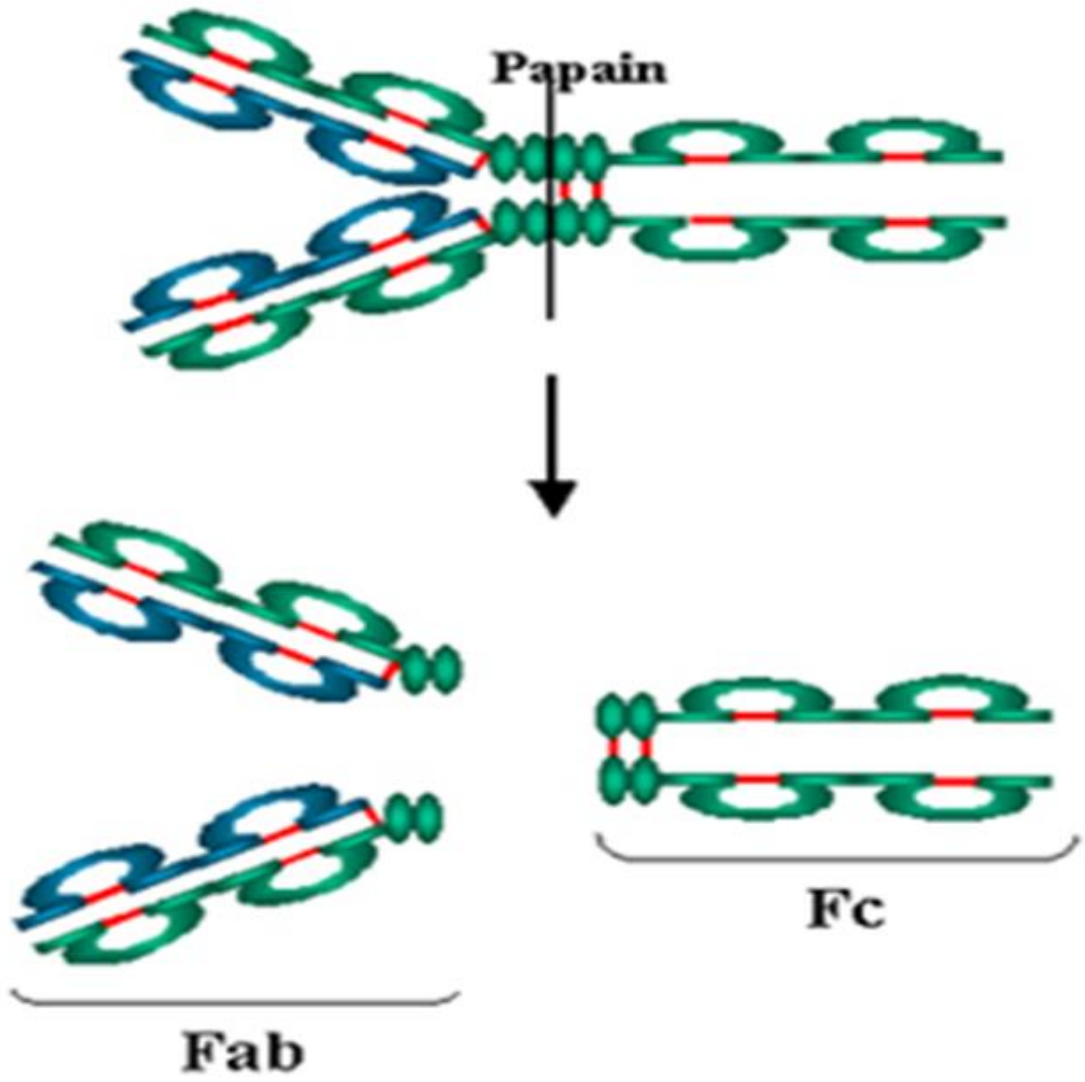


Immunoglobulin fragments by Papain

- **Fab** -These fragments are called the Fab fragments because they contained the **antigen binding** sites of the antibody.
- Each Fab fragment is monovalent whereas the original molecule was divalent.
- The combining site of the antibody is created by both V_H and V_L.

Immunoglobulin fragments by Papain

- **B. Fc**
  Digestion with papain also produces a fragment that contains the remainder of the two heavy chains each containing a C_{H2} and C_{H3} domain.
- This fragment was called Fc because it was easily crystallized.



Structure-function relationship

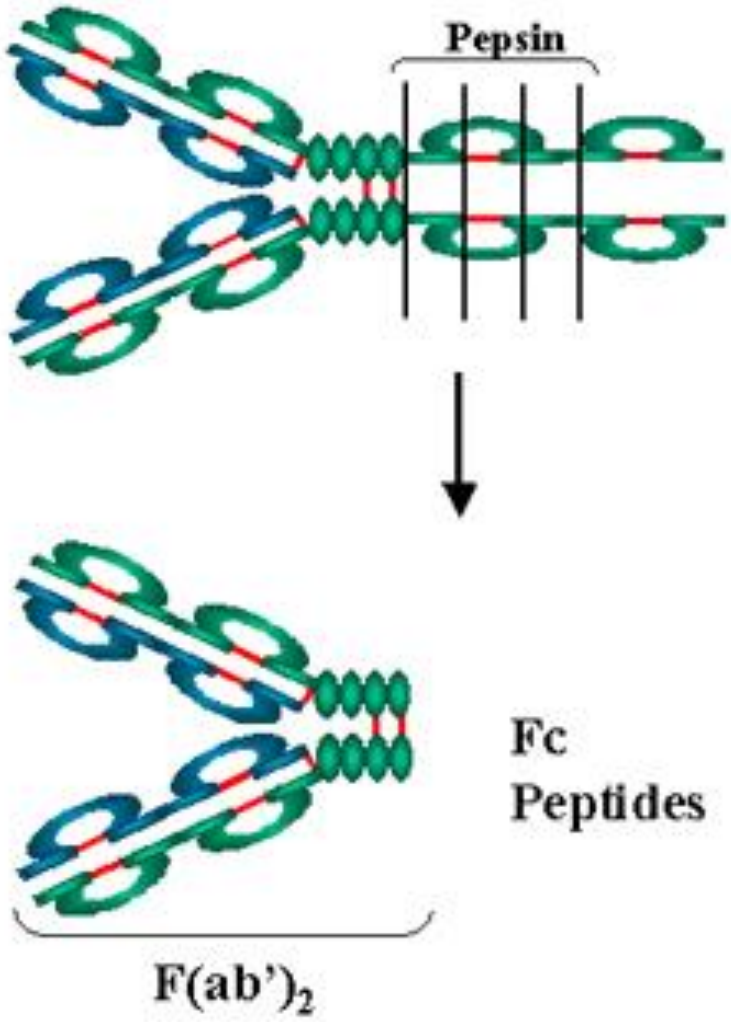
Antigen binding function of Immunoglobulins is carried out by Fab part,

Effector functions -The effector functions are mediated by Fc part of the molecule.

Different functions are mediated by the different domains in this fragment.

