Antigens and Antibodies

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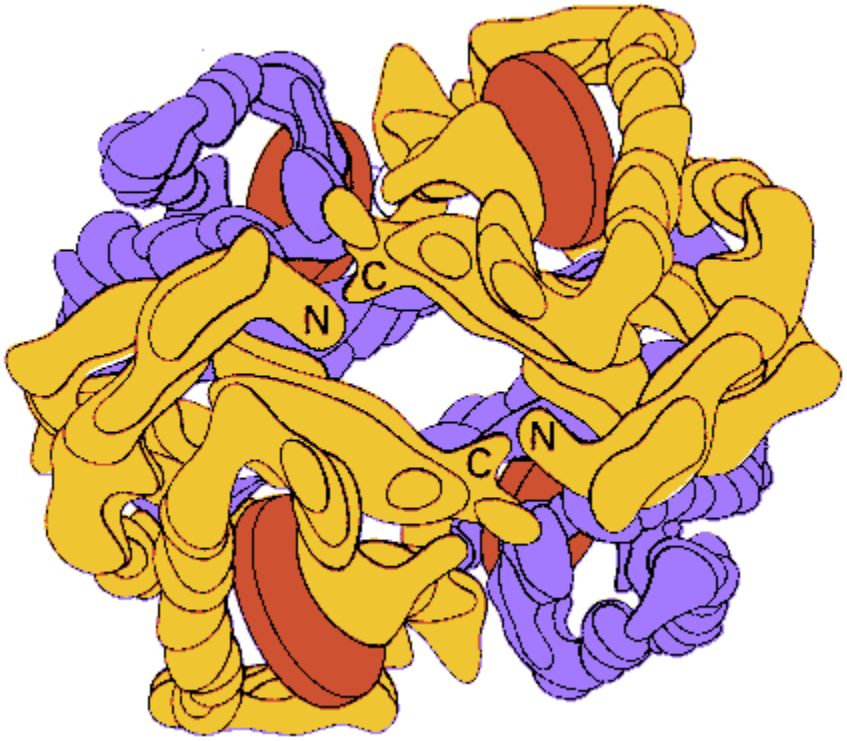
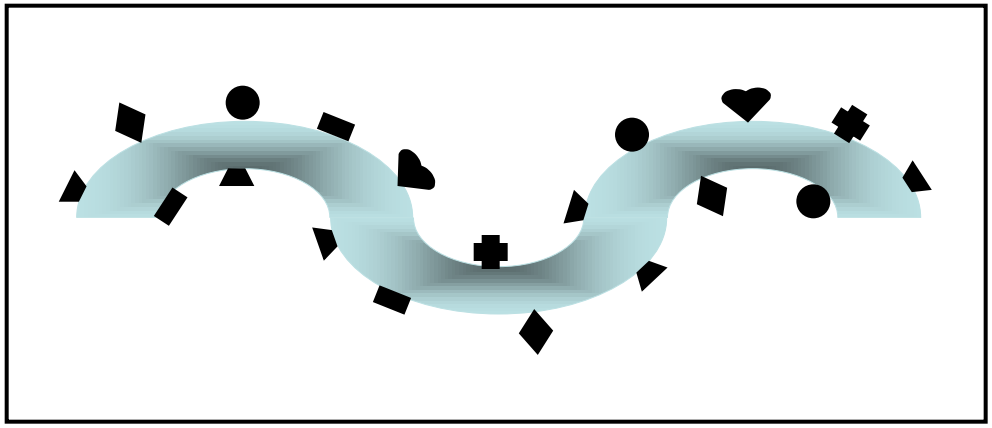
Antigen





- **Antigens** are substances that induce a specific immune response and subsequently react with the products of a specific immune response.
- The word originated from the notion that they can stimulate **anti**body **gen**eration.

Antigen

- There are two general types of antigens (Ags):

A. Exogenous (external): originate from outside the body, as viruses, bacteria and parasite. entered the body by inhalation, ingestion, or injection. For example by endocytosis or phagocytosis, This type of Ag presented by antigen presenting cells (APCs).

Antigen

B. Endogenous (internal) : are antigens that have been generated within previously normal cells as a result of abnormal cell metabolism, or because of viral or intracellular bacterial infection and bacterial toxins.

- Superantigen: A class of antigens that cause activation of large number of T cells leads to the release of high levels of cytokines and severe outcome to the host, including shock and death, e.g. toxic shock syndrome include:
  - Staphylococcal enterotoxins
  - Staphylococcal toxic shock toxin
  - Staphylococcal exfoliating toxin
  - Streptococcal pyrogenic exotoxins

- These antigenic molecules may have several antigenic determinants, called epitopes, and each epitope can bind with a specific antibody. Thus, a single antigen can bind to many different antibodies with different binding sites.

Determinants Recognized by the Innate Immune System

. PAMPs – Pathogen Associated Molecular Patterns; on microbe

. PRRs – Pattern Recognition Receptors; on cells

Properities of Immunogenicity

A-Foreignness: To be immunogenic, molecules must be recognized as “non-self” foreign.

B-Molecular size: 10.000-100.000 Dalton MW is imunogenetically

C-Chemical structural complexity: proteins are the most potent immunogens and most polysaccharides are weak antigens. complexity is required e.g. amino acid homopolymers are less immunogenic than heteropolymers.

D-Genetic factors of host: Some substances are immunogenic in one species or individual but not in other. The species or individuals may lack or have altered genes that code for the recognition of antigens.

E-Route of entry, Dose and Physical form.

Generally, the subcutaneous or intramuscular route is better than the intravenous or intragastric route.

Dose, increasing the dose or repeating it gives a better immune response.

Physical form, particulate Ags are more immunogenic than soluble and denatured Ag more immunogenic than the native form.

