Immunology
3ed stage
Dr .Thabit moath
Le No 19

Immunology of transplantation

Graft or Transplant: Transfer of living cells, tissues and organs from one part of the body to another or from one individual to another.
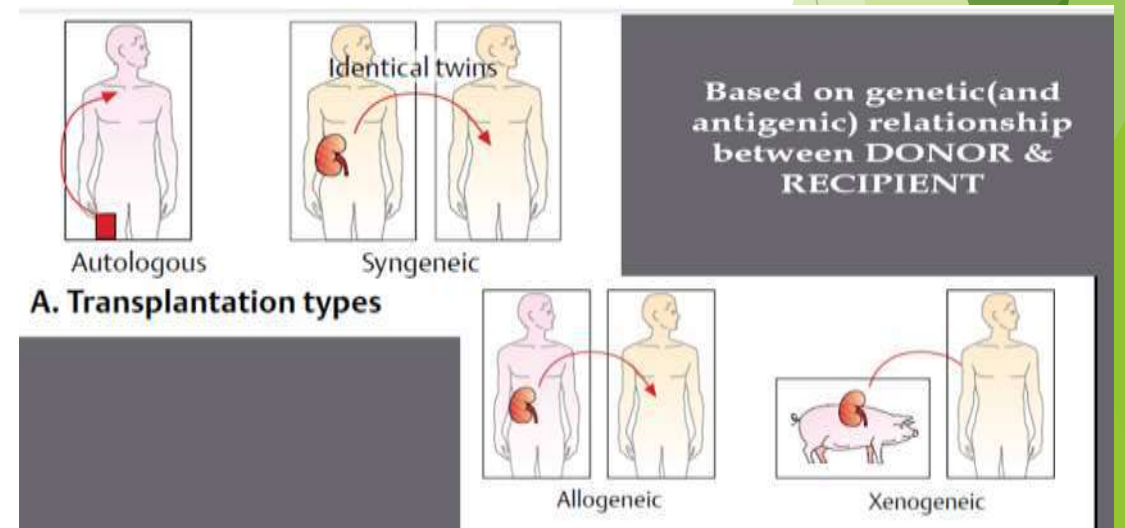
The transplantation mainly based on:

1. Organ or tissue transplanted

2. Anatomical site of origin of transplant & site of its placement:

a. Orthotopic: normal sites

b. Heterotopic: abnormal sites

1. Genetic compatibility and antigenic relationship.

2. Fresh or stored transplanted tissue :

Types of Transplantation:-

1. Autologous .

2. Syngeneic.

3. Allogenic .

4. Xenogeneic .

Auto grafting (Autologous) -

Transfer of self-tissue from one body site to another in the same individual, it should:

Autologous   Syngeneic
A. Transplantation types

Identical twins

Based on genetic(and antigenic) relationship between DONOR & RECIPIENT
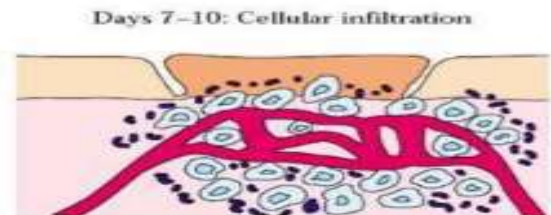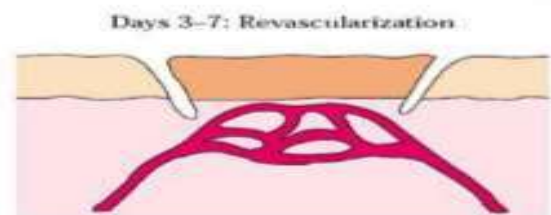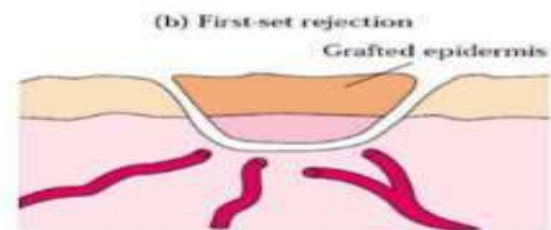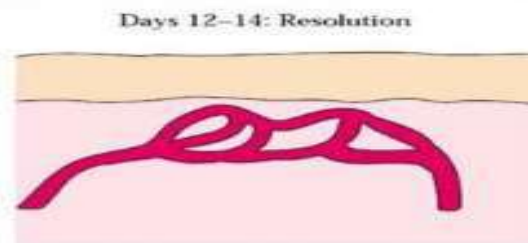
Allogeneic   Xenogeneic

Genetic homology of the tissue- immune system does not respond (Skin,hiar grafts)   Auto graft acceptance epidermis:-

1. After 3- 7 days have revascularization of blood vesicles.

2. 7 – 10 days healing.

3. 12 -14 neutrophil resolution,

Allograft reaction:-

First Set Response :- Skin graft from a genetically unrelated animal of same species . Initial acceptance , Thrombosed and necrosed Mainly by T lymphocytes .

-     After 3- 7 days have revascularization of blood vesicles .

-     7 – 10 cellular infiltration ,

-     10 -14 thromboses and necrosis ,

-      > 14 day damaged blood vesicles and rejection the implanted tissues.

(a) Autograft acceptance
Grafted epidermis
Blood vessels

Days 3–7: Revascularization

Days 7–10: Healing
Neutrophils

Days 12–14: Resolution

(b) First-set rejection
Grafted epidermis

Days 3–7: Revascularization

Days 7–10: Cellular infiltration

Days 10–14: Thrombosis and necrosis
Blood clots
Damaged blood vessels

Second Set Response :- If an animal has rejected a graft by the first set response, another graft from the same donor is applied – rejected in an accelerated manner , Mainly by antibodies

Effecter mechanism of allograft rejection:-

- **Hyper acute Rejection**

a. Pre-existing specific antibodies in high titers in the host circulation bind to donor endothelial antigens.

b. Activates Complement Cascade.

c. Characterized by thrombotic occlusion of graft

d. Graft remains pale

e. Rejected within minutes or hours, even without an attempt at vascularization

Hyperacute Rejection

Endothelial cell

Blood vessel

Alloantigen (e.g., blood group antigen)

Circulating alloantigen-specific antibody
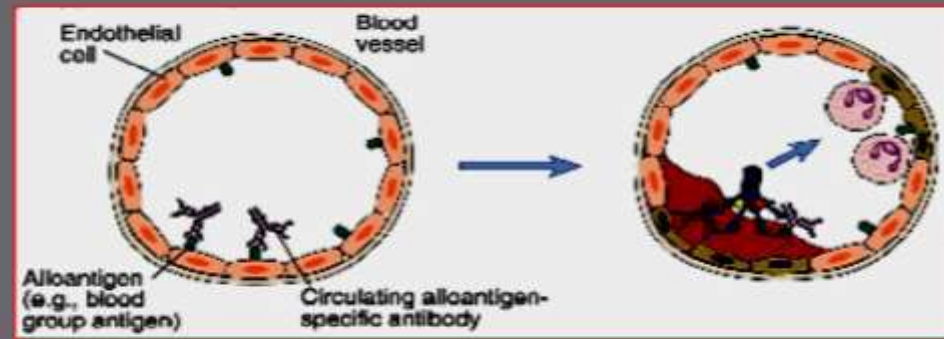
1. Preformed Ab, 2. complement activation,
3. neutrophil margination, 4. inflammation,
5. Thrombosis formation

Immunological Enhancement: -

Humoral antibodies can **act in opposition to CMI by inhibiting graft rejection.**

- **Afferent inhibition**: Combine with antigens released from graft so that they are unable to initiate an immune response

- **Central inhibition**: Antibodies may combine with lymphoid cell, by a negative feedback, render them incapable of responding to the antigens of the graft.

- **Efferent inhibition**: By coating the surface of cells in the graft so that sensitized lymphocytes are kept out of contact with them .

Acute Rejection: -

- Vascular and parenchymal injury mediated by T cells and antibodies that usually begin after first week of transplantation if **no immunosuppressant therapy**

- Incidence is high (30%) for the first 90 days.

Acute Rejection
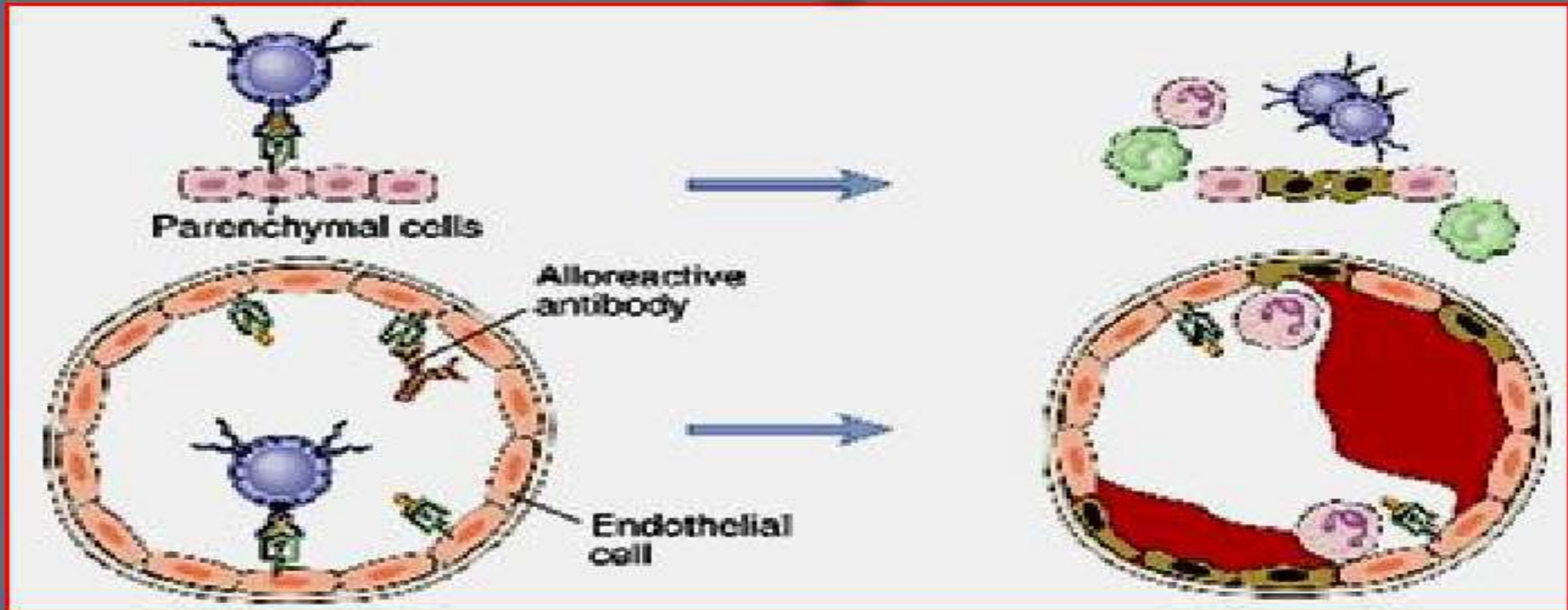
T-cell, macrophage and Ab mediated, myocyte and endothelial damage, Inflammation

Chronic Rejection :-

- Occurs in most solid organ transplants

- Heart, Kidney, Lung, Liver

- Characterized by :

a. Fibrosis

b. Vascular abnormalities

c. Loss of graft function over a prolonged period.