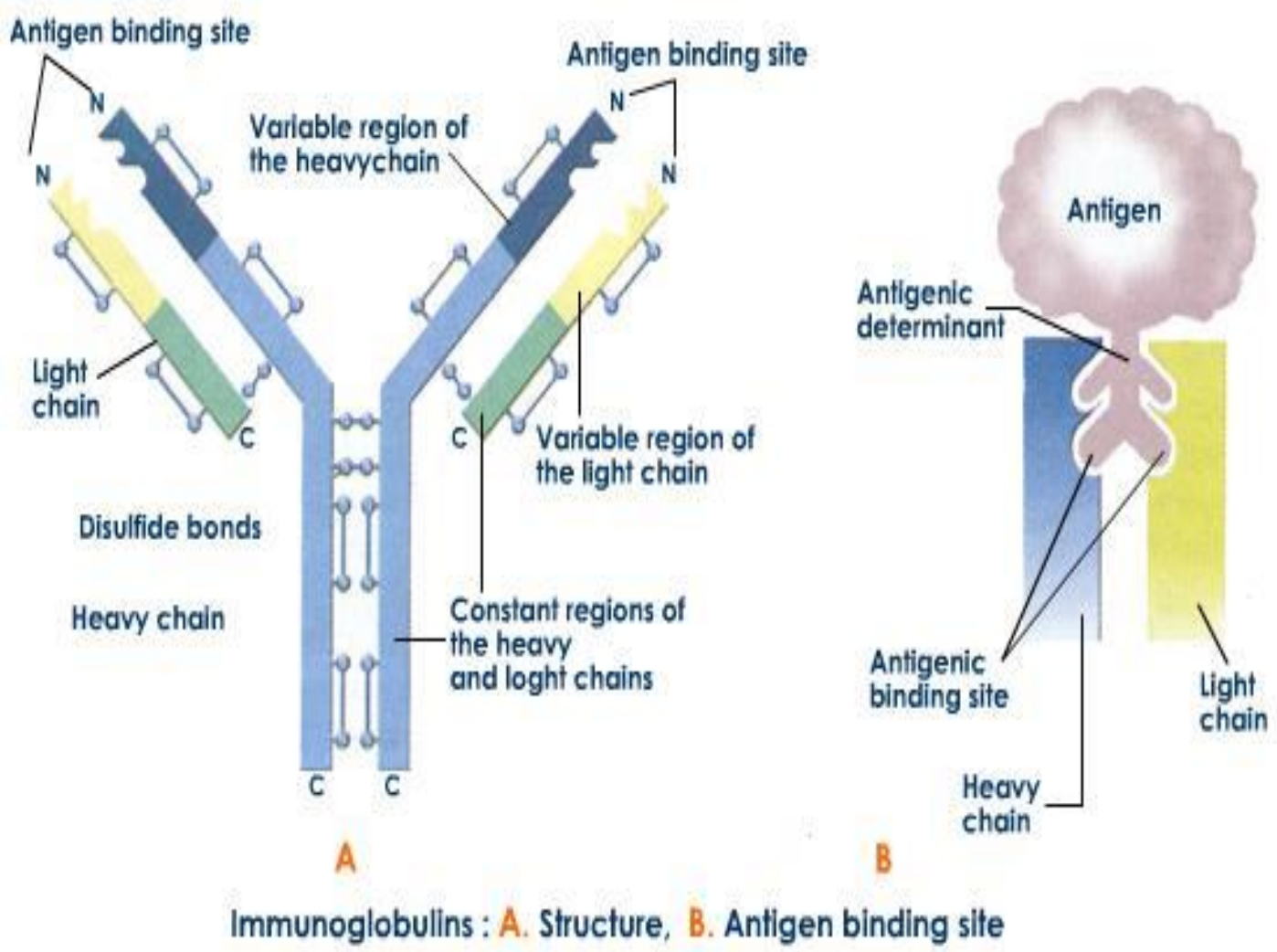
Immunoglobulin fragments by Pepsin

- **F(ab')$_2$ -**Treatment of immunoglobulins with pepsin results in cleavage of the heavy chain after the H-H inter-chain disulfide bonds resulting in a fragment that contains both antigen binding sites . This fragment is called F(ab')$_2$because it is divalent.
- **The Fc region** of the molecule is digested into small peptides by pepsin. The F(ab')$_2$binds antigen but it does not mediate the effector functions of antibodies.



General functions of immunoglobulins

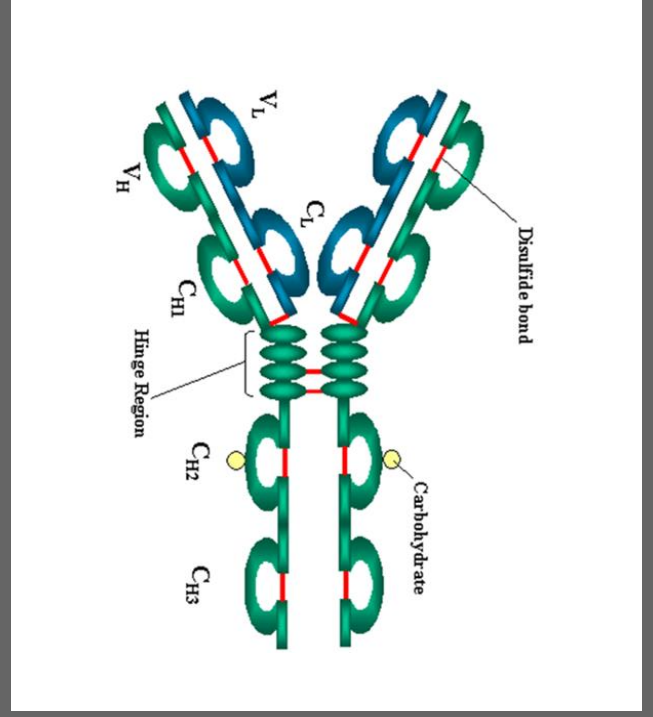
- **A. Antigen binding**
- Antigen binding by antibodies is the primary function of antibodies and can result in protection of the host.
- Each immunoglobulin binds to a specific antigenic determinant.

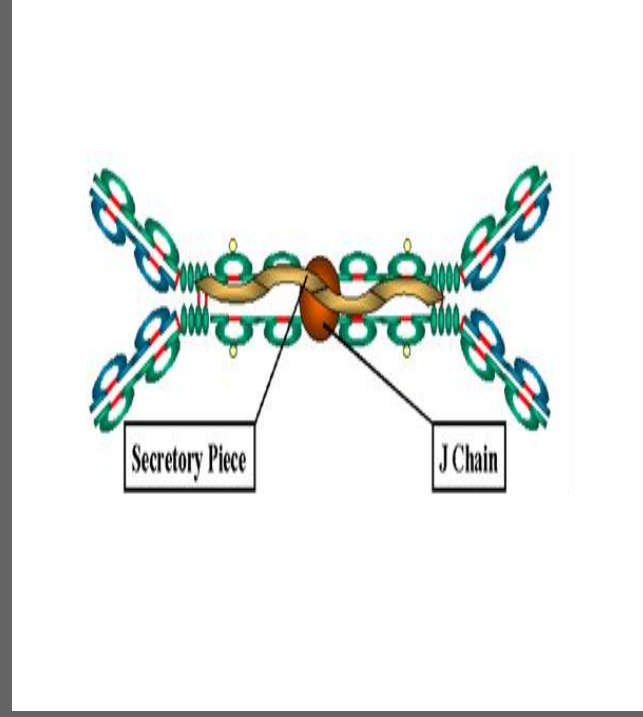


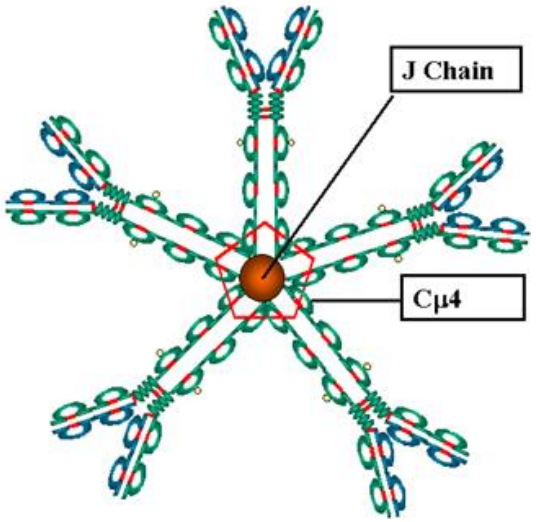
Immunoglobulins : A. Structure, B. Antigen binding site

Valency of antibody

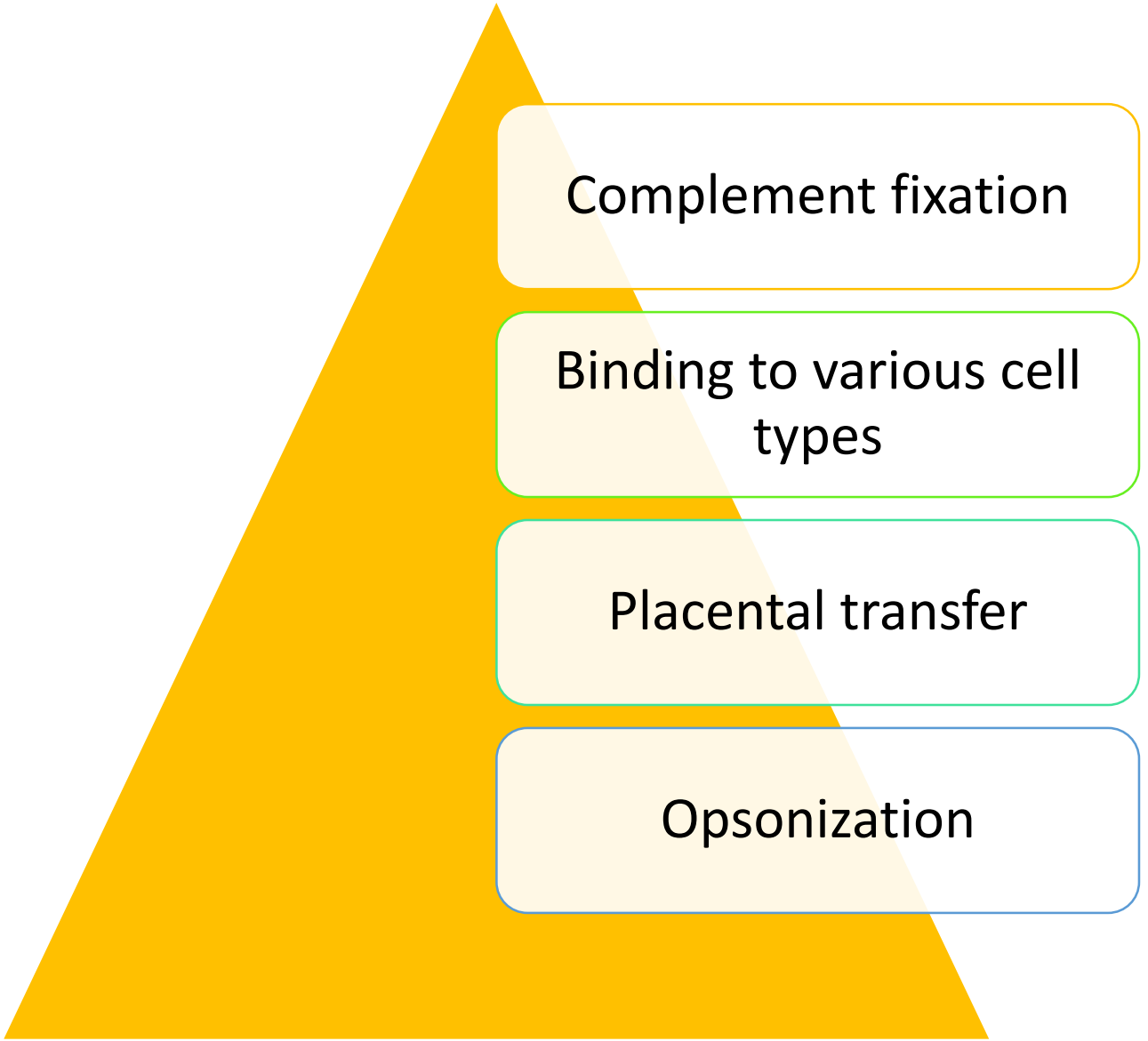
- The valency of antibody refers to the number of antigenic determinants that an individual antibody molecule can bind.
- The valency of all antibodies is at least two and in some instances more.