Immunoglobulins

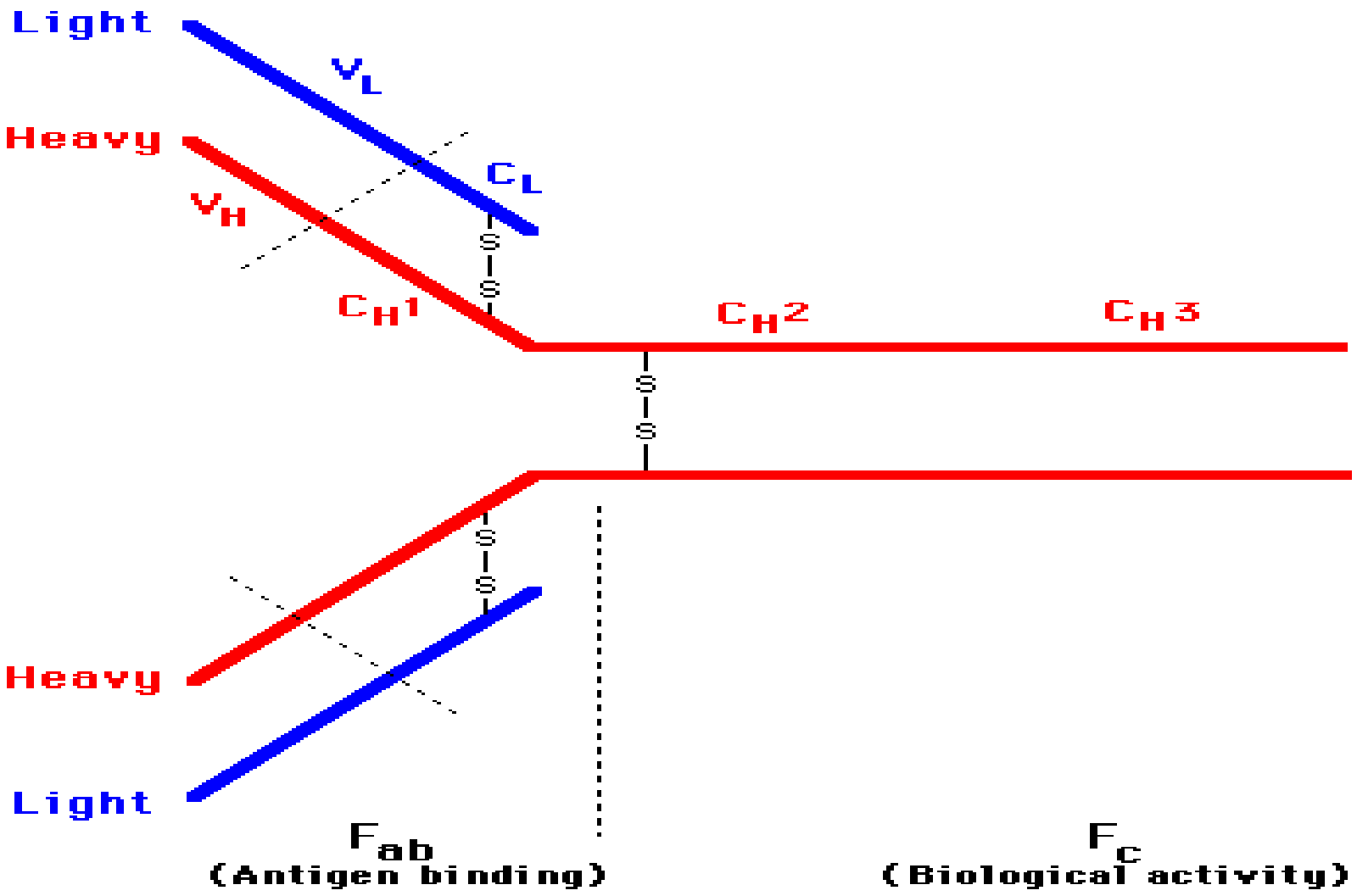
Structure and Function

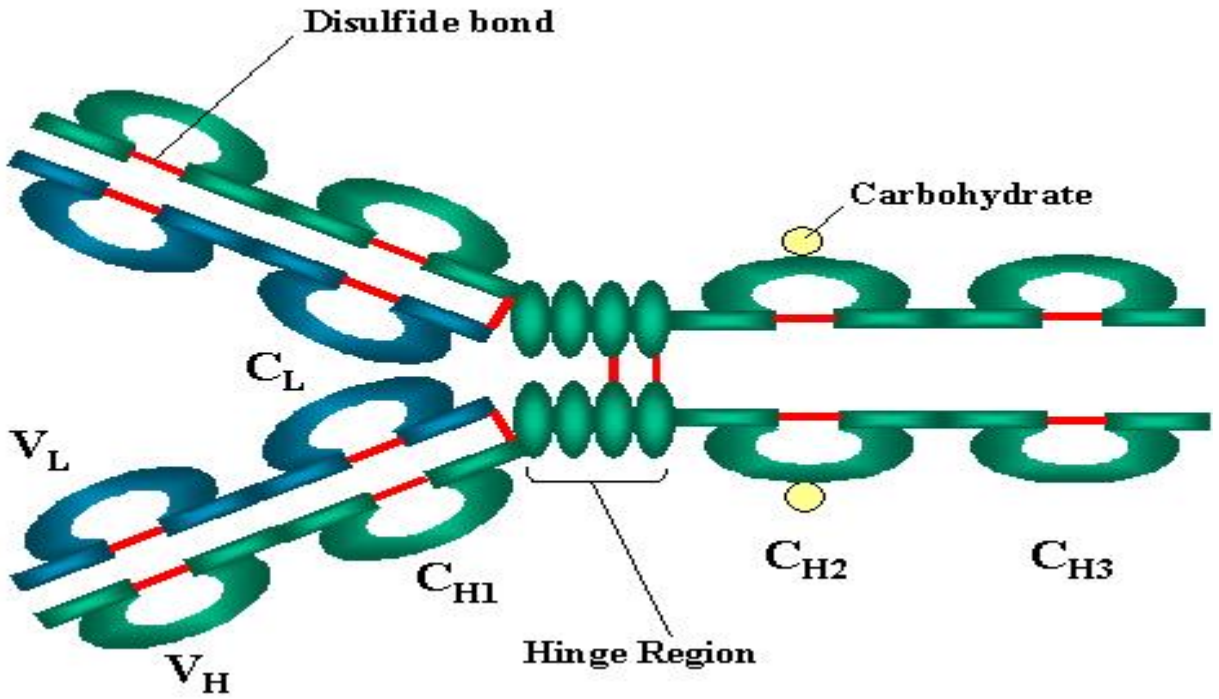
Immunoglobulin (Ig):

Immunoglobulins (antibodies) are glycoprotein molecules that are produced by **plasma cells** in response to an immunogen and which function as antibodies.