Chronic Rejection

Chronic rejection

Vascular smooth muscle cell

APC

Cytokines

Macrophage

Cytokines

Alloantigen-specific CD4⁻ T cell
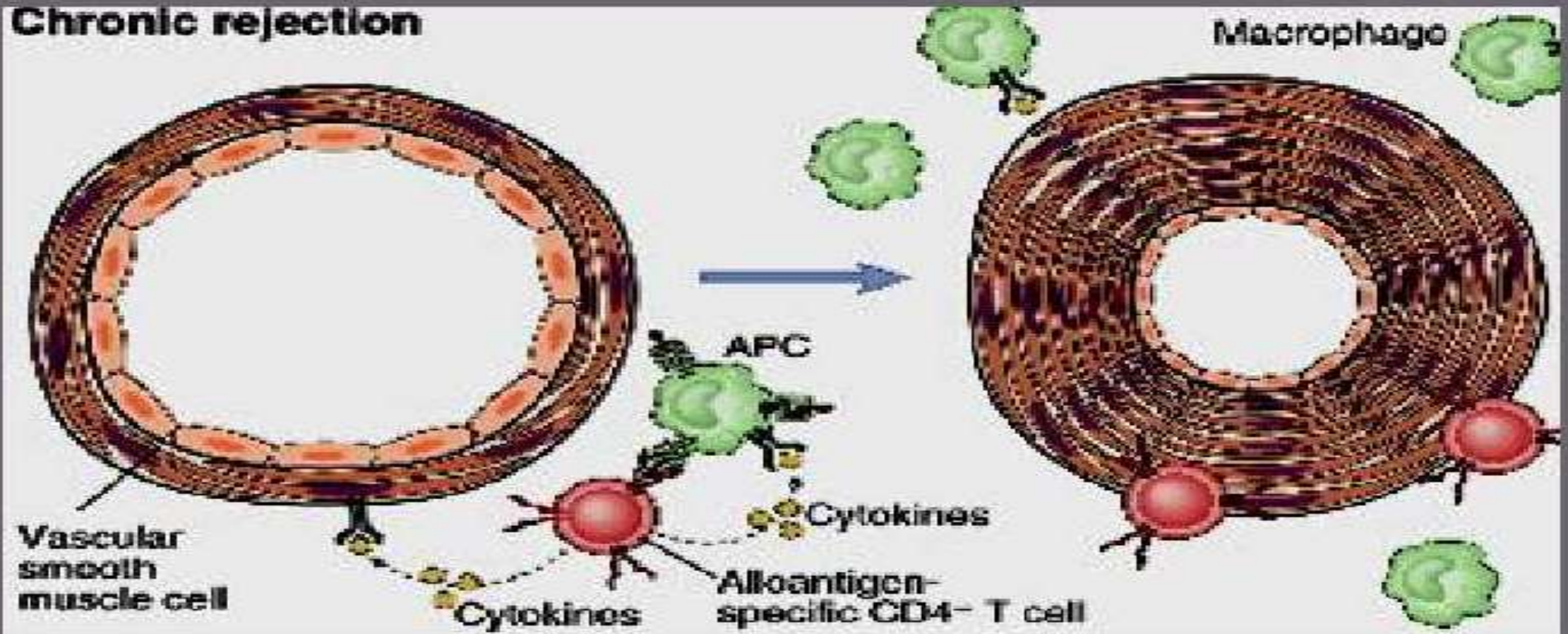
Macrophage – T cell mediated

Concentric medial hyperplasia

Chronic DTH reaction

Histocompatibility antigens.

Antigens that participate in graft rejection are called transplantation or histocompatibility antigens :

- ABO blood group

- HLA system (MHC restricted allograft Rejection)

Histocompatibility Testing :-

- ABO blood grouping

- HLA compatibility:

- Tested by HLA typing and tissue matching

- HLA typing identifies the HLA antigens expressed on the surface of leucocytes

Methods of HLA – Typing :-

- Microcytotoxicity   test .

- Molecular methods

a. PCR using sequence specific primers.

- Tissue matching .

can be employed to approximate the risk of a given recipient of having a positive crossmatch. This is to a likely organ donor taken from a similar population.

A Cell based antibody screening

Cells + Serum with no donor specific Ab

Cells + Serum with donor specific Ab

Wash

+ Complement and vital dye

B Solid phase antibody screening (bead based)

Beads + Serum with no donor specific Ab

Beads + Serum with donor specific Ab

Wash

+ Fluorescent conjugated anti-IgG

%Beads

Negative   Positive

Fluorescence →

Lymphocyte with HLA antigens on cell surface

HLA antibody

Membrane attack complex

Anti human globulin (AHG)

Bead with HLA antigens on cell surface

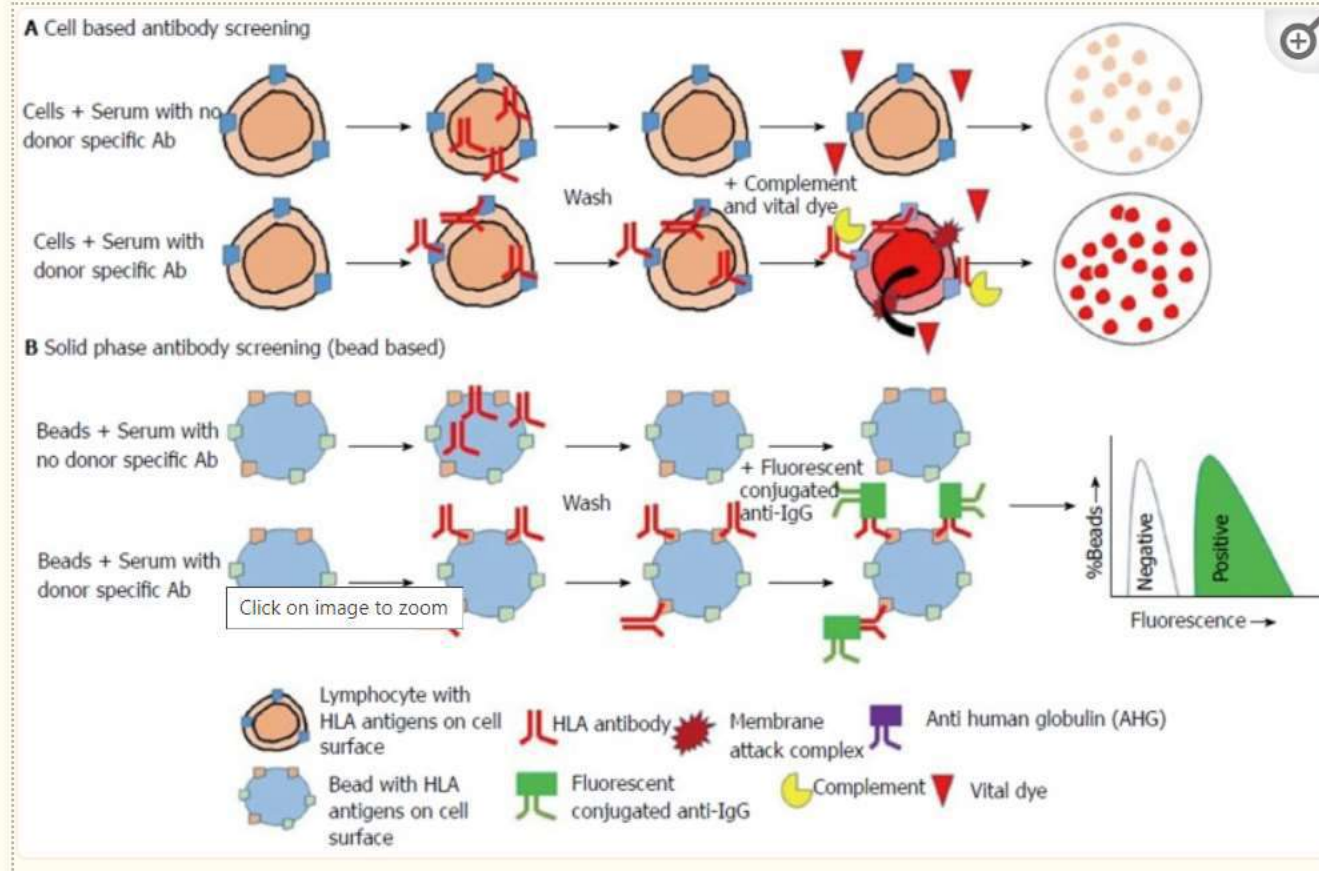Fluorescent conjugated anti-IgG

Complement

Vital dye

Figure 1

Schematic diagram of lymphocyte based antibody screening and solid phase (bead based) antibody screening. A: Cell based antibody screening; B: Solid phase (bead based) antibody screening.

Micro Cytotoxicity: -

Tests for complement mediated lysis of peripheral blood lymphocytes with a standard set of typing sera. Micro-cytotoxicity assay, utilizes serum with known anti-HLA antibodies that recognize particular HLA loci (HLA-A, HLA-B, HLA-C, HLA-DQ, HLA-DR /not DP) in order to match genetically similar individuals in hopes of performing a tissue transplantation.

Graft-versus-host (GVH) reaction: -

Graft rejection is due to the reaction of the host to the grafted tissue, Host-versus-graft response , The contrary situation, in which the graft mounts an immune response against the antigens of the host, is known as: Graft-versus-host (GVH) reaction.

Essential Component Required for (GVH)

The GVH reaction occurs when the following conditions are present:

1. The graft contains immunocompetent T cells.

2. The recipient possesses transplantation antigens that are absent in the graft.

3. The recipient must not reject the graft.

When grafted tissue has mature T cells, they will attack host tissue leading to GVHR. Major problem for bone marrow transplant.

Methods to overcome GVHR :

- Treat bone marrow to deplete T cells.

- Use autologous bone marrow.

- Use umbilical cord blood.

Caused by the reaction of grafted mature Tcells in the marrow inoculum with alloantigens of the host

- **Acute GVHD   :-** Characterized by epithelial cell death in the skin, GI tract, and liver .