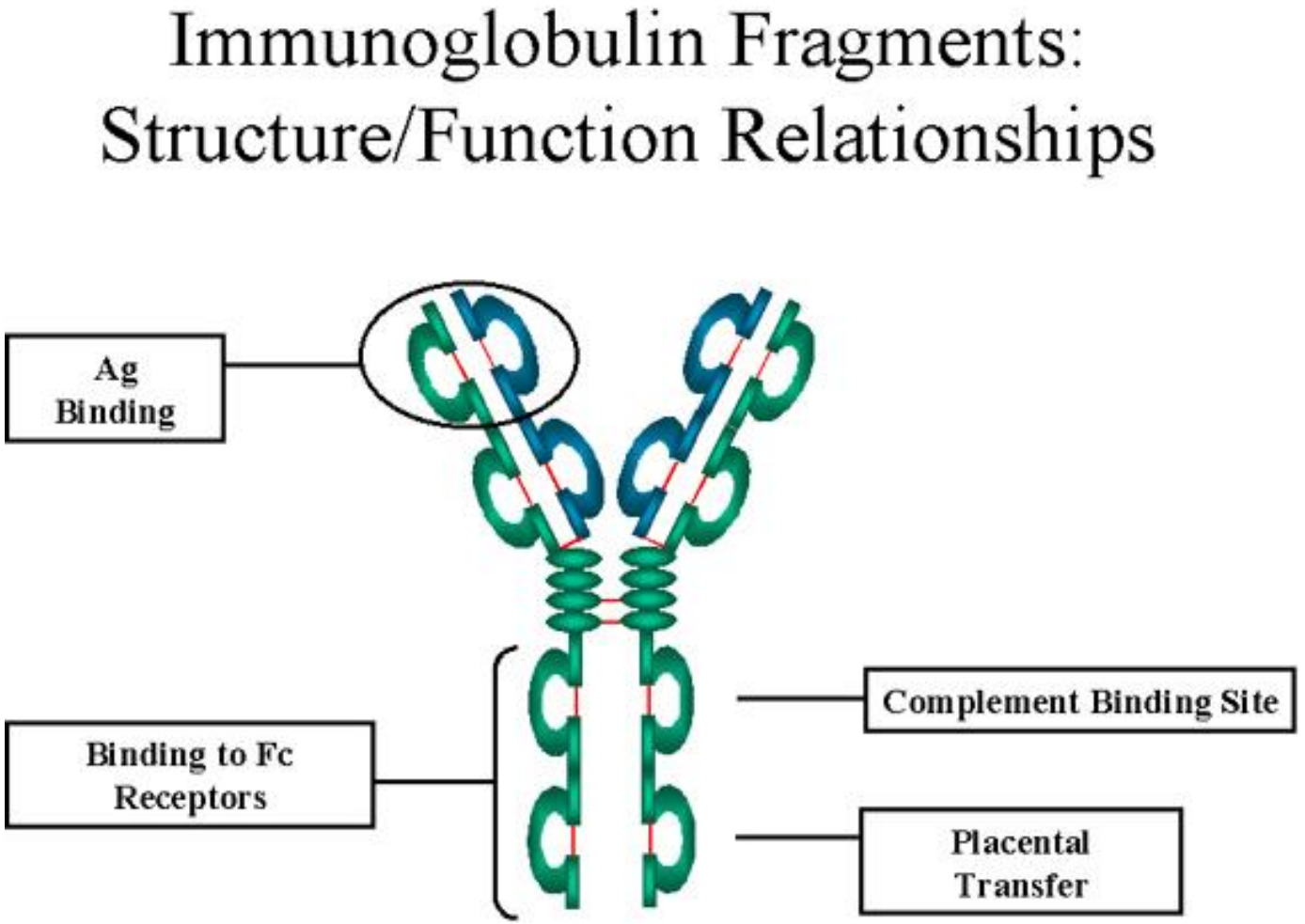




B. Effector Functions

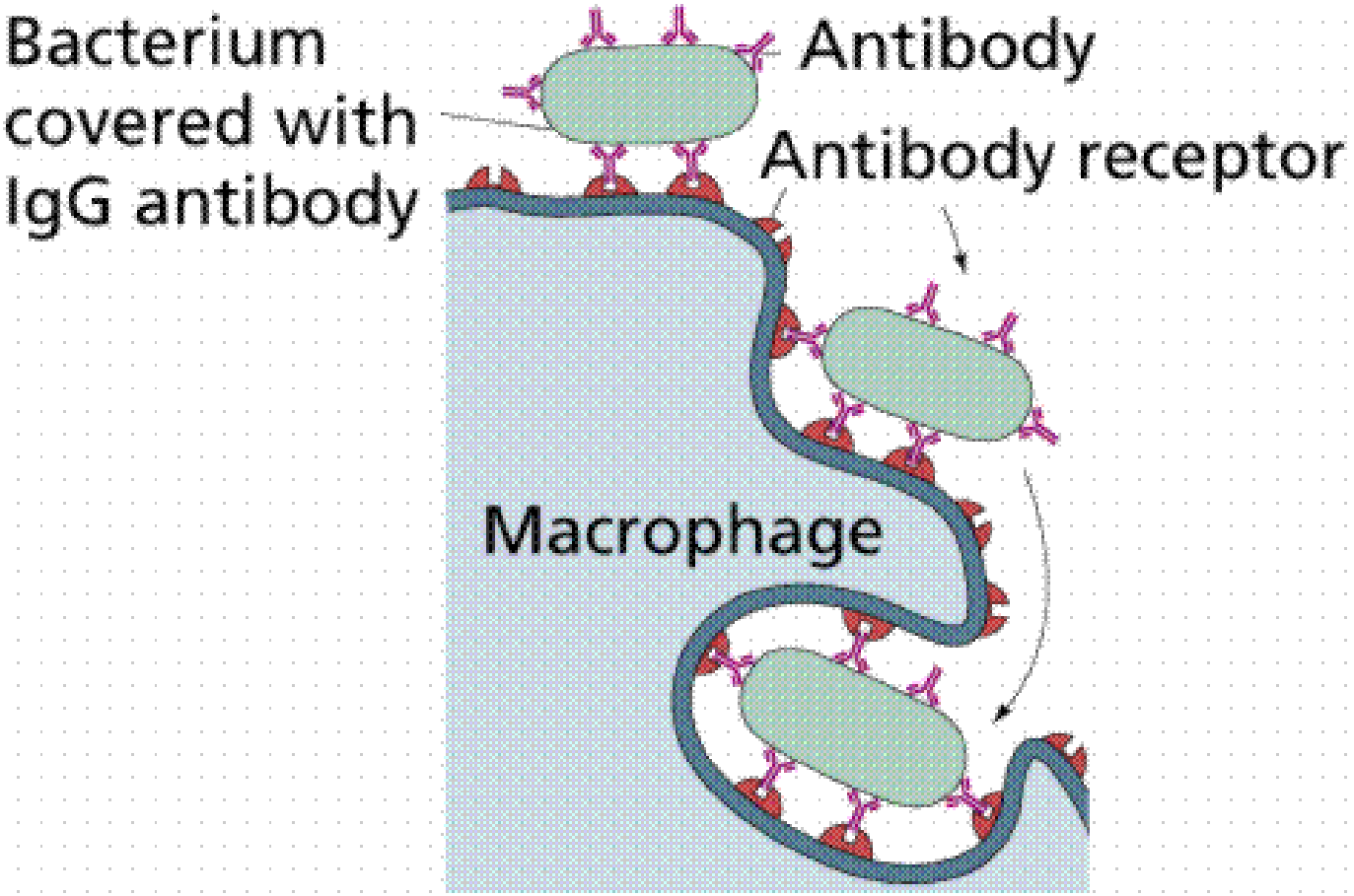


General functions of Immunoglobulins



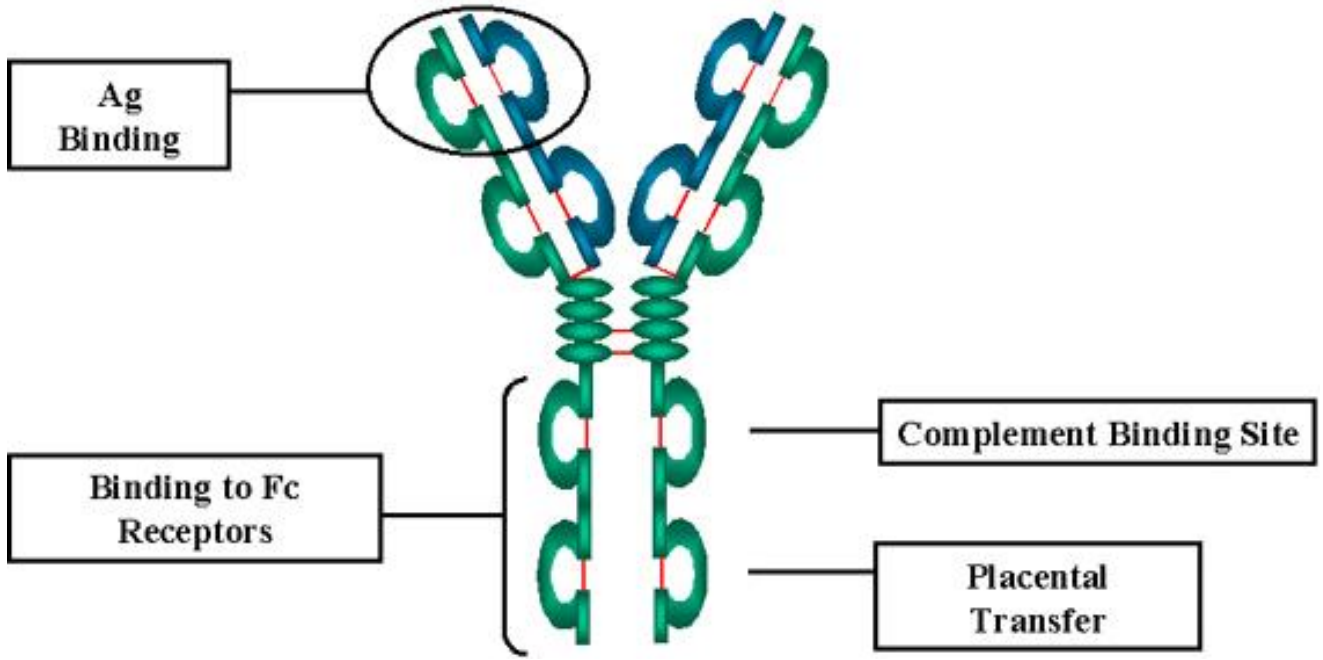
Binding to various cell types

- Phagocytic cells, lymphocytes, platelets, mast cells, and basophils have receptors that bind immunoglobulins.
- This binding can activate the cells to perform some function.



Functions of Immunoglobulins

- Some immunoglobulins also bind to receptors on placental trophoblasts, which results in transfer of the immunoglobulin across the placenta.
- As a result, the transferred maternal antibodies provide immunity to the fetus and newborn.

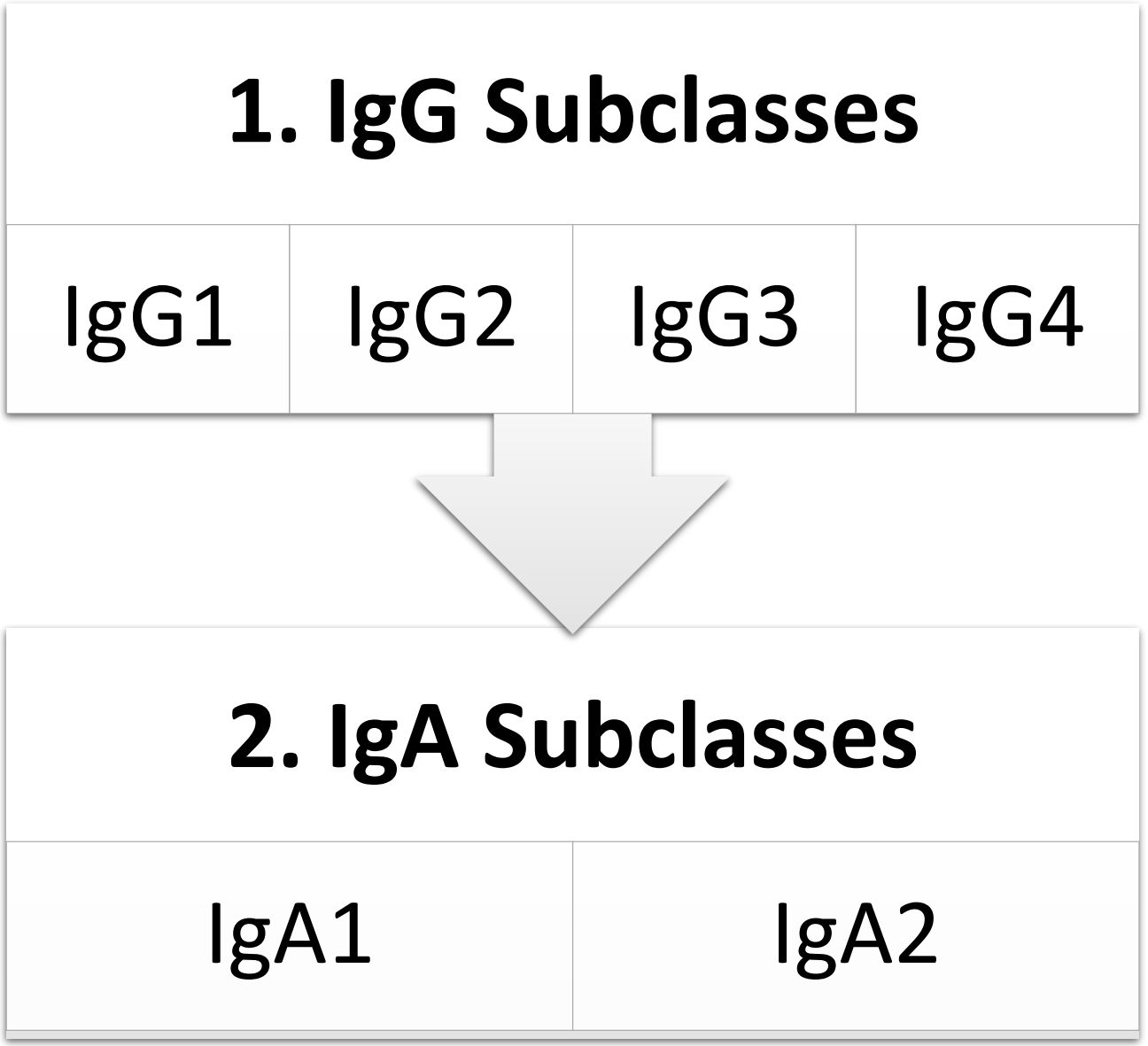


Immunoglobulin classes