BASIC STRUCTURE OF IMMUNOGLOBULINS

Although different immunoglobulins can differ structurally, but all Igs built from the same basic units.


Light
V_L
Heavy
C_L
V_H
C_{H1}
C_{H2}
C_{H3}
S
S
S
S
S
S
Heavy
Light
F_{ab}
(Antigen binding)
F_C
(Biological activity)


Disulfide bond
Carbohydrate
C_L
V_L
C_{H1}
C_{H2}
C_{H3}
Hinge Region
V_H

A. Heavy and Light Chains

All immunoglobulins have a four chain structure as their basic unit. They are composed of two identical light chains and two identical heavy chains.

B. Disulfide bonds

1. Inter-chain disulfide bonds - The heavy and light chains and the two heavy chains are held together by inter-chain disulfide bonds. The number of inter-chain disulfide bonds varies among different immunoglobulin molecules.

2.Intra-chain disulfide bonds - Within each of the polypeptide chains there are intra-chain disulfide bonds.

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

The heavy and light chain could be divided into two regions based on variability in the amino acid sequences.

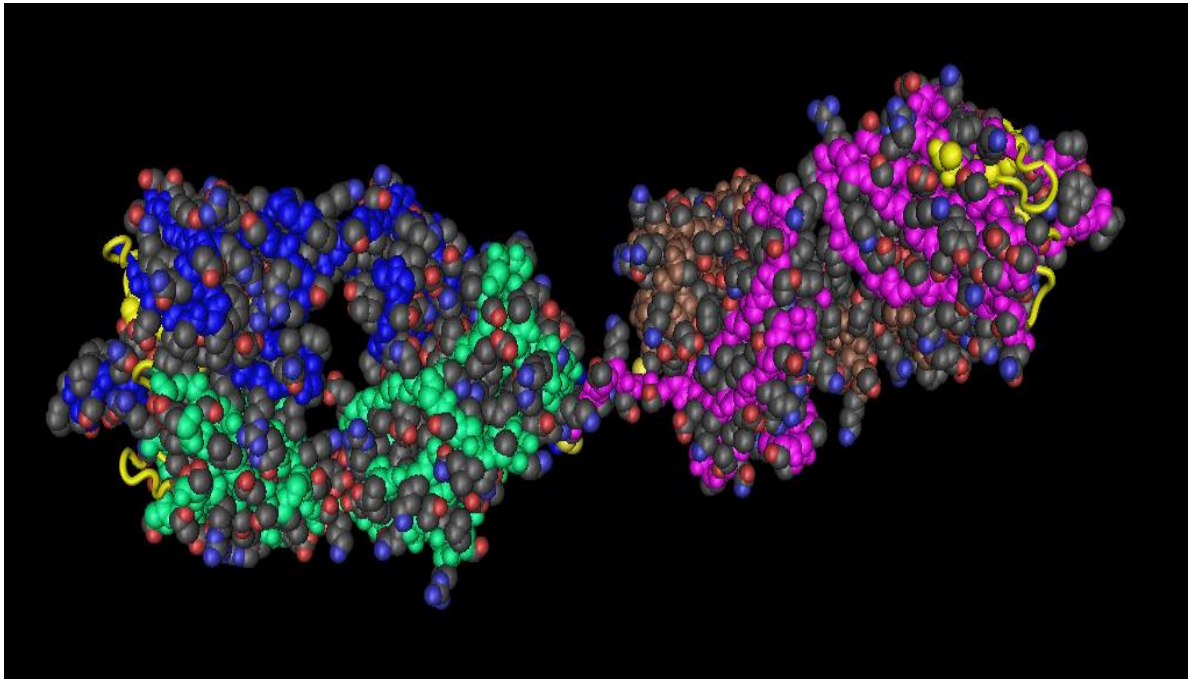
1. Amino acid terminal: in which the amino acid is variable, called **variable region**.
2. Carboxyl terminal: the amino acid is rather than constant, called **constant region.**

D. Hinge Region

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

E. Domains

- The amino acid sequence in both light and heavy chain is not a linear sequence but there are domes and loops due to the presence of intra chain disulfide bonds these globular areas called domains.



IMMUNOGLOBULIN FRAGMENTS

Digestion of Igs with papain breaks the immunoglobulin molecule at the hinge region, and produces three fragments, two identical fragments (Fab) and one fragment (Fc).

A. Fab: **F**ragment **a**ntigen **b**inding - these fragments were called the Fab fragments because they contained the antigen binding sites of the antibody. It is also called **paratope**, is a part of an antibody which recognizes and binds to an antigen.

- The combining site of the antibody is created by both VH and VL.

B. Fc : **F**ragment **C**rystallization: This fragment was called Fc because it was easily crystallize

IMMUNOGLOBULIN CLASSES, SUBCLASSES AND TYPES

A. 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

B. Immunoglobulin Subclasses

The classes of immunoglobulins can be divided into subclasses based on small differences in the amino acid sequences in the constant region of the heavy chains.

1. IgG Subclasses

a) IgG1 - Gamma 1 heavy chains

b) IgG2 - Gamma 2 heavy chains

c) IgG3 - Gamma 3 heavy chains

d) IgG4 - Gamma 4 heavy chains

2. IgA Subclasses •

a) IgA1 - Alpha 1 heavy chains •

b) IgA2 - Alpha 2 heavy chains •

C. 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.

**Kappa light chains •

**Lambda light chains •

GENERAL FUNCTIONS Of IMMUNOGLOBULINS

A. Primary function: Antigen binding

- Immunoglobulins bind specifically to one or a few closely related antigens.