- **Chronic GVHD :-** Characterized by atrophy and fibrosis of one or more of these same target organs as well as the lungs+
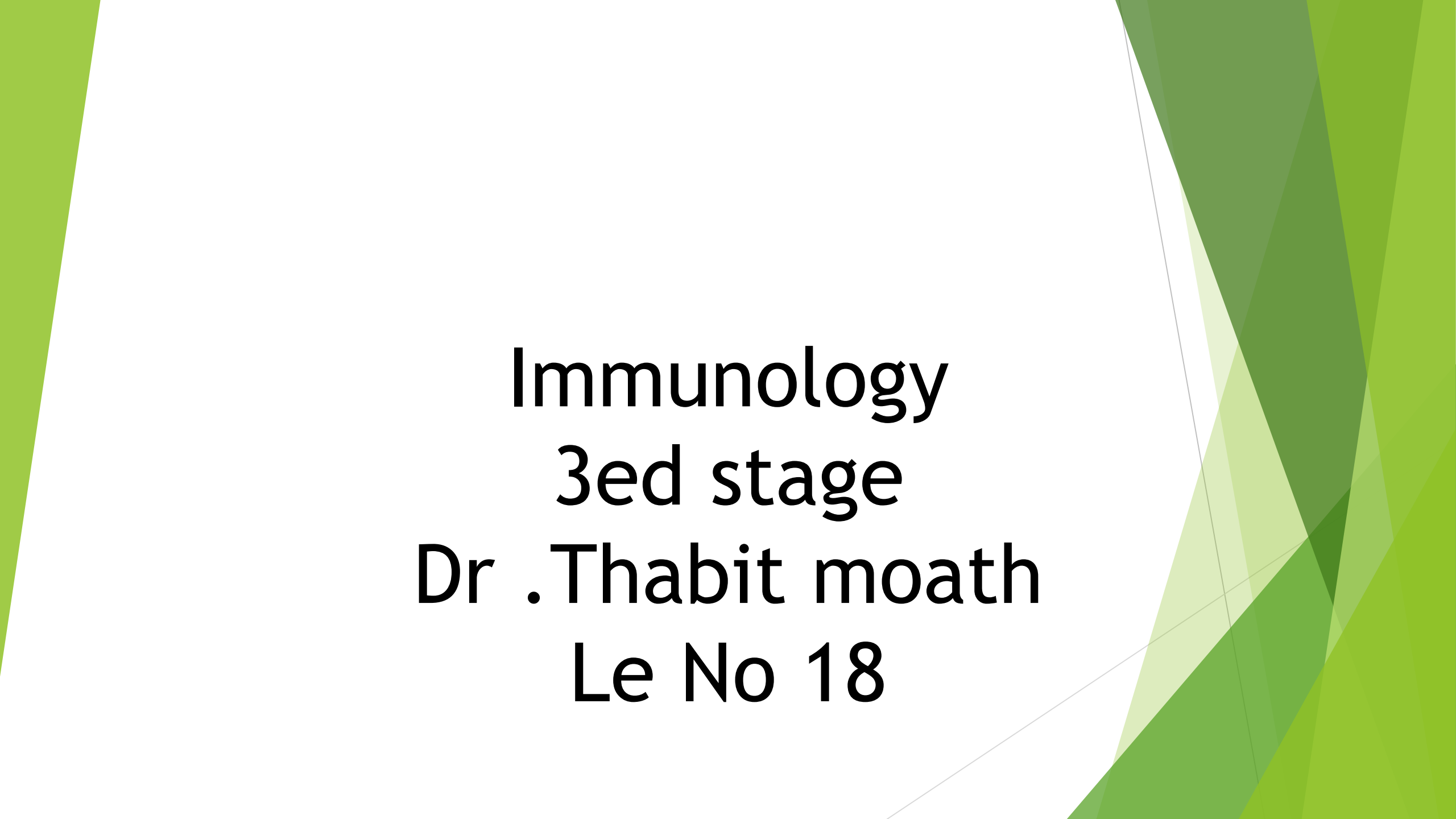
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Hypersensitivity

Hypersensitivity (also called hypersensitivity reaction or intolerance) is a set of **undesirable reactions produced by the normal immune system**, including allergies and autoimmunity. They are usually referred to as an over- reaction of the immune system and these reactions may be damaging, **uncomfortable**, or occasionally **fatal**. Hypersensitivity reactions require a **pre-sensitized (immune) state of the host**. They are classified in four groups after the proposal of P. G. H. Gell and Robin Coombs in 1963.
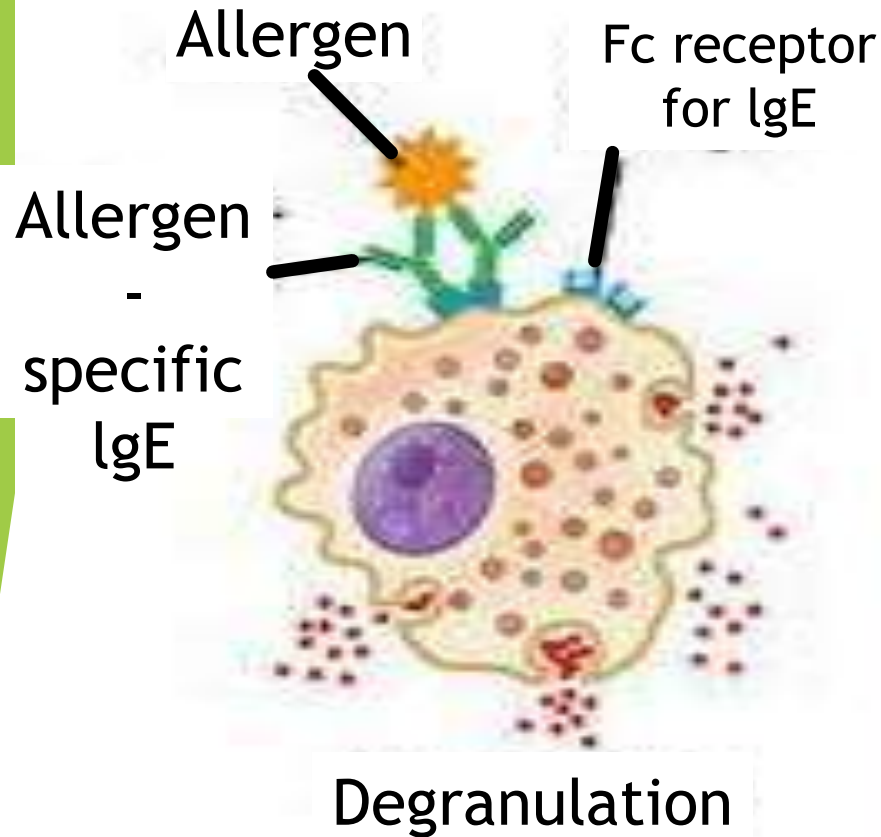
Type I: IgE mediated immediate reaction

**Type II: Antibody-mediated reaction (IgG or IgM antibodies)**

Type III: Immune complex-mediated reaction

Type IV: Cytotoxic, cell-mediated, delayed hypersensitivity reaction .

Type I

Allergen

Fc receptor for lgE

Allergen - specific lgE

Degranulation

- lgE-Mediated Hypersensitivity

- lgE is bound lo mast cells via its Fc portion. When an allergen binds to these antibodies. crosslinking of IgE induces degranulation.

- Causes localized and systemic anaphylaxis.
  seasonal allergies including hay fever.
  food allergies such as those to shellfish and   peanuts. hives. and eczema
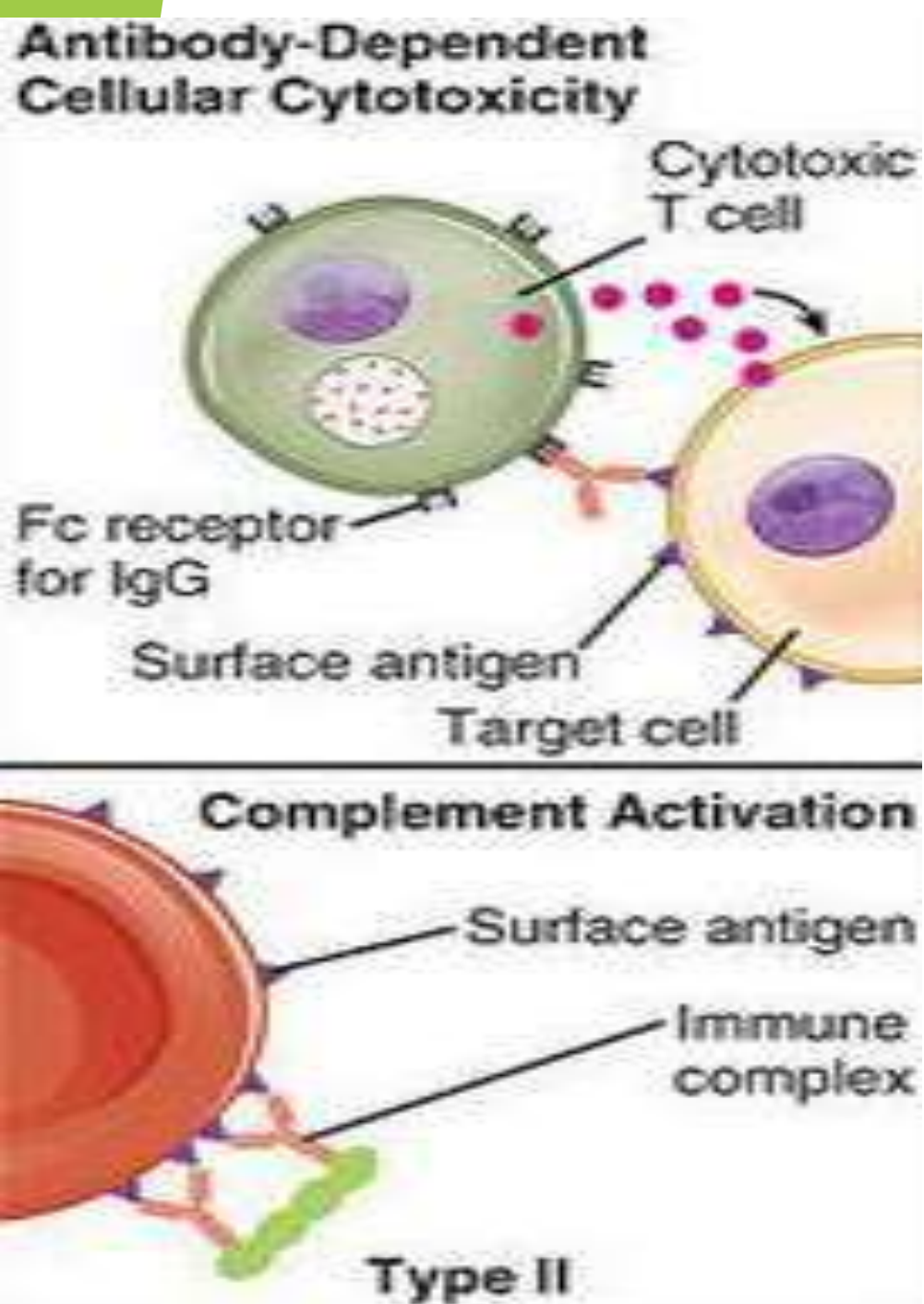
Type II

- lgG-Mediated Cytotoxic Hypersensitivity

- Cells are destroyed by bound antibody, either by activation at complement or by a cytotoxic T cell with an Fc receptor tor the antibody {ADCC}

- Red blood cells destroyed by complement and antibody during a transfusion ot mismatched blood type or during erythroblastosis fetalis