- The immunoglobulins can be divided into five different classes, based on differences in the amino acid sequences in the constant region of the heavy chains.

1. IgG - Gamma heavy chains
2. IgM - Mu heavy chains
3. IgA - Alpha heavy chains
4. IgD - Delta heavy chains
5. IgE - Epsilon heavy chains

Immunoglobulin Subclasses



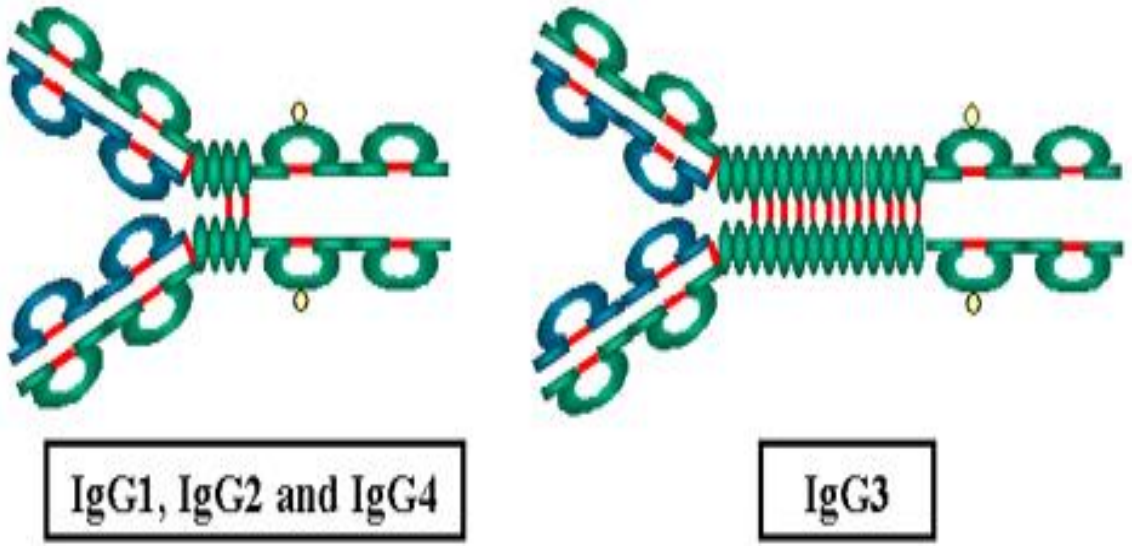
Immunoglobulin Types

- Immunoglobulins can also be classified by the type of light chain that they have.
- Light chain types are based on differences in the amino acid sequence in the constant region of the light chain.

1. Kappa light chains
2. Lambda light chains

Immunoglobulin G (IgG)- Structure

- All IgG's are monomers (7S immunoglobulin).
- The subclasses differ in the number of disulfide bonds and length of the hinge region.