B. Secondary function: Effector Functions

- The binding of an antibody to an antigen has no direct biological effect. Rather, the significant biological effects are secondary "effector functions" of antibodies.
- Not every immunoglobulin will mediate all effector functions. Such effector functions include:
  -

- **1. Agglutination:** Agglutination of particulate antigen, including bacteria and viruses. IgM is suitable for this function.
- **2. Opsonization:** coating of bacteria with antibody's Fab region (IgG). This facilitates phagocytosis by cells possessing Fc receptor, e.g. neutrophil, polymorphonuclear leucocytes"..
- **3. Neutralization:** Neutralization of toxins released by bacteria e.g. tetanus toxin is neutralized when specific IgG antibody binds, thus preventing the toxin binding to it’s receptor. In the case of viruses, antibodies can hinder their ability to attach to receptors on host cells.

4. Complement activation (classical pathway): by IgM and IgG, leads eventually to death of bacteria by the terminal complement components which make holes in the cell wall, leading to an **osmotic death**.

5. Precipitation: Precipitation of soluble antigens by immune complex formation. They can be removed by phagocytic cells, and can fix complement.

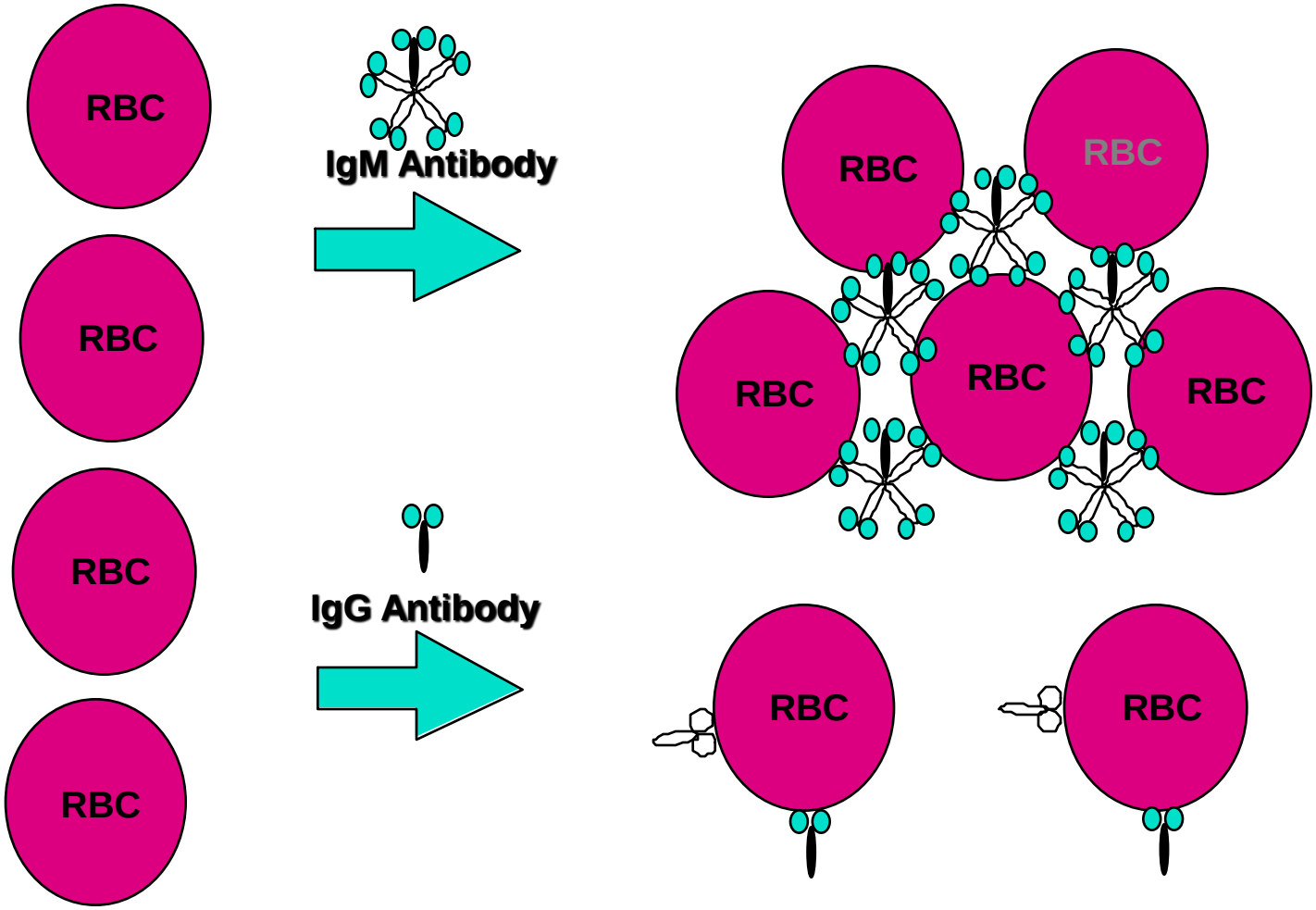
6. Antibody dependent cell mediated cytotoxicity (ADCC):

Antibodies bind to organisms via their Fab region. Large granular lymphocytes NK cells, attach via Fc receptors, and kill these organisms not by phagocytosis but by release of toxic substances called perforins.