Antibody-Dependent Cellular Cytotoxicity

Cytotoxic T cell

Fc receptor for IgG

Surface antigen

Target cell

Complement Activation

Surface antigen

Immune complex

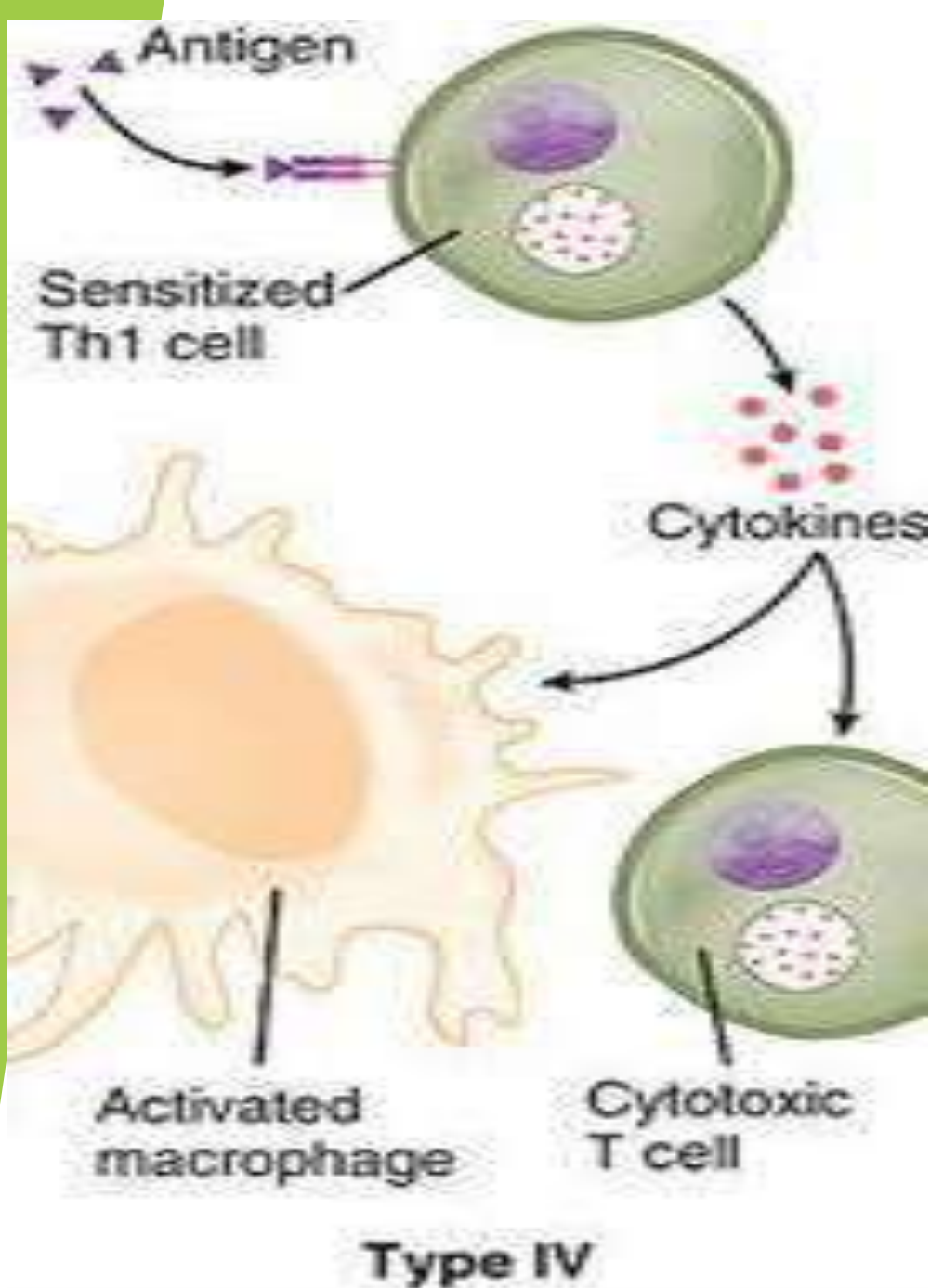Type II

Type III

Free-floating immune complex

Complement activation

Neutrophil

Type III

- Immune Complex-Mediated Hypersensitivity

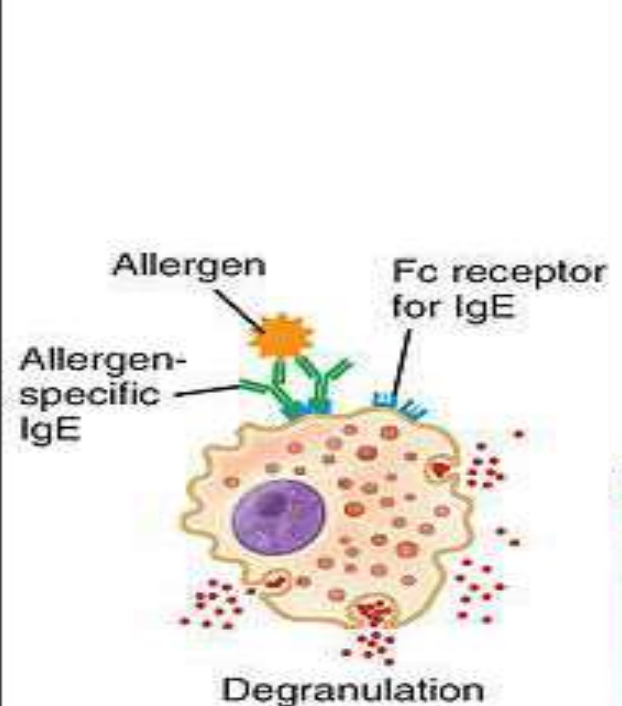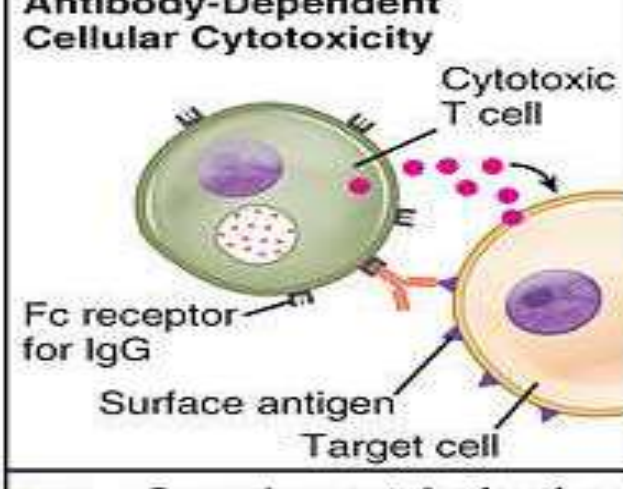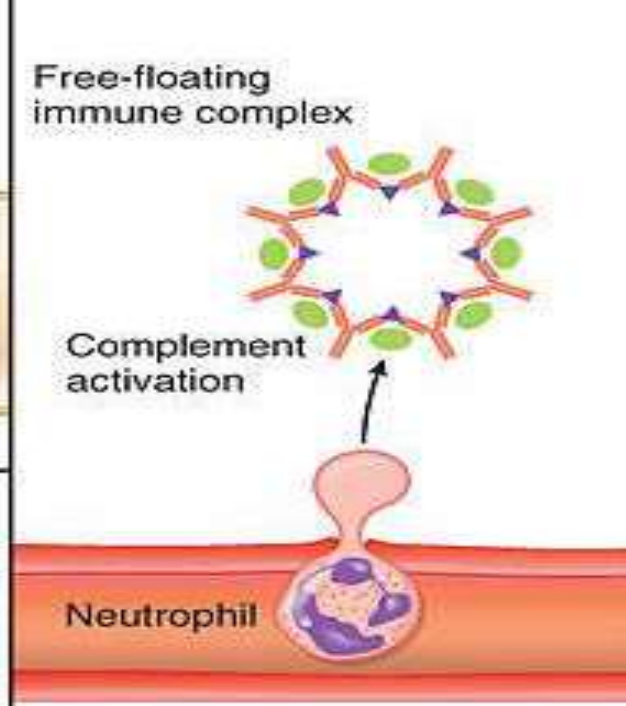Antigen-antibody complexes are deposited in tissues. causing activation at complement. which attracts neutrophils to the site

- Most common forms at immune complex disease are seen in glomerulonephritis. rheumatoid arthritis. And endemic lupus erythematosus

Type IV

Antigen
Sensitized Th1 cell
Cytokines
Activated macrophage
Cytotoxic T cell
Type IV

- Cell-Mediated Hypersensitivity

- Th1 cells secrete cytokines. which activate macrophages and cytotoxic T cells and can cause macrophage accumulation at the site

- Most common terms are contact dermatitis, tuberculin reaction, autoimmune diseases such as diabetes mellitus type I, multiple sclerosis, and rheumatoid arthritis

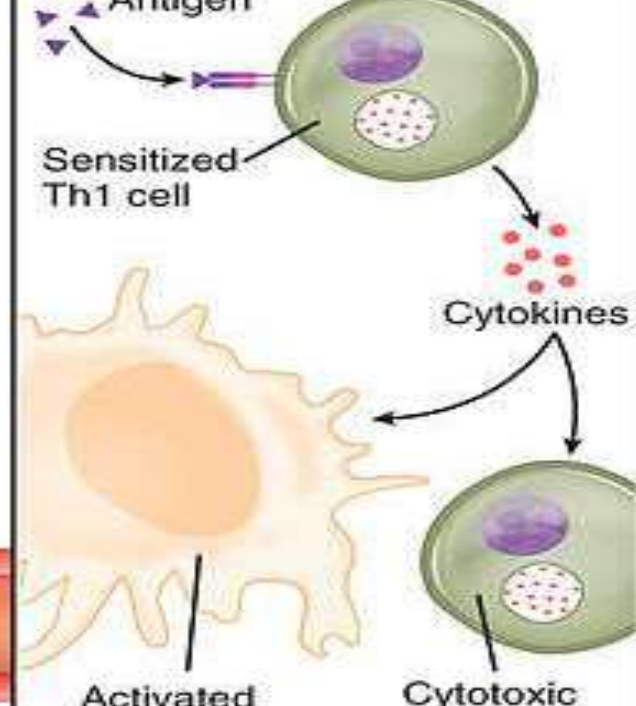| Type I	Type II	Type III	Type IV
IgE-Mediated Hypersensitivity	**IgG-Mediated Cytotoxic Hypersensitivity**	**Immune Complex-Mediated Hypersensitivity**	**Cell-Mediated Hypersensitivity**
IgE is bound to mast cells via its Fc portion. When an allergen binds to these antibodies, crosslinking of IgE induces degranulation.	Cells are destroyed by bound antibody, either by activation of complement or by a cytotoxic T cell with an Fc receptor for the antibody (ADCC)	Antigen–antibody complexes are deposited in tissues, causing activation of complement, which attracts neutrophils to the site	Th1 cells secrete cytokines, which activate macrophages and cytotoxic T cells and can cause macrophage accumulation at the site
Causes localized and systemic anaphylaxis, seasonal allergies including hay fever, food allergies such as those to shellfish and peanuts, hives, and eczema	Red blood cells destroyed by complement and antibody during a transfusion of mismatched blood type or during erythroblastosis fetalis	Most common forms of immune complex disease are seen in glomerulonephritis, rheumatoid arthritis, and systemic lupus erythematosus	Most common forms are contact dermatitis, tuberculin reaction, autoimmune diseases such as diabetes mellitus type I, multiple sclerosis, and rheumatoid arthritis

The antigen-**specific IgE** antibodies can then bind to high-affinity receptors located on the surfaces **of mast cells** and **basophils**. **Reexposure** to the antigen can then result in the antigen binding to and cross-linking the bound IgE antibodies on the mast cells and basophils. This causes the release and formation of chemical mediators from these cells.