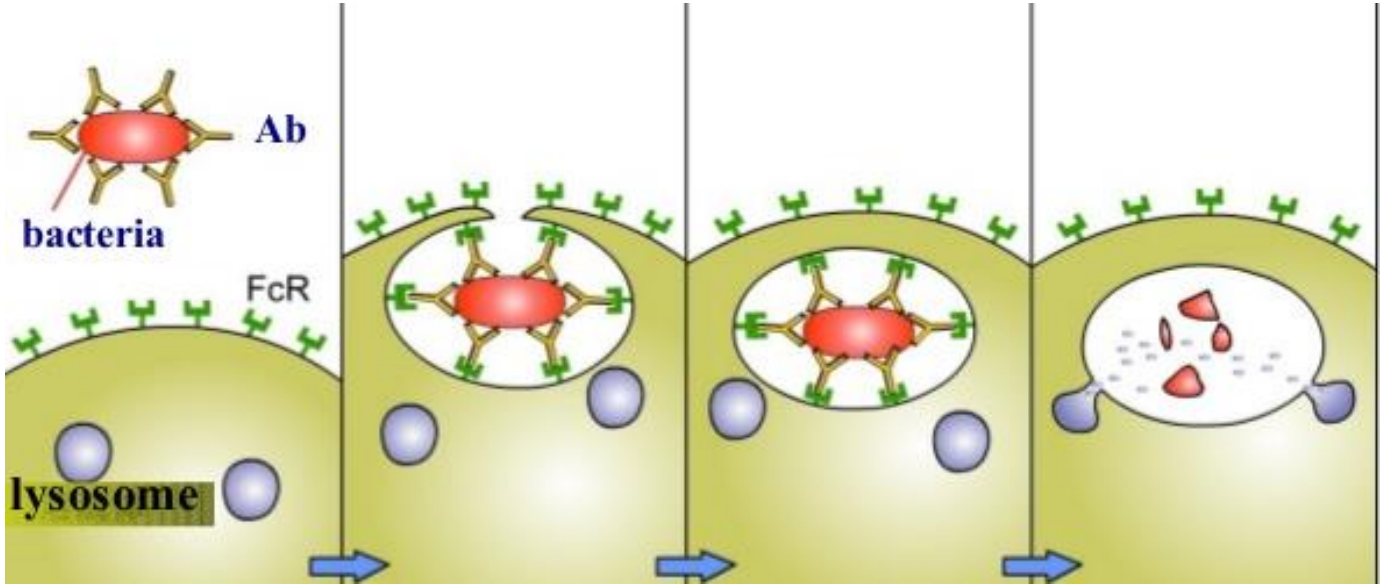


Immunoglobulin G (IgG)-Properties

- Major Ig in serum
- Major Ig in extravascular spaces
- Only Ig that crosses placenta
- Complement fixation
- Opsonization

Opsonization

- The term **opsonin** is used to describe substances that enhance phagocytosis.
- IgG is a good opsonin.
- The antibody prepares the antigen for killing by the phagocytic cells.
- Macrophages, monocytes and neutrophils and some lymphocytes have Fc receptors for the Fc region of IgG.
- A consequence of binding to the Fc receptors on such cells is that the cells can now internalize the antigen better.



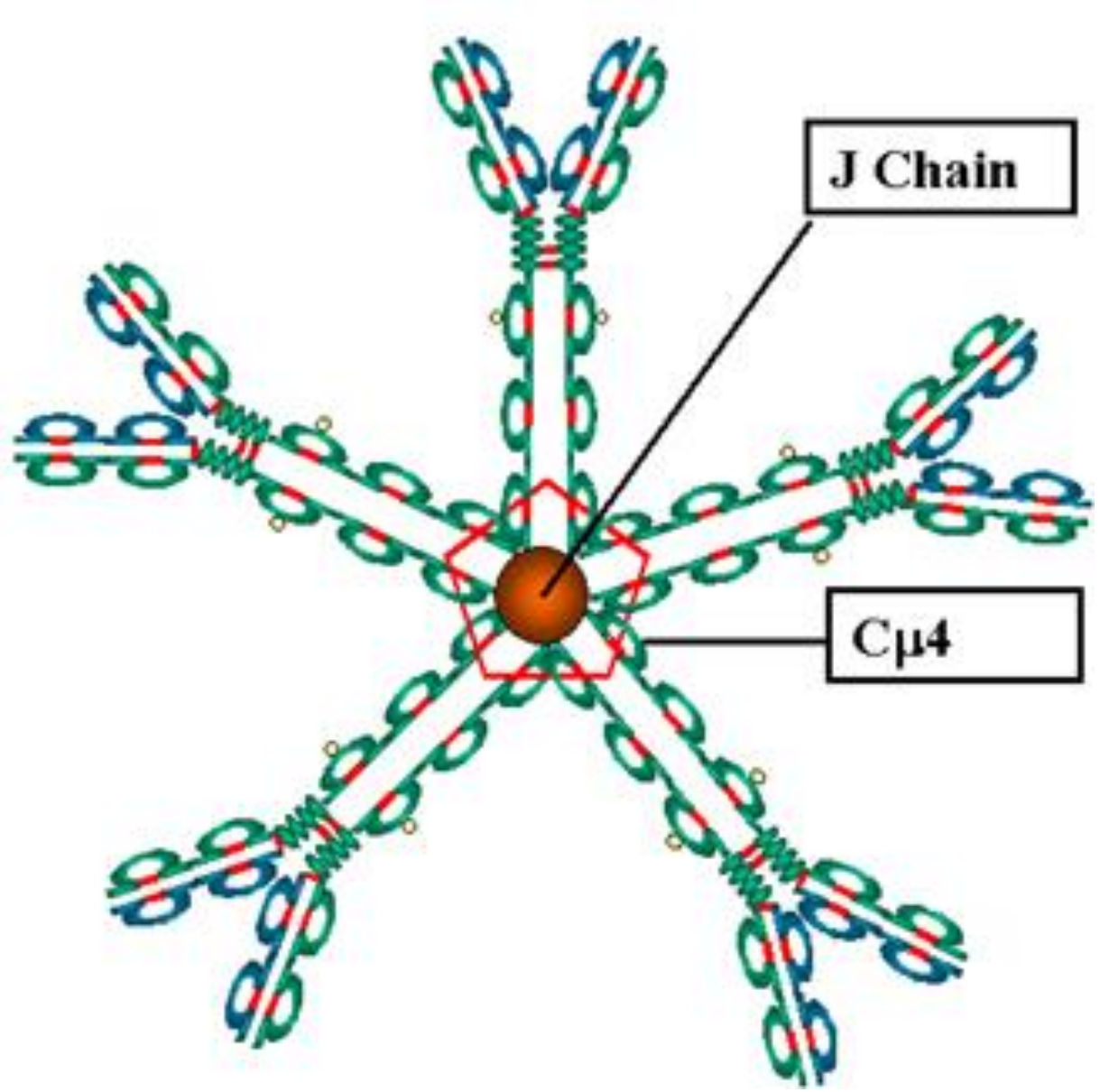
IgG- Function

- Secreted in high quantities in secondary exposures
- Cross the placenta
- Major functions / applications
  - neutralize microbes and toxins
  - opsonize antigens for phagocytosis
  - activate the complement
  - protect the newborn

- 4-fold rise or fall indicates active infection
- A single positive sample indicates past exposure