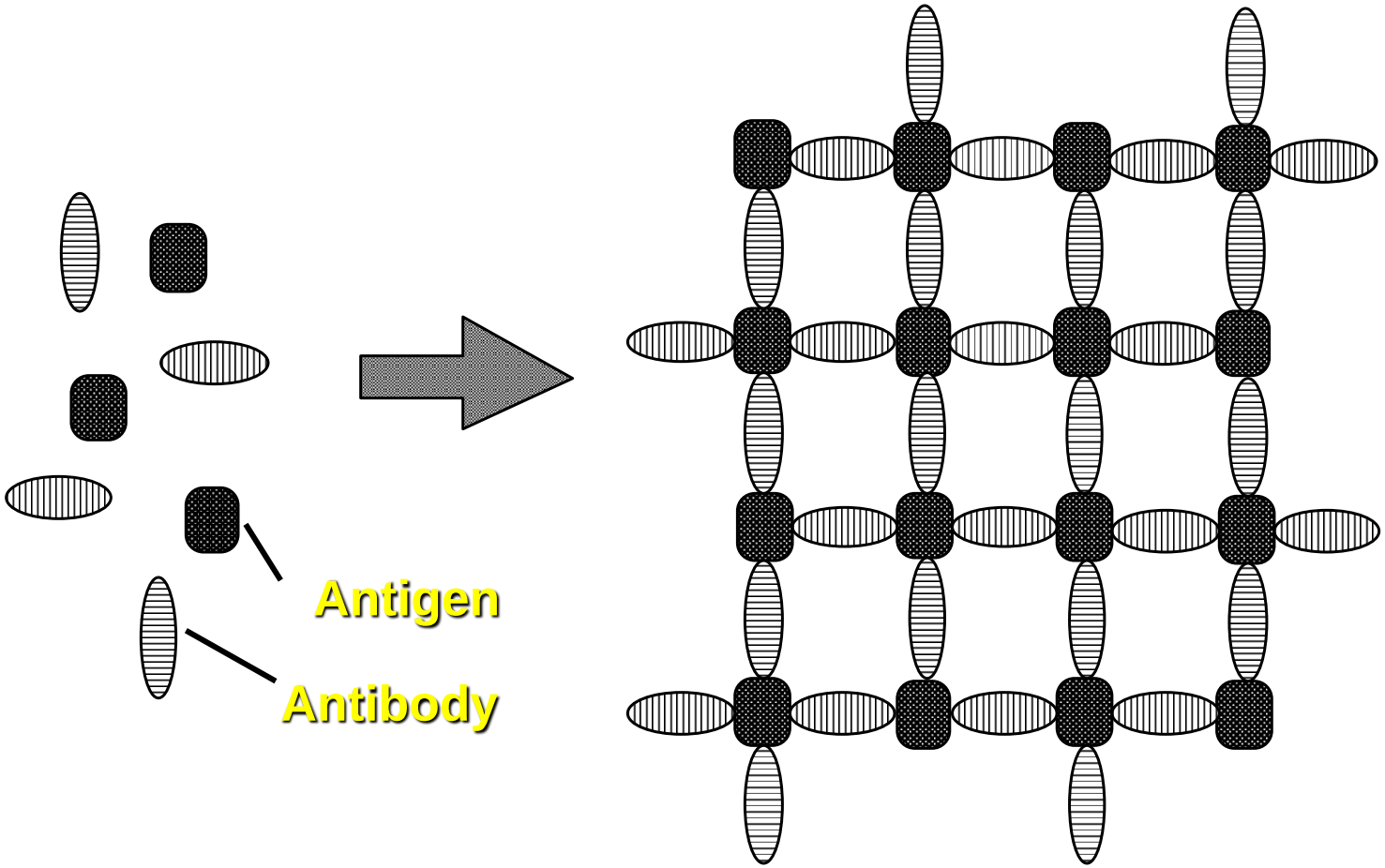
Antibody dependent cell mediated cytolysis (ADCC) - Antibodies serve as bridge b/w the infected target cell and effector cell (Natural killer cells).

7. Mucosal protection: mediated byIgA, it acts chiefly by inhibiting pathogens from gaining attachment to mucosal surfaces.