The major mediators and their functions are described as follows:

1. **Histamine**:

2. **Tryptase**:

3. **Proteoglycans**:

4. Chemotactic factors:

Diagnosis of hypersensitivity

Laboratory testing may include:

1- Allergen-specific IgE blood testing: this is testing that is used to help diagnose allergies. The test measures the amount of **allergen-specific IgE antibodies in the blood** in order to detect an allergy to a particular substance.

Other types of allergy tests:

1. ***Skin prick or scratch tests*** are done in an allergist's or dermatologist's office and must be done by a trained professional. They are often used to detect airborne allergies such as pollens, dust, and molds. Because of the potential for a severe reaction, skin prick tests are not usually used for food allergies. The person being tested must not have significant eczema or be taking antihistamines or certain antidepressants for several days before the skin prick test. False positives can arise with even a non-allergic person if the dosage of the allergen is high enough.

Intradermal allergy skin tests, using injections that form a bubble under the skin, may be done but they are not widely accepted **because of a high false-positive** rate
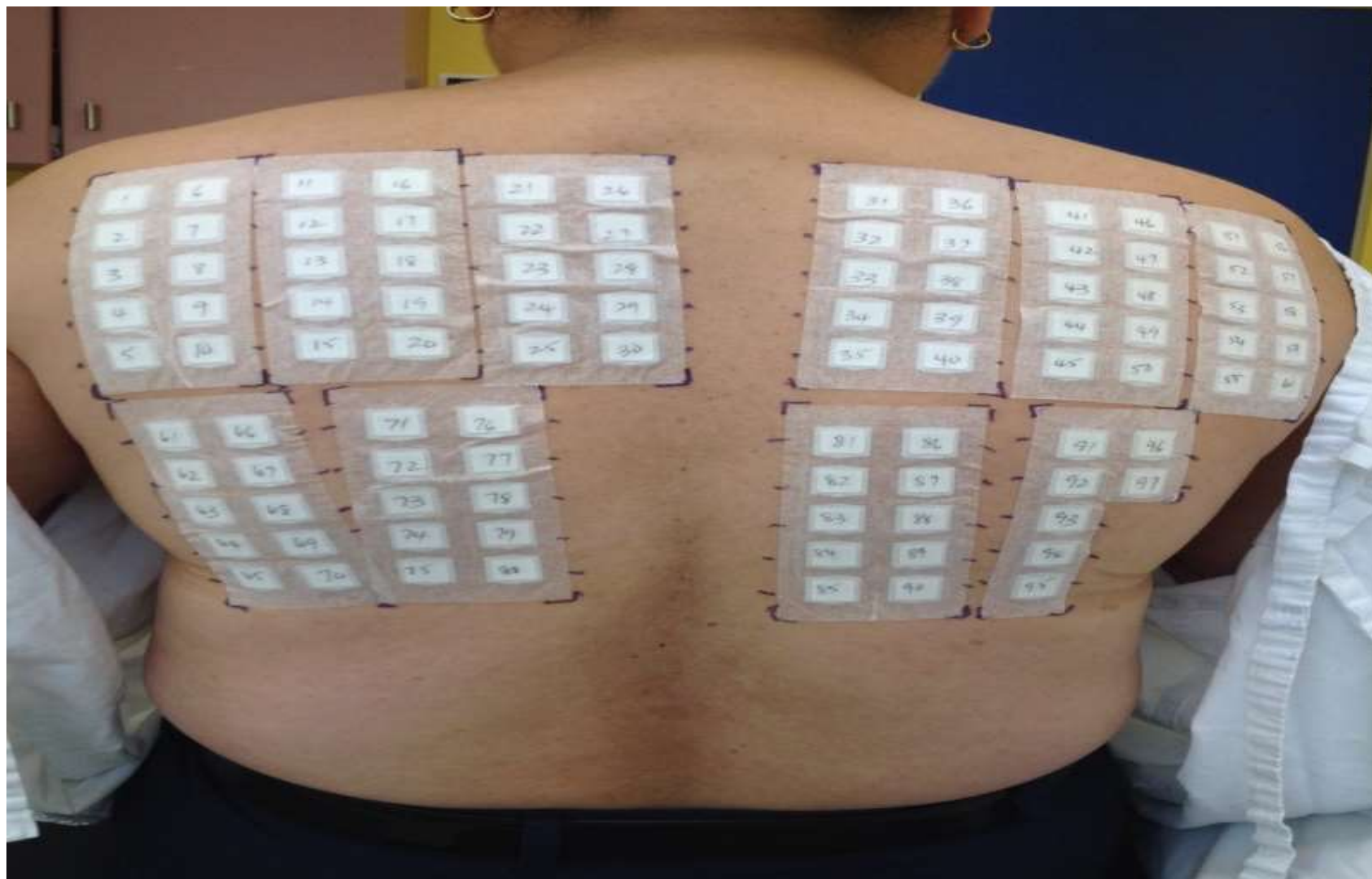
Patch testing:- Delayed hypersensitivity skin and patch tests are the easiest methods of testing for type IV delayed hypersensitivity. A concentration of the suspected allergen is applied to the skin under a nonabsorbent adhesive patch and left for 48 hours. If burning or itching develops more rapidly, the patch is removed. A positive test consists of redness with some hardening and swelling of the skin and sometimes vesicle (blister-like) formation. Some reactions will not appear until after the patches are removed, so the test sites are also checked at 72 and 96 hours
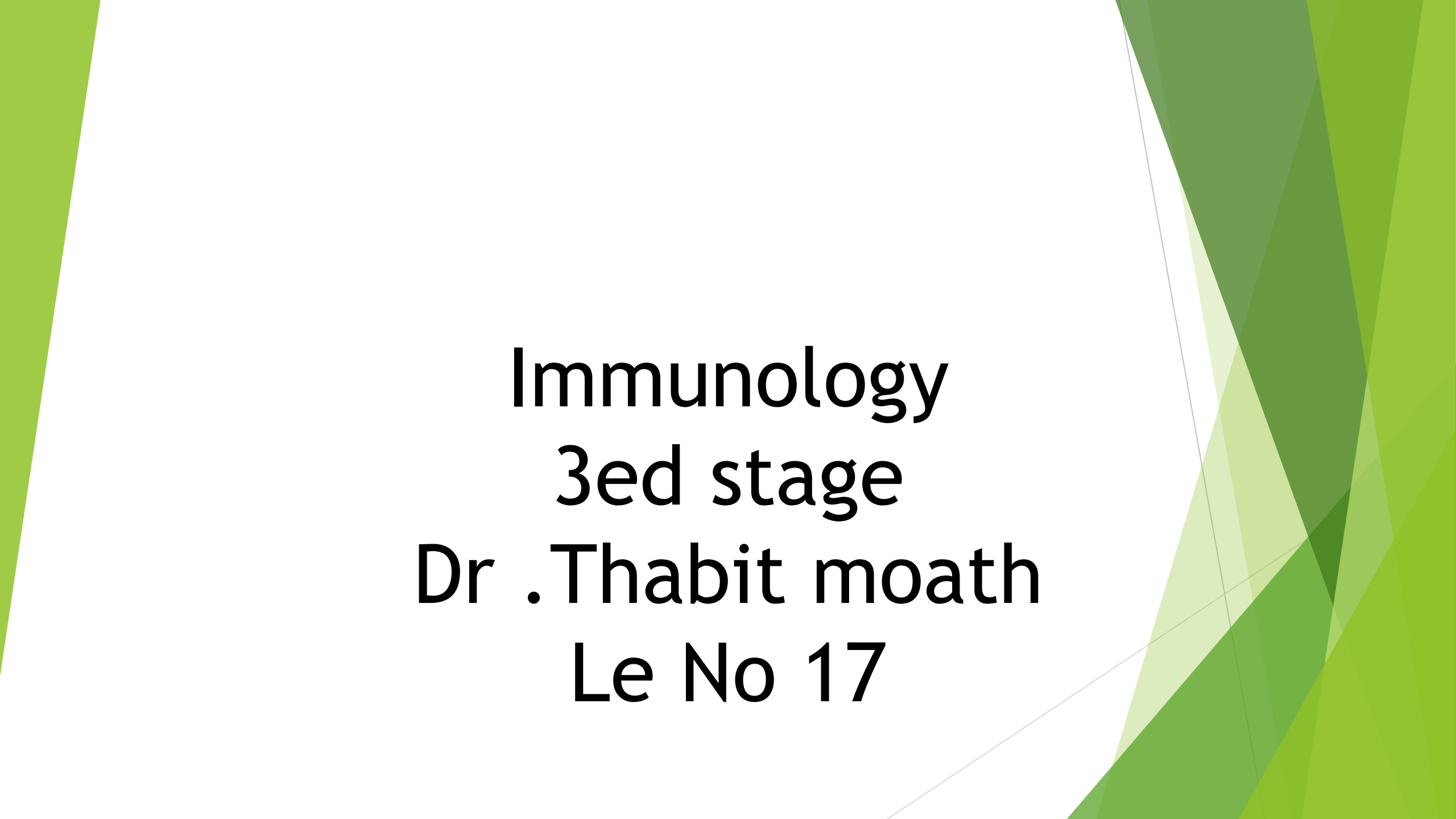
4. ***Oral food challenges*** **are** considered the "gold standard" for diagnosing food allergies. They are labor-intensive and require close medical supervision because reactions can

    be severe, including life-threatening anaphylaxis. Food challenges involve giving a person small amounts of unmarked potential food **allergens in capsule or intravenous** form and watching for allergic reactions. Negatives are confirmed with larger meal-sized portions of food.

4. *Food elimination* is another way to test for food allergies: eliminating all suspected foods from the diet, then reintroducing them one at a time to find out which one(s) are causing the problem.

2- **Total IgE testing** is sometimes done to look for an ongoing allergic process. It is a **blood test that detects the total amount of IgE protein** (including allergy antibodies) but does not identify specific allergens. Conditions besides allergies can also cause the IgE level to rise.

3- **Complete blood count (CBC) and WBC differential**—these tests include the measurement of eosinophils, a type of white blood cell. The level of eosinophils may be increased in a person with allergies.

4- **Histamine and/or tryptase blood tests** may be used to help diagnose anaphylaxis or mast cell activation.

Immunology
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Tumors and Tumor Markers

Tumor immunology

The proliferation of normal cells is carefully regulated. However, such cells when exposed to chemical carcinogens, irradiation and certain viruses may undergo mutations leading to their transformation into cells that are capable of uncontrolled growth, producing a tumor or neoplasm.

The tumor may be:

- Benign, if it is not capable of indefinite growth and the host survives.

- Malignant, if the tumor continues to grow indefinitely and spreads (metastases). This uncontrolled growth may be due to up regulation of **oncogenes** (cancer-inducing genes) and/or down regulation of **tumor suppressor genes** (that normally inhibit tumor growth often by inducing cell death).

Tumor associated antigens

There are 2 main types of tumor antigens:

- Tumor-**specific** transplantation antigens (TSTA) which are **unique to tumor cells and not expressed on normal cells.**

- Tumor **associated** transplantation antigens (TATA) that are expressed by **tumor cells and normal cells.**

Tumor-associated developmental antigens or onco-fetal antigens:-

These include **alpha-fetoprotein (AFP)** and **carcino-embryonic antigen (CEA)** found secreted in the serum. AFP is found in patients with **hepatocellular carcinoma** whereas CEA is found in **colon cancer**. These are important in diagnosis. **AFP is produced only as a secreted protein whereas CEA is found both on cell membranes and in secreted fluids.**

Viruses that cause human tumors include:

DNA viruses

- Papova (papilloma, polyoma) viruses: Papilloma virus causes cervical cancer.