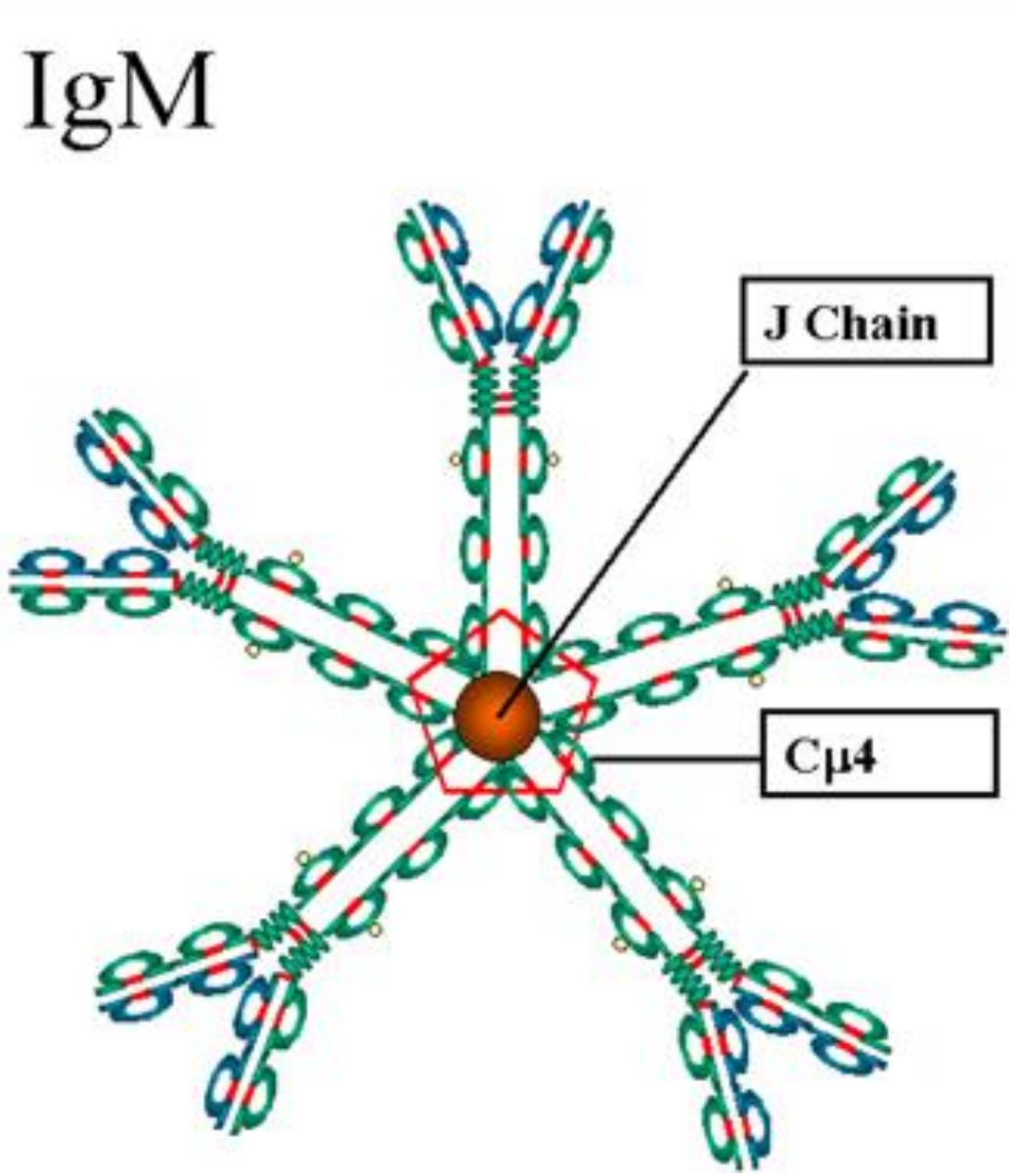
IgM- Structure

- IgM normally exists as a pentamer (19S immunoglobulin) but it can also exist as a monomer.
- In the pentameric form all heavy chains are identical and all light chains are identical.
- The valence is theoretically 10.



IgM- Structure

- IgM has an extra domain on the mu chain (CH4) and it has another protein covalently bound via a S-S bond called the J chain.
- This chain functions in polymerization of the molecule into a pentamer.



IgM- Properties

Third most common serum Ig.

First Ig to be made by the fetus

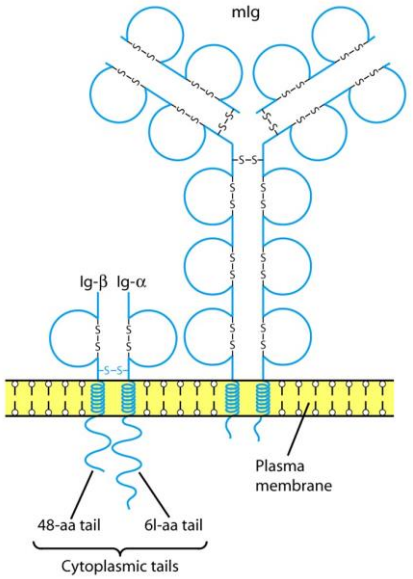
First Ig to be made by a virgin B cells when stimulated by antigen

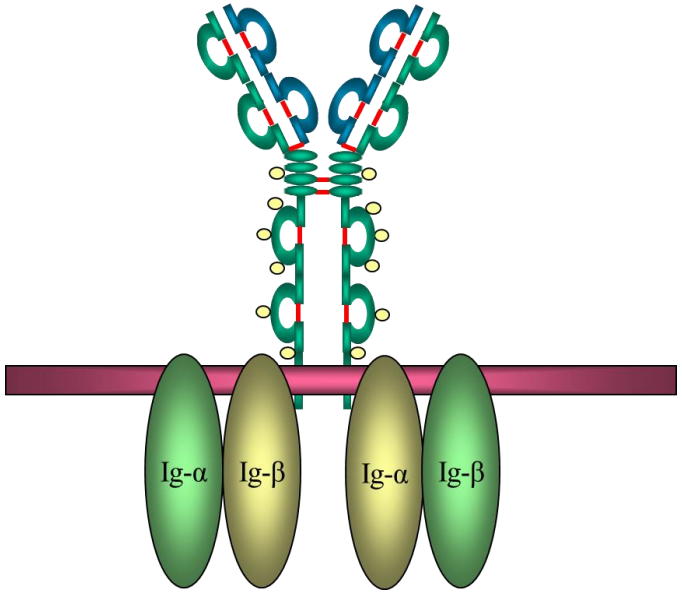
IgM- Properties (contd.)

A good complement fixing Ig.

Very efficient in leading to the lysis of microorganisms.

A good agglutinating Ig.





IgM- Properties (contd.)

- **Cellular binding**-IgM binds to some cells via Fc receptors.
- **B cell surface Ig**
- Surface IgM exists as a monomer and lacks J chain but it has an extra 20 amino acids at the C-terminus to anchor it into the membrane . Cell surface IgM functions as a receptor for antigen on B cells.

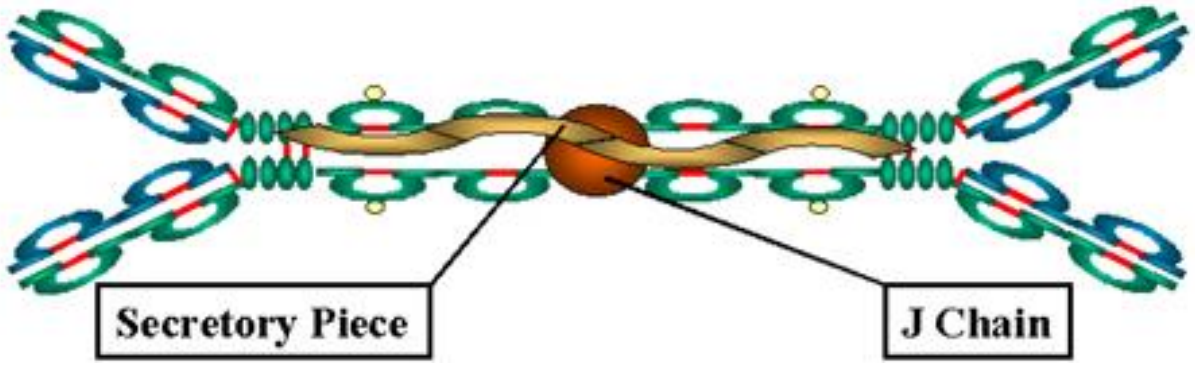
IgM

- Secreted initially during primary infection
- Cannot cross the placenta
- Major functions / applications
  - secreted first during primary exposure
  - activates the complement
  - used as a marker of recent infection

- Presence in newborn means infection
- Single positive sample in serum or CSF indicates recent or active infection
- Used to detect early phase of infection