Agglutination



Immune Precipitation



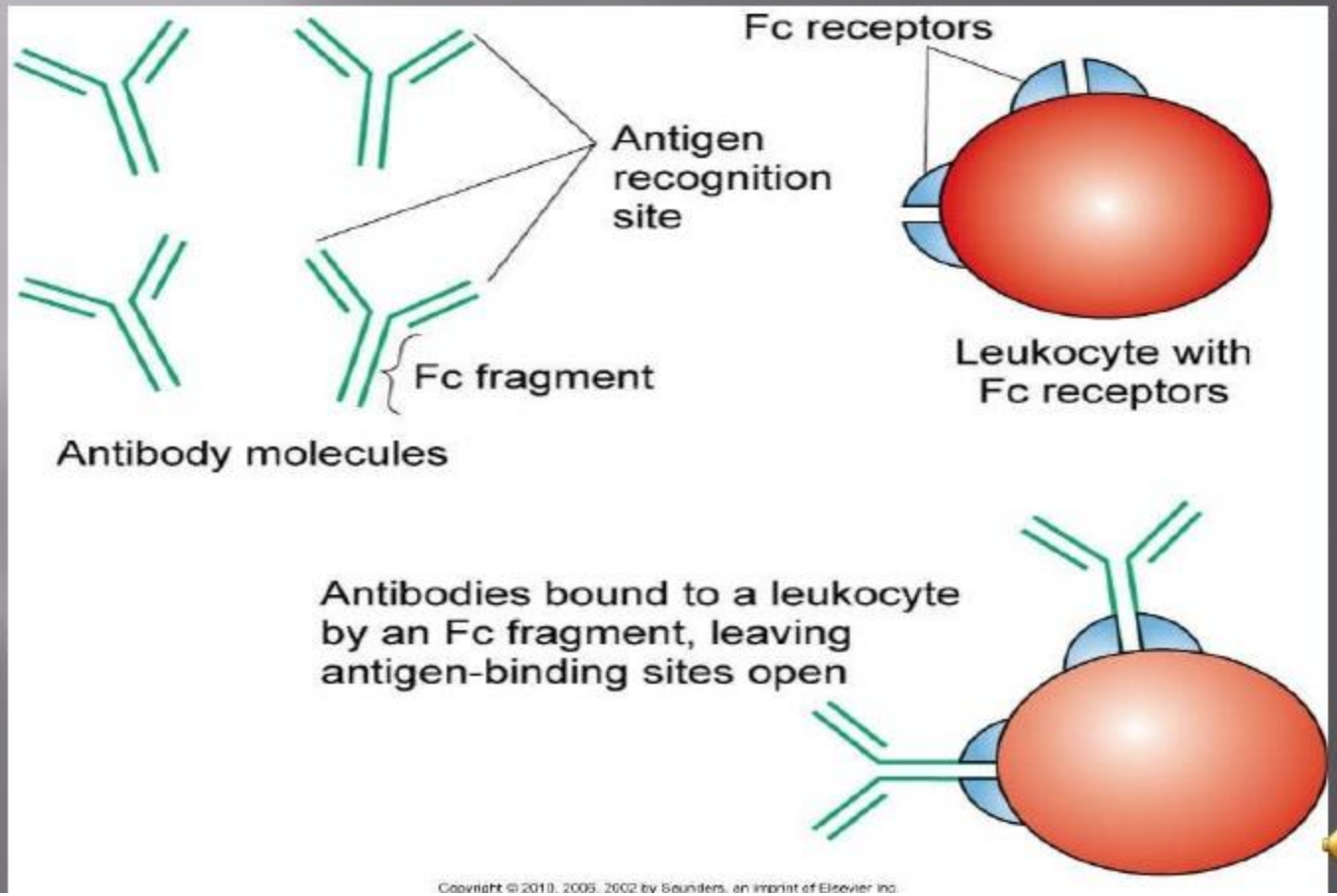
neutralisation of toxins

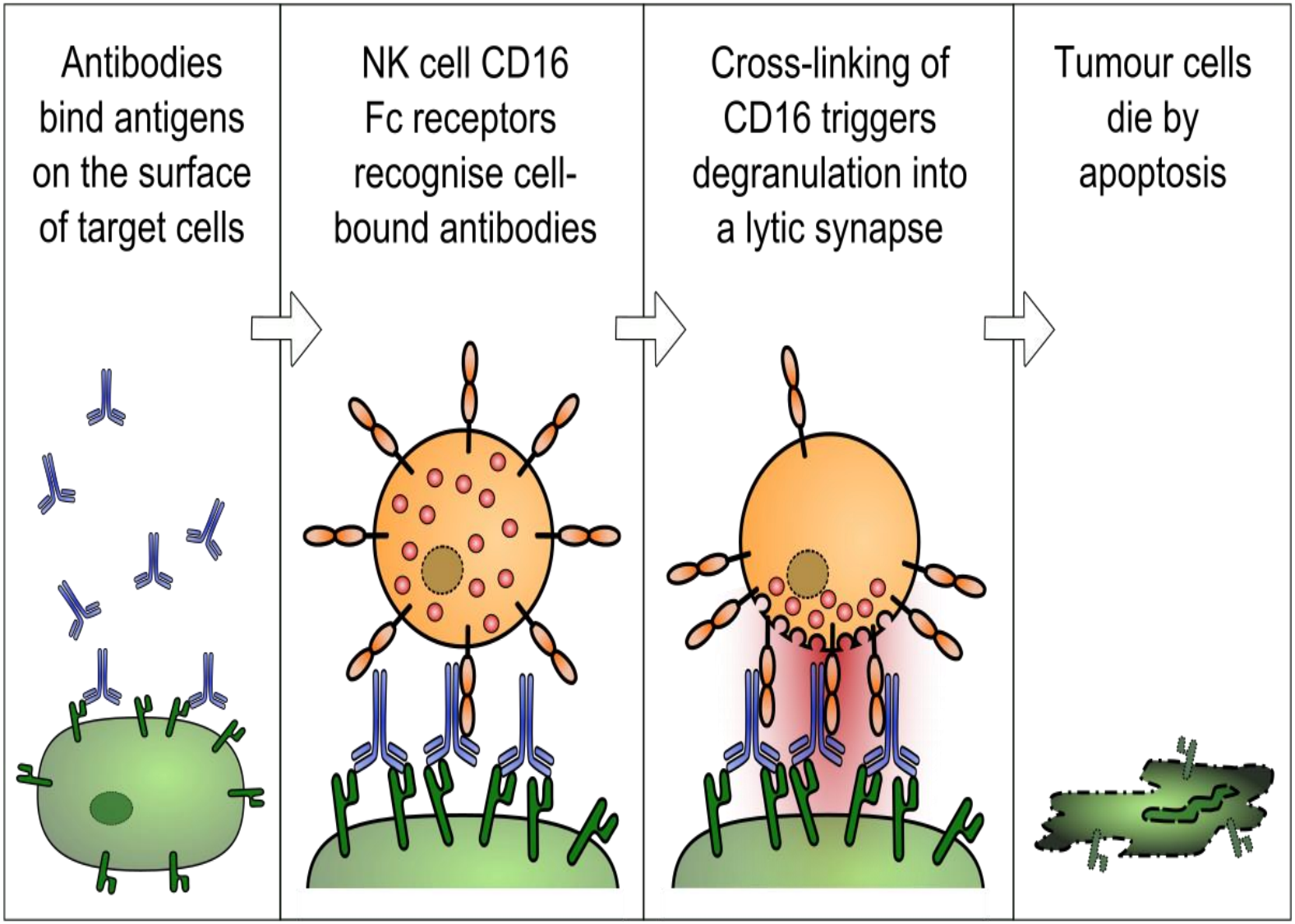
opsonisation of pathogens

activation of complement

activation of effector cells

Antibody-Mediated Immunity Action



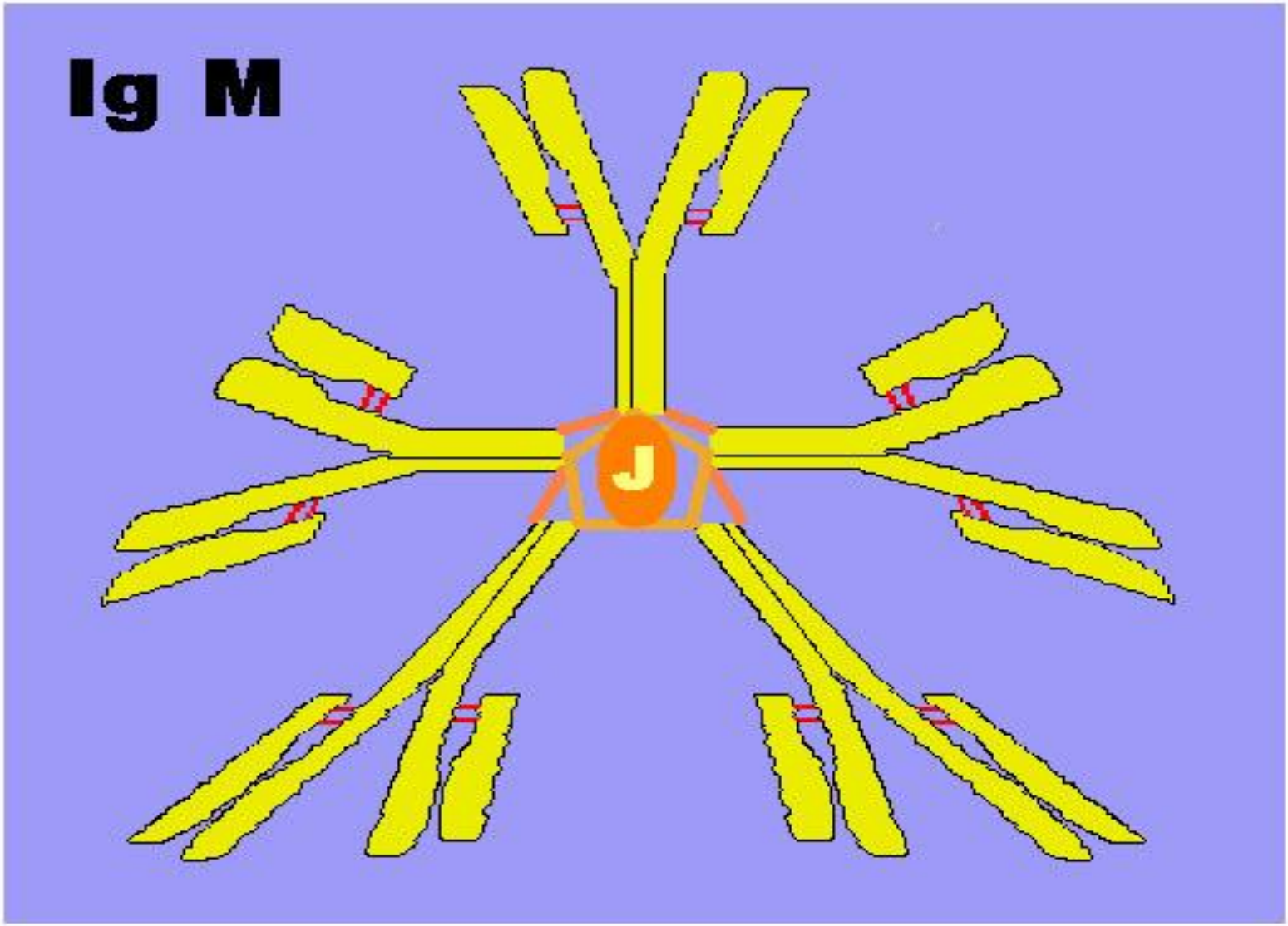

Antibodies bind antigens on the surface of target cells
NK cell CD16 Fc receptors recognise cell-bound antibodies
Cross-linking of CD16 triggers degranulation into a lytic synapse
Tumour cells die by apoptosis

Properties of Igs
IgG

- Monomer, 70-75% of total immuniglobulin
- Secreted in high quantities in secondary exposures
- Cross the placenta
- Most protective because it is capable of carrying out all effector functions of immunoglobulins.
- **Major functions / applications**
  - neutralize microbes and toxins
  - opsonize antigens for phagocytosis
  - activate the complement
  - protect the newborn

IgM

- Normally pentamer linked by **J chain**, but it can also exist as a monomer on the surface of B cells
- Secreted initially during primary infection
- Cannot cross the placenta
- **Major functions / applications**
  - secreted first during primary exposure
  - activates the complement
  - functions as Ag receptor on B cell
  - the first Ig made by the fetus and the first Ig to be made by a virgin B cells when it is stimulated by antigen.
  - used as a marker of recent infection


Ig M
J

IgA

- Monomeric in serum
- Dimeric with secretory component **(J chain)** in secretion.
- **Major function**
  - neutralizes microbes and toxins
- When IgA is found in secretions is also has another protein associated with it called the secretory piece or T piece. the secretory piece is made not in plasma cell, but in epithelial cells and 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.**