- Hepatitis virus: Hepatitis B virus causes hepatocellular cancer.

- Adenoviruses may also be tumorigenic

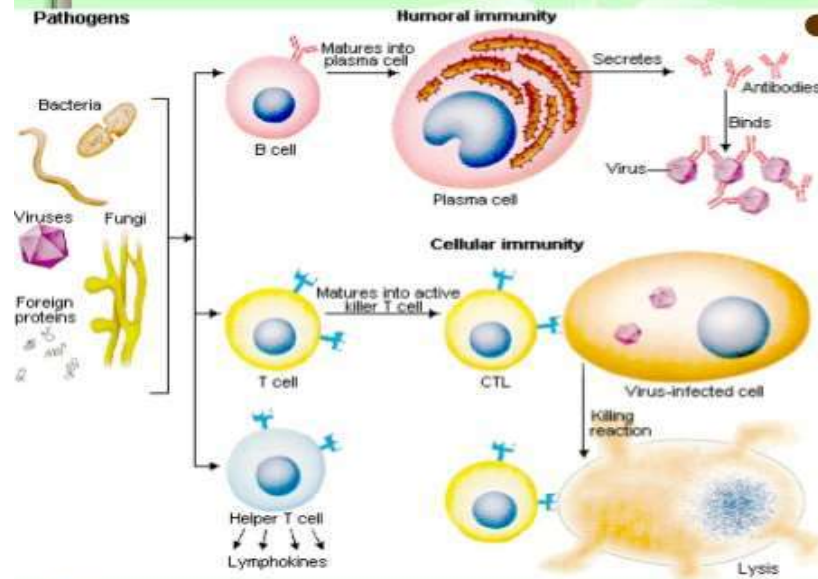- Certain types of Herpes Virus (CMV and EBV)

RNA viruses

- Retroviruses: Human T- lymphotropic viruses (HTLV-I and HTLV-II) causes T-cell leukemias.

Immunity against Tumor : There is several anti-tumor immune reactivity in human;

1. **Cell-mediated immunity** plays a critical role in **tumor rejection.**

2. <u>**The T helper (Th)** cells recognize the tumor antigens that may be shed from tumors and internalized, processed and presented in association with class II MHC on antigen presenting cells. These Th cells, will produce cytokines.   provide help to B cells in antibody production.</u>

3. **Cytokines such as IFN-gamma** γ may also activate **macrophages** to be tumoricidal

4. The Th cells also provide help to **tumor-specific cytotoxic T cells (CTLs) CD8$^+$** and **NK cell** by inducing their proliferation and differentiation.

5. **The CTLs recognize tumor antigens** in the context of class **I MHC** and mediate tumor cell lysis.

6. <u>In tumors with decreased MHC antigens, natural killer (NK) cells are important in mediating tumor rejection.</u>

Immune Response to Tumours

Pathogens

Humoral immunity

Bacteria

B cell → Matures into plasma cell → Plasma cell → Secretes → Antibodies → Binds → Virus

Viruses Fungi

Foreign proteins

Cellular immunity

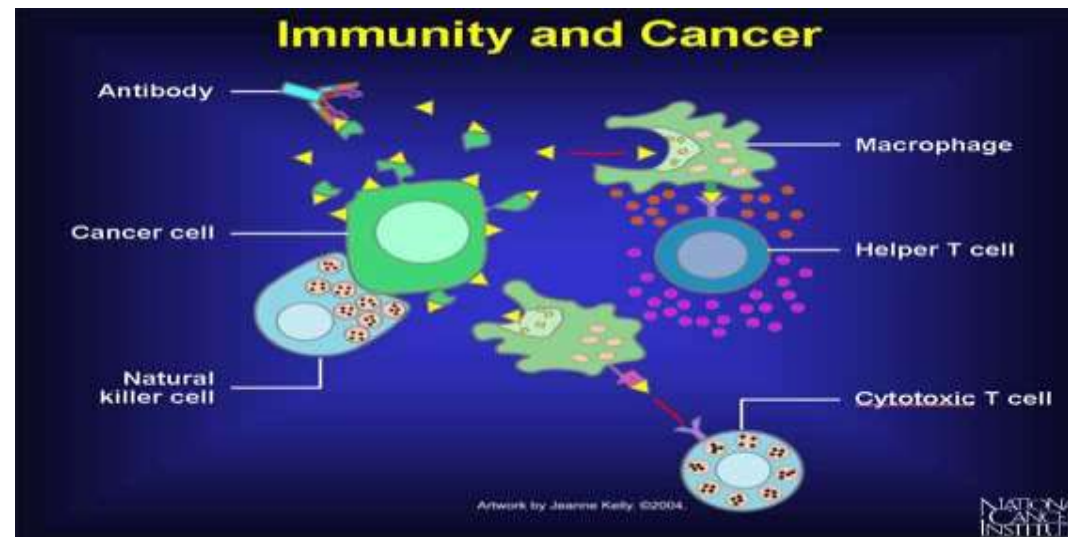T cell → Matures into active killer T cell → CTL → Virus-infected cell → Killing reaction → Lysis

Helper T cell → Lymphokines

- The immune response to tumors includes
  - CTL-mediated lysis,
  - NK-cell activity,
  - macrophage-mediated tumor destruction, and
  - destruction mediated by ADCC.

Taradi

Interactive immunology   tumor immunology   41

Tumor Marker (TM):-

T.M or also known as biomarkers are indicators of cellular, biochemical, molecular, or genetic alterations by which neoplasia can be recognized. It is measurable biochemical that are associated with a malignancy, **they are substances found at higher than normal levels in some peoples with cancer.**

Classification:- The T.M. is either produced by the tumor cells (tumor derived) or by the body in response to tumor cells (Tumor associated).

1. **Tumor Specific Antigens**: - Present on tumor cells and not on any normal cells.
2. **Tumor Associated Antigens: -** Are present on tumor cells and also on some normal cells in response to the tumor.

Clinically associated useful of TMs :-

• Diagnostic and distinguish benign from malignant disease

• Correlate with the amount of tumor present (so-called tumor burden)

• Allow subtype classification to more accurately stage patients

• Be prognostic, either by the presence or absence of the marker or by its concentration

• Guide choice of therapy and predict response to therapy

<u>Type of T.Ms. (specific and /or sensitive)</u>

As like other laboratory Testing, T.Ms. test must be both **specific and sensitive.**

Specificity: - If either the T.Ms. it self or the test used to detect or measure it is not specific enough, there is a chance that the results could suggest a tumor is presents, or growing despite treatment, when it is not (a false positive). High specificity – not present in other diseases, non-tumors and in healthy subjects

Sensitivity: - If the T.Ms. or the test is not sensitive enough, the result may suggest a tumor is not present when it actually is or that is responding to treatment, when it is not (A false Negative). High sensitivity – detectable at the beginning of the disease

Main Examples of T.Ms. in cancers:-

Protein Tumor Markers :

- Carcinoembryonic antigen (CEA)
- α- Fetoprotein (AFP) .
- Carbohydrate Antigen (CA . 19-9) . (CA . 125). monitoring of ovarian CA
- Prostate-Specific Antigen (PSA) .
- CA 15-3 – monitoring of breast CA
- CA 72 CA 72-4– monitoring of gastric CA
- CA 19 CA 19-9 9 for monitoring of pancreas CA (and bile ducts)
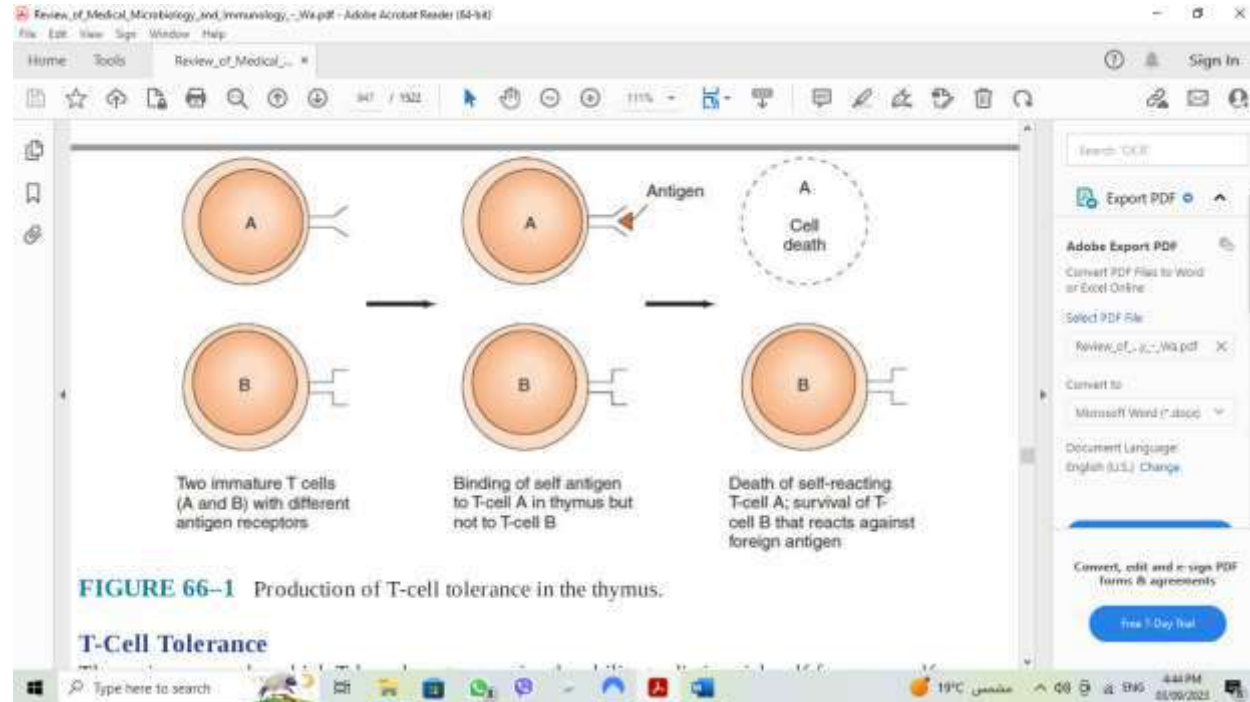- Human Chorionic Gonadotropin in Testicular Germ Cell Tumors

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Tolerance is **specific immunologic unresponsiveness** (i.e., an immune response to a certain antigen does not occur, although the immune system is otherwise functioning normally.
 In general, antigens that are present during embryonic life are considered "self" and **do not stimulate** an immunologic response (i.e., we are tolerant to those antigens). The lack of an immune response in the fetus is caused by the **deletion of self-reactive T-cell precursors** in the thymus. On the other hand, antigens that are not present during the process of maturation (i.e., that are encountered first when the body is immunologically mature) are considered "nonself" and usually elicit an immunologic response. **Although both B cells and T cells participate in tolerance, it is T-cell tolerance that plays the primary role**