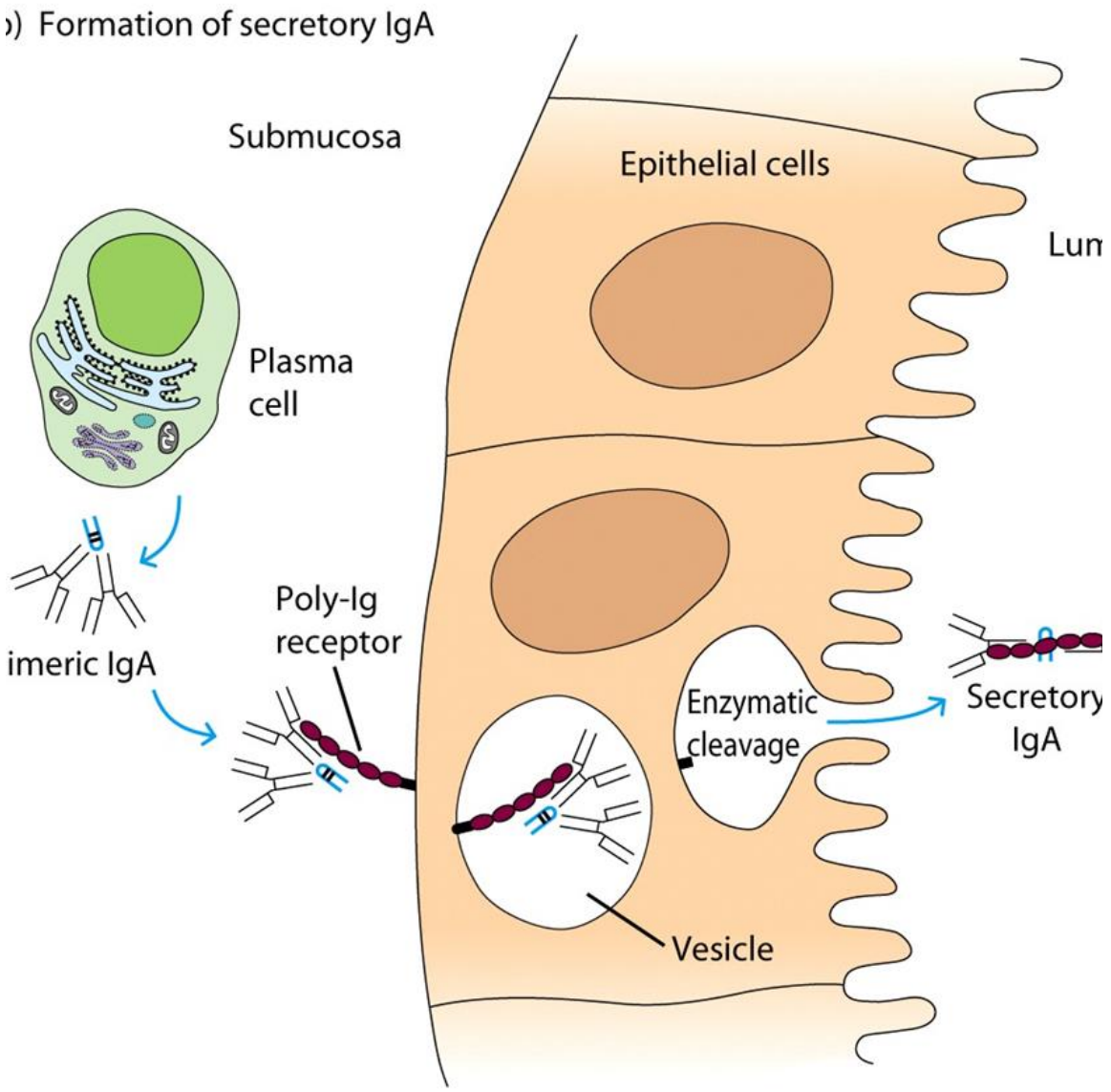
Ig A- Structure

- Serum IgA is a monomer,
- IgA found in secretions(sIgA)is a dimer.
- J chain is associated with dimeric form.
- A **secretory piece or T piece is also associated with secretory Ig A.**
- sIgA is sometimes referred to as 11S immunoglobulin



Secretory piece of Ig A

- Unlike the remainder of the IgA which is made in the plasma cell, the secretory piece is made in epithelial cells and is added to the IgA as it passes into the secretions
- The secretory piece helps IgA to be transported across mucosa and also protects it from degradation in the secretions.



IgA-Properties

a) 2nd most common serum Ig.

b) IgA is the major class of Ig in secretions - tears, saliva, colostrum, mucus. Since it is found in secretions secretory IgA is important in local (mucosal) immunity.

c) Normally IgA does not fix complement, unless aggregated.

d) IgA can bind to some cells - PMN's and some lymphocytes.

IgA

- Monomeric in serum
- Dimeric with secretory component in the lumen of the gastro-intestinal tract and in the respiratory tract
- Major function / application
  - neutralizes microbes and toxins

- Sero-diagnosis of tuberculosis
- Respiratory syncytial virus tests

IgD-Structure and Properties

1. Structure

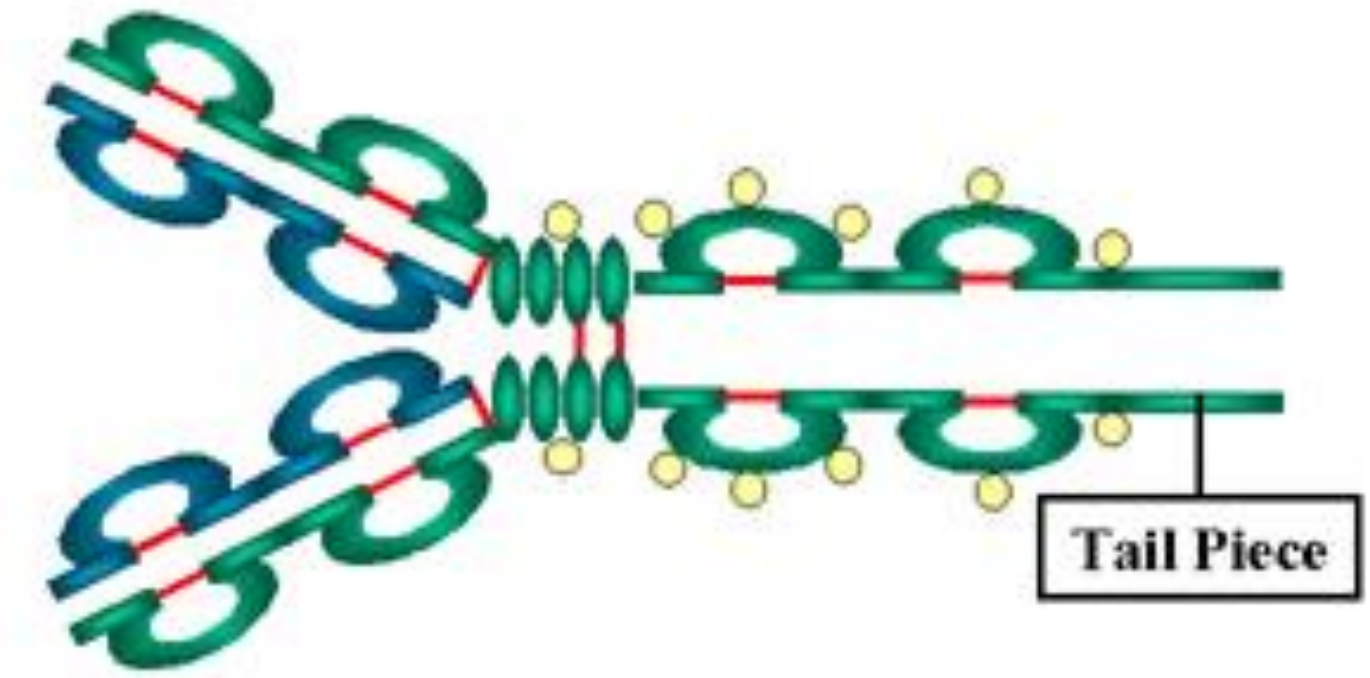
IgD exists only as a monomer.

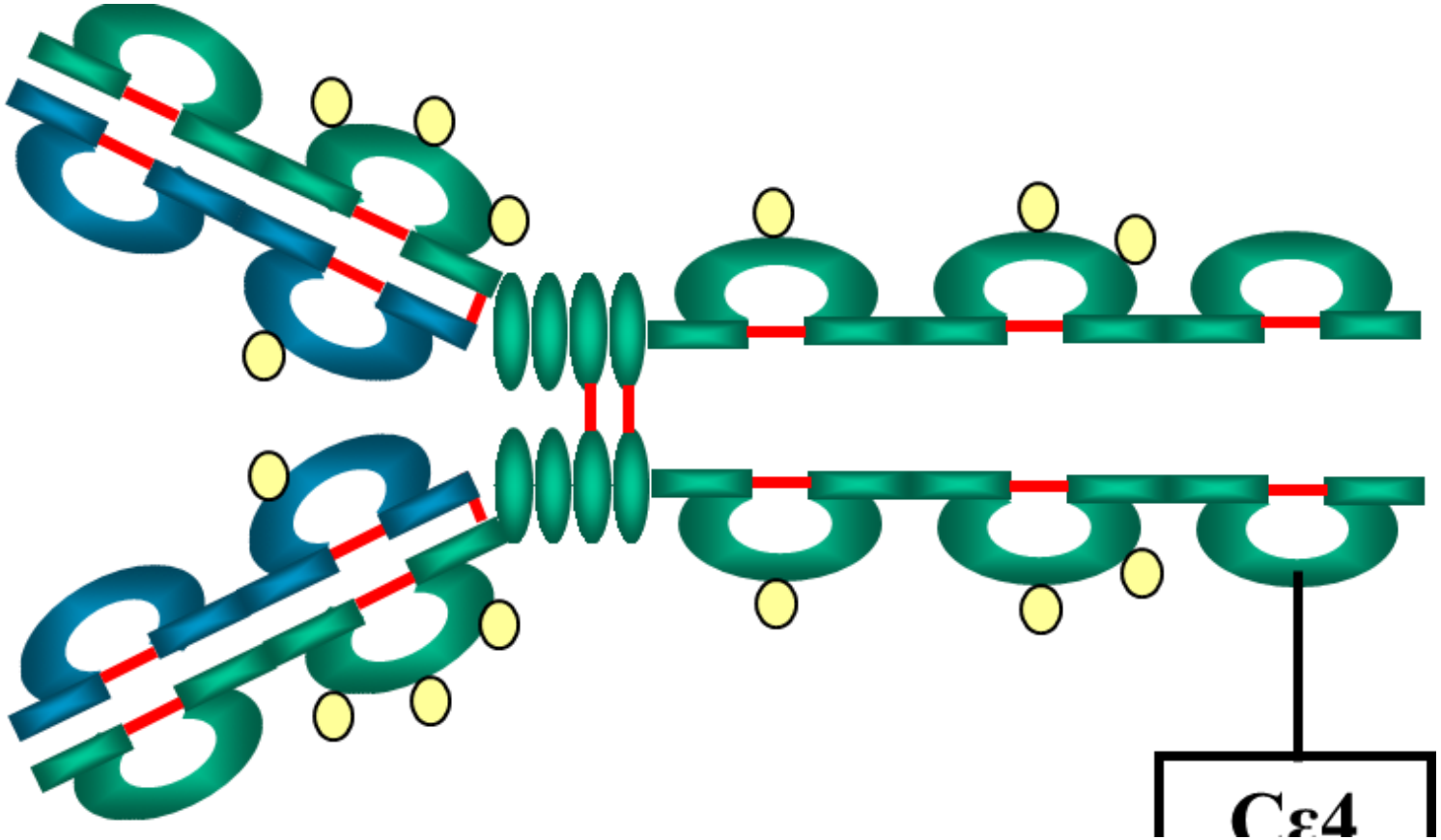
2. Properties

a) IgD is found in low levels in serum; its role in serum is uncertain.

b) IgD is primarily found on B cell surfaces where it functions as a receptor for antigen.