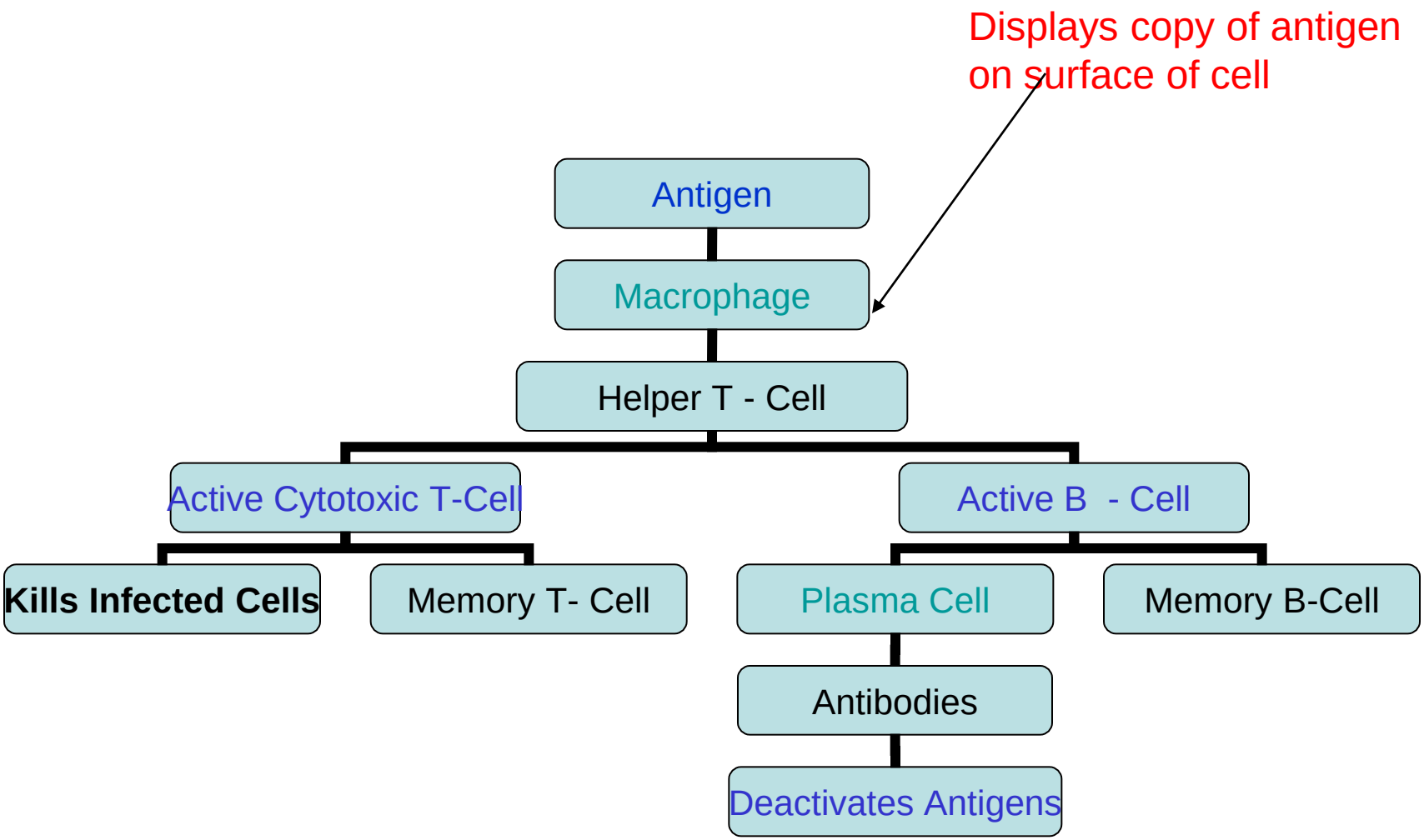
IgD

- Monomeric
- **Major functions**
  - present on the surface of B lymphocytes as Ag receptor

IgE

- Mediates type I hypersensitivity
- Monomeric
- **Major functions**
  - Mediates type I hypersensitivity
  - Plays a role in immunity to helminthic parasites

Immune Response Summary



Cellular Immunity

Antibody Immunity

I-First exposure (primary humoral immune response)

a- Lag phase: in the first few days, Ab level in serum is zero and no Ab production.

Lag phase duration depend on, nature of Ag, dose, route of entry and immune status.

b-Log phase: Ab produce and increasing.

c-Plateau phase or stationary phase: Ab production stop at certain level.

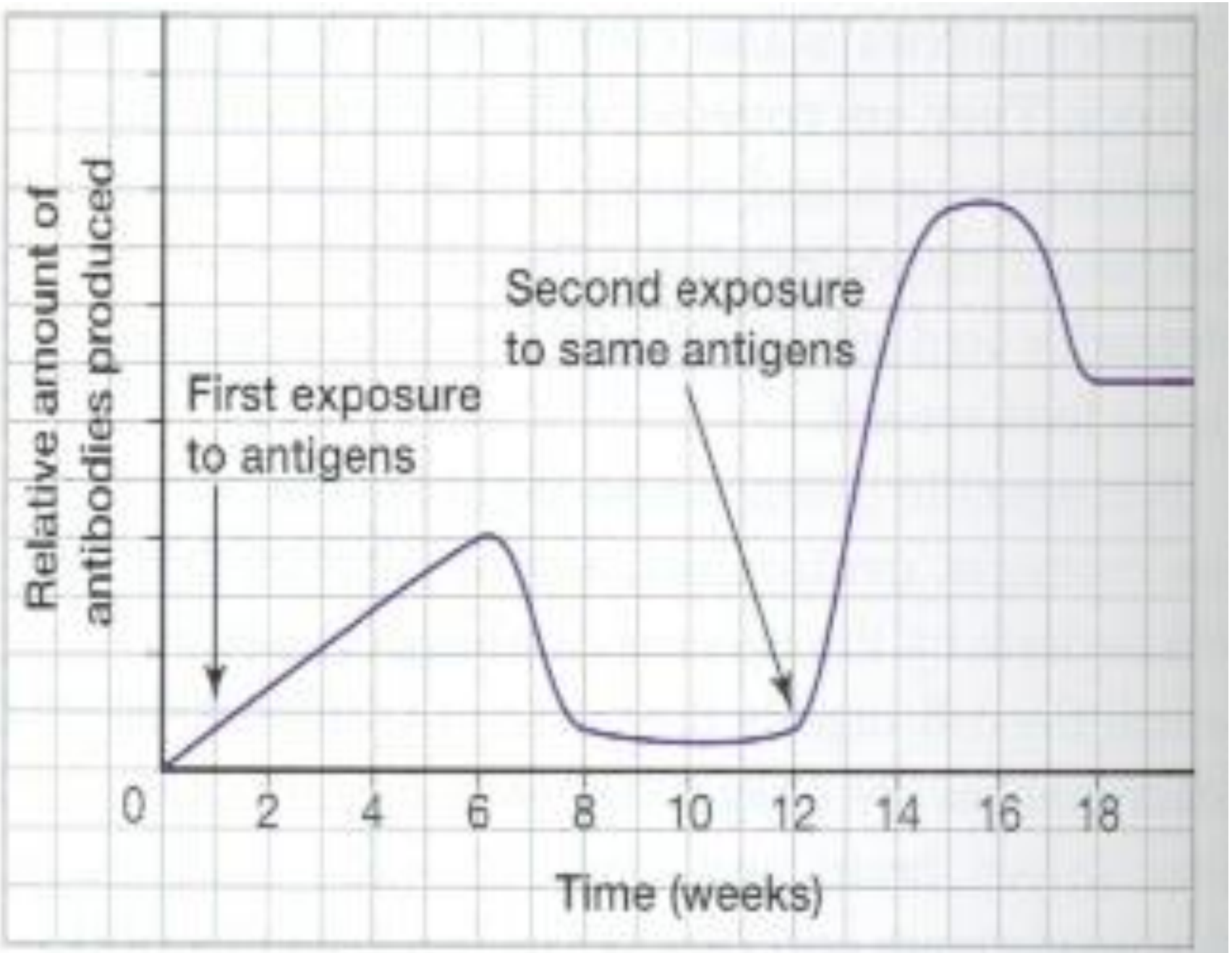
d-Decline phase: Ab titer decrease because of combination with Ag.

II-Second exposure (secondary humoral immune response)

a-There is memory cells (no need for lag phase).

b-There is high level of Ab.

c- Only those cells with identical receptors will be stimulated (high affinity).


Relative amount of antibodies produced
First exposure to antigens
Second exposure to same antigens
0
2
4
6
8
10
12
14
16
18
Time (weeks)

IgM – IgG sequential response