Two immature T cells (A and B) with different antigen receptors

Binding of self antigen to T-cell A in thymus but not to T-cell B

Death of self-reacting T-cell A; survival of T-cell B that reacts against foreign antigen

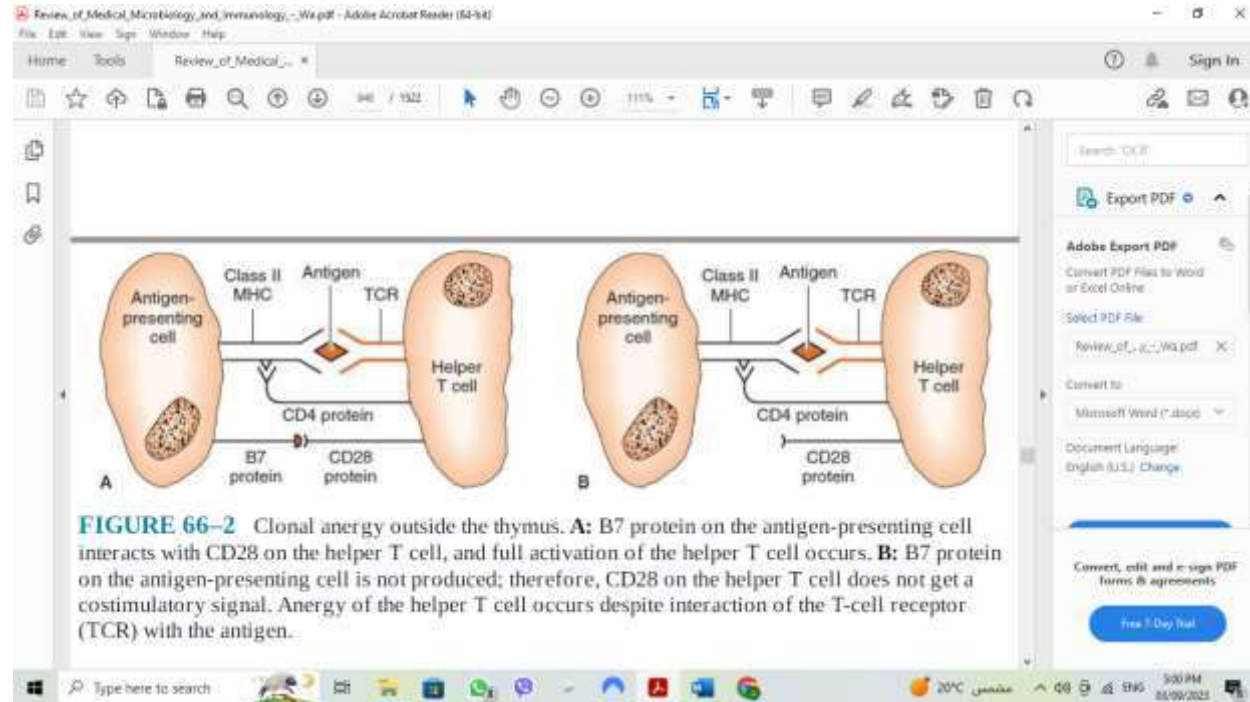**FIGURE 66–1** Production of T-cell tolerance in the thymus.

T-Cell Tolerance

- The main process by which T lymphocytes acquire the ability to distinguish self from no self occurs in the fetal thymus This process, **called clonal deletion**
- involves the killing of Tcells ("**negative selection**") that react against antigens present in the fetus at that time.
- The self-reactive cells die by a process of programmed cell death called **apoptosis**.
- Tolerance to self acquired within the thymus is called **central tolerance,** whereas tolerance acquired outside the thymus is called **peripheral tolerance**.
- Peripheral tolerance is necessary because some antigens are not expressed in the thymus and therefore some self-reactive T cells are not killed in the thymus.

mechanisms involved in peripheral tolerance:

1. Some self-reactive T cells are killed, some are not activated, and others are **suppressed by regulatory T cells** producing inhibitory cytokines.
2. **Clonal anergy** is the term used to describe self-reactive T cells that are not activated because proper costimulation does not occur Clonal anergy outside the thymus. **A: B7 protein on the antigen-presenting cell** interacts **with CD28 on the helper T cell,** and full activation of the helper T cell occurs. **B:** B7 protein on the antigen-presenting cell is not produced; therefore, CD28 on the helper T cell does not get costimulatory signal. Anergy of the helper T cell occurs despite interaction of the T-cell receptor (TCR) with the antigen.

FIGURE 66–2 Clonal anergy outside the thymus. **A:** B7 protein on the antigen-presenting cell interacts with CD28 on the helper T cell, and full activation of the helper T cell occurs. **B:** B7 protein on the antigen-presenting cell is not produced; therefore, CD28 on the helper T cell does not get a costimulatory signal. Anergy of the helper T cell occurs despite interaction of the T-cell receptor (TCR) with the antigen.
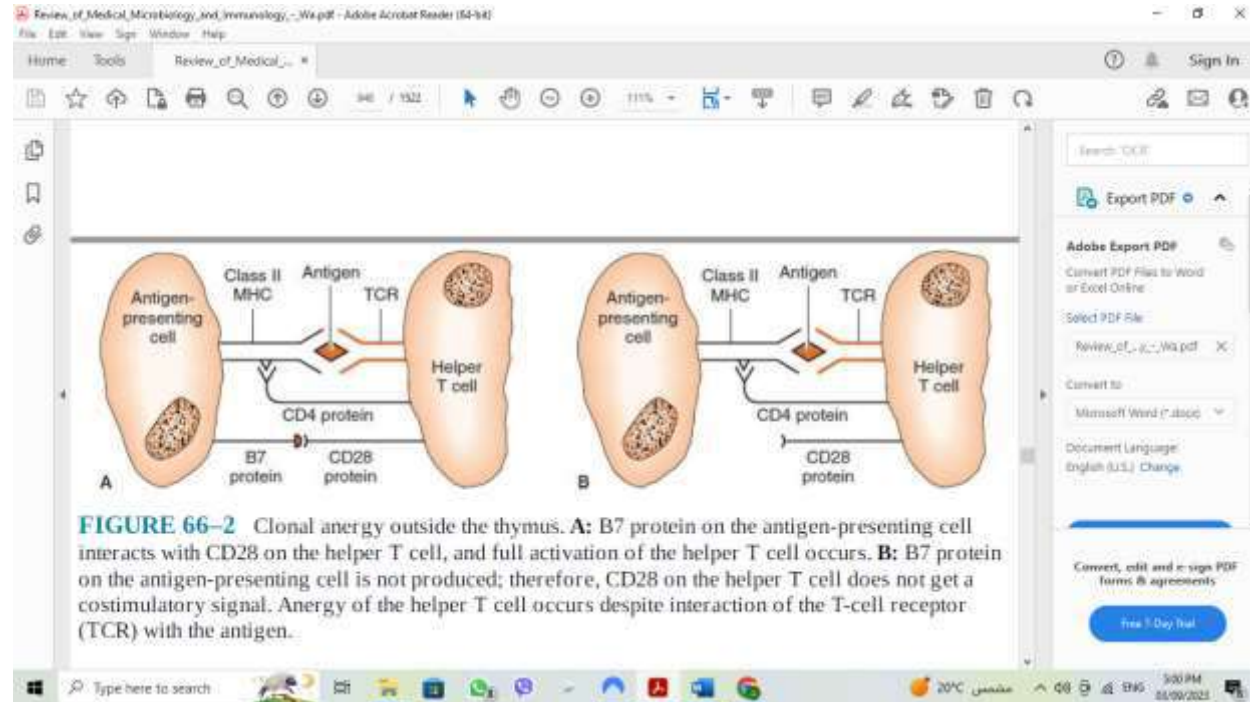
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Clonal ignorance refers to self-reactive T cells that ignore self antigens. These self-reactive T cells are either kept ignorant by physical separation from the target antigens (e.g., the blood–brain barrier) or ignore self antigens because the antigens are present in such small amounts
Although T cells that are clonally anergic are nonreactive, they can become reactive and initiate an autoimmune disease if conditions change later in life

B-Cell Tolerance

B cells also become tolerant to self by two mechanisms:

(1) clonal deletion, probably while the B-cell precursors are in the bone marrow,

(2) clonal anergy of B cells in the periphery.

However, tolerance in B cells is less complete than in T cells, an observation supported by the finding that most autoimmune diseases are mediated by antibodies.

B cells bearing an antigen receptor for a self protein can escape clonal deletion (apoptosis) by a process called **receptor editing**
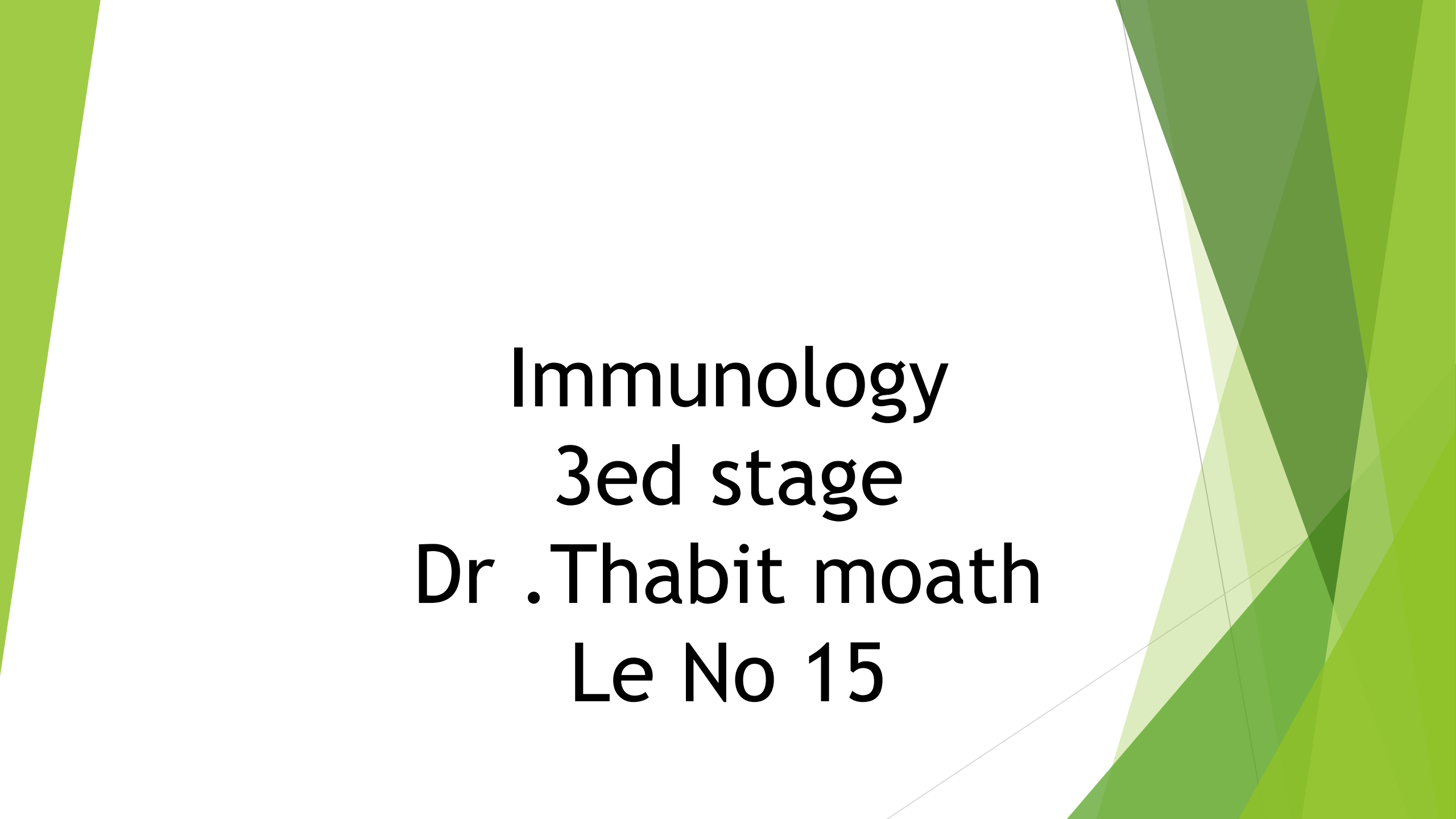
INDUCTION OF TOLERANCE

Whether an antigen will induce tolerance rather than an immunologic response is largely determined by the following:

(1) The immunologic **maturity** of the host (e.g., neonatal animals are immunologically immature and do not respond well to foreign antigens; for instance, neonates will accept allografts that would be rejected by mature animals).