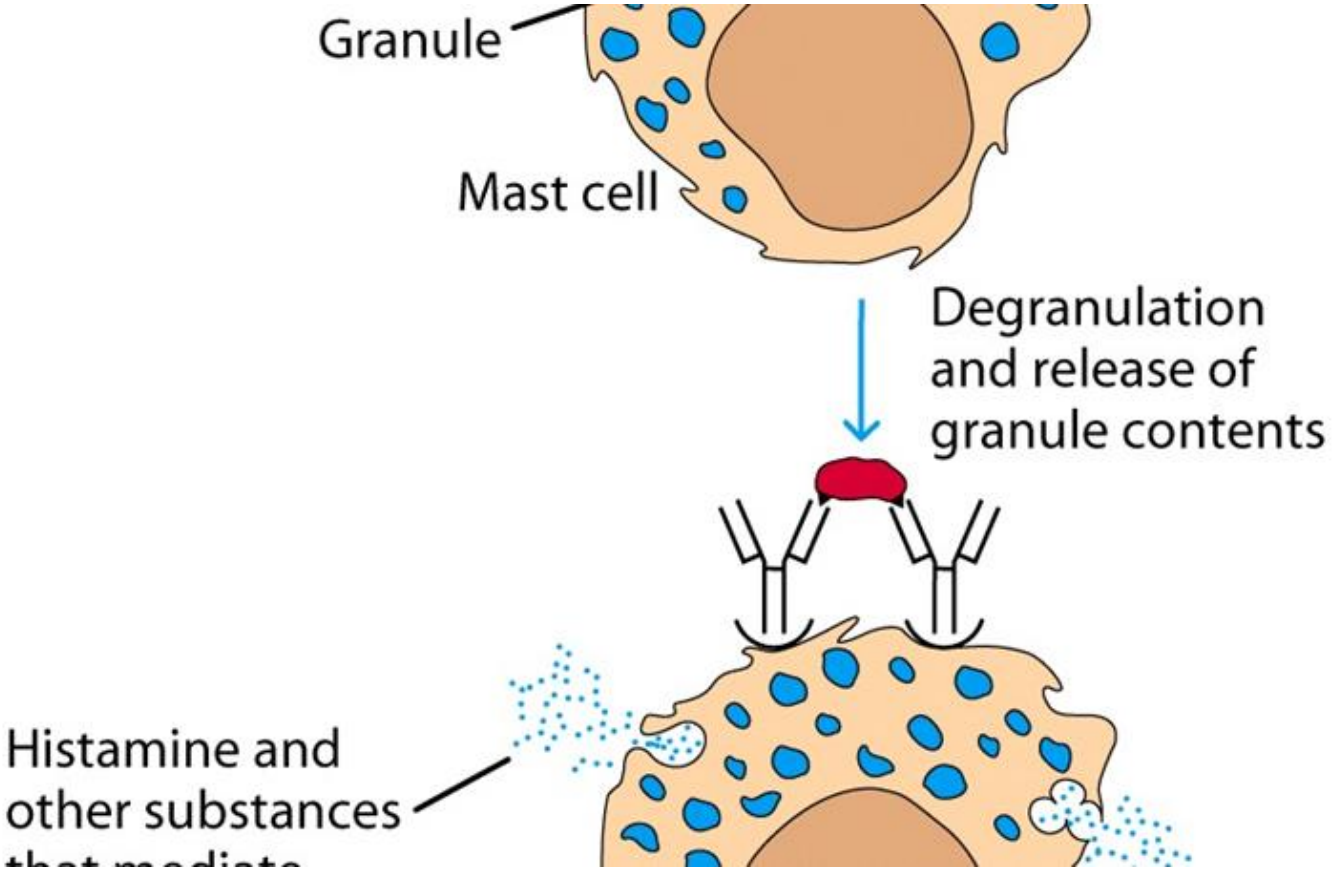
c) IgD does not bind complement





IgE-Structure

- IgE exists as a monomer and has an extra domain in the constant region.



Ig E-Properties-Homocytotropism

- a) IgE is the least common serum Ig since it binds very tightly to Fc receptors on basophils and mast cells even before interacting with antigen.
- b) IgE does not fix complement.

Ig E and allergic reactions

c) Involved in allergic reactions

As a consequence of its binding to basophils and mast cells, IgE is involved in allergic reactions.

Binding of the allergen to the IgE on the cells results in the release of various pharmacological mediators that result in allergic symptoms.

Ig E and parasitic helminth diseases

IgE also plays a role in **parasitic helminth diseases**.

Since serum IgE levels rise in parasitic diseases, measuring IgE levels is helpful in diagnosing parasitic infections.

Eosinophils have Fc receptors for IgE and binding of eosinophils to IgE-coated helminths results in killing of the parasite.

IgE

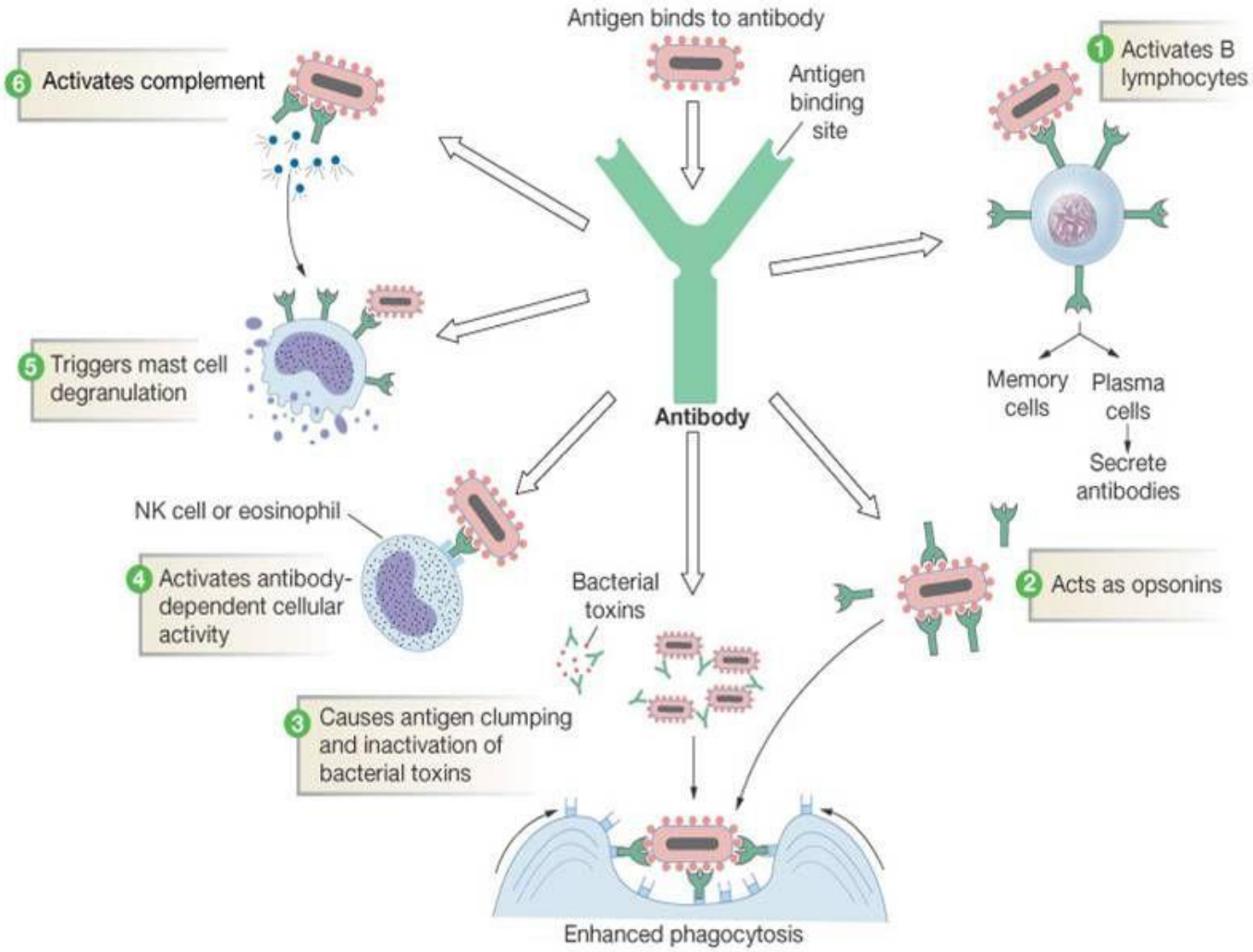
- Mediates type I hypersensitivity
- Monomeric
- Major functions / applications
  - associated with anaphylaxis
  - plays a role in immunity to helminthic parasites

Serodiagnosis of infectious and non infectious allergies (e.g., allergic bronchopulmonary aspergillosis, parasitic diseases)

Summary

- Immunoglobulins are glycoproteins
- There are five immunoglobulins based on variations in the heavy chain.
- IgG is the only antibody for placental transfer of immunity.
- Ig M is the most potent agglutinating antibody.
- Ig A acts as a mucosal barrier.
- IgE is the antibody for allergies.
- IgD and IgM are present on the surface of B lymphocytes.

Functions of Immunoglobulins- an overview



Thank you