(2) The **structure** and **dose** of the antigen (e.g., a very simple molecule induces tolerance more readily than a complex one, and very high or very low doses of antigen may result in tolerance instead of an immune response). Purified polysaccharides or amino acid copolymers injected in very large doses result in "immune paralysis"—a lack of response.

Other aspects of the induction or maintenance of tolerance are as follows:

(1) T cells become tolerant more readily and remain tolerant longer than B cells.

(2) Administration of a cross-reacting antigen tends to terminate tolerance.

(3) **Administration of immunosuppressive drugs enhances tolerance (e.g., in patients who have received organ transplants).**

(4) Tolerance is maintained best if the antigen to which the immune system is tolerant continues to be present.

Immunology
3ed stage
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Le No 15

I.   **Viral infections:**

Are Obligatory intercellular pathogens that replicate within nucleic acid and protein synthetic machineries of the host cell, it able to infect a variety of cells by utilizing normal cell surface molecules as receptors to enter cell.

- **Innate immunity to viral infections is largely through:**

1. **Produce of antiviral proteins such as interferons (e.g. IFN-α, IFN-β) that lead resistance to viral replication**

2. **NK cell activation.**

- **Adaptive immune response to viral infection through:**

1. **Block the virus binding and entry into host cells by neutralization antibody.**

2. **Antibody or complement opsonize viral particles and promote clearance by phagocytosis.**

3. **CTL eliminates the infection by killing infected cells.**

Viruses have developed various ways to evade immune responses, include:

1. Evading the action of IFNs.

2. Inhibition of antigen presentation: By down-regulating/reduce of MHC expression, and inhibiting transporter molecule needed for antigen processing.

3. Interfering with complement function: By produces C3, C4b-binding proteins that inhibiting both the classical and alternative pathways.

4. Constant alteration in viral antigens: By antigenic drift/shift (point mutation) that prevents a secondary immune response from developing.

5. Modulation of viral antigen from the surface of infected cells by capping and shedding following interaction with virus-specific antibody allows virus-infected cells to escape ADCC by NK cells.

6. Suppression of antiviral immune responses: by activating regulatory T cells or destroying essential lymphoid tissues (e.g. HIV).

Host resistant to infectious pathogens:

Parasitic infections (protozoa or helminthes):

I. Parasitic infections (protozoa or helminthes):

Parasites are uni/multicellular eukaryotic pathogens, and broadly comprise:

(1) **Protozoa** may live in the gut e.g. amoebae, in the blood e.g. African trypanosomes, inside erythrocytes e.g. *Plasmodium* spp., within macrophages of the liver and/or spleen (e.g. *Leishmania* spp.), or in muscle (e.g. *Trypanosoma cruzi*).

(2) **Helminthes or worms** may infect many organs of human as trematodes or flukes e.g. *Schistosomes* live in blood vessels, cestodes e.g. tapeworms and nematodes or roundworms (e.g. *Trichinella spiralis*, hookworms, pinworms, *Ascaris* spp. inhibit the gut and filarial worms may live in lymphatic tissues).

(3) **Arthropods** include Lice, mites, ticks and other arthropods may infect human. These parasites complete part or all of their life cycle within human.

Like other pathogens, parasites must highly survive from potent immune response because; they possess great diversity of strategies for avoiding immune detection, suppressing cellular immunity and deflecting immune attack mechanisms by of the following:

✓ **Antigenic variation onto parasitic surfaces mechanism:**

It depends on variants of antigens on parasite surface:

1. The African *trypanosome* and *Plasmodium* of malaria have able to **antigenic variation** **t**hat protects them from host defense.

2. *Schistosoma mansoni* acquire **surface layer from host that does not distinguish them,** which promote parasite to travels through the host. *Schitosoma spp.* have tough tegument, *Entamoeba* and (*Trichinella spiralis*)

1. **surround them with protective cysts** which protects them from defenses.

- ✓ **Seclusion intracellularly mechanism**

  It depends on hiding of parasite inside cell that avoid antibody and complement to activates and avoid destruction or killed them by **oxygen metabolites and lysosomal enzymes:**

- ☻ *Leishmania* **sp.** activates complement binds, opsonization, phagocytosed by macrophage, enclosed in the phagolysosome vacuoles, but ability to **produce antioxidant enzymes that inhibitors of lysosomal enzymes.**

- ☻ *T. gondii* are taken up by phagocytes and **prevents phagosome from fusing with lysosome**.

- ☻ *Trypanosoma cruzi* (Chagas' disease) **abnormal phagocytosis** caused by lysosomes to cluster and enters directly.

- ✓ :

✓ **Molecules that inhibit nonspecific host effector:**

Its depend onto parasitic produce antioxidant enzymes against toxic substance produced by activated immune cells as hydrogen peroxide, superoxide ions, hydroxyl radicals:

🕐 Oxygen scavenging enzymes to protect themselves.

🕐 Major enzymes are superoxide dismutase, catalase, and peroxidase.

All protozoan and helminthes parasites may contain at least one of these **antioxidant enzymes.**

✓ **Molecules that inhibit Complement activation:**

- *Leishmania* **avoid membrane attack complex (MAC)** <u>by having surface proteins, that prevent allow of MAC formation</u> because they are too far away from the membrane surface.

- *T. cruzi* express a molecule (DAF) that **prevents assembly of the complement cascade,** which lead no opsonization by complement and no MAC formation.

- ***Taenia solium* produces paramyosin** that **blocks assembly of the complement** cascade which lead no opsonization by complement and no MAC formation.

- ✓ **Host immunosuppression:**

- *Trypanosoma cruzi, T. gondii,* and *Leishmania* **induce expression of TGF-β :Transforming growth factor beta, that down regulates both TH1 and TH2 responses.**

- *T. gondii* and *Leishmania* **enters macrophage without inducing IL-12 that means noTH1 response.**

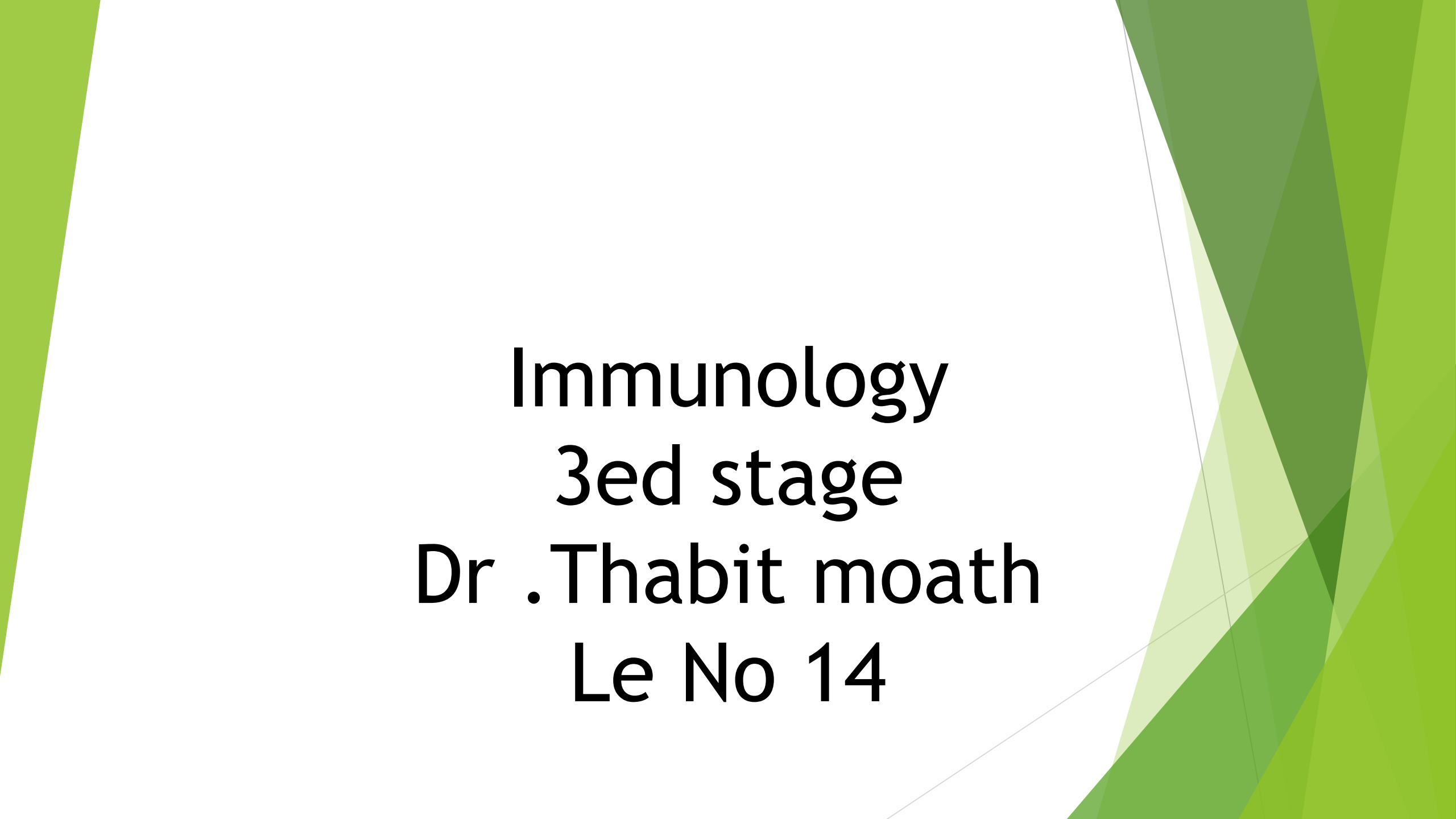- *Shistosoma* eggs **induce IL-10 that diminishes IFN-γ mediated macrophages activation.**

Immune response against parasitic infection:

⏰ **Macrophages activation by Th1 cytokines.**

⏰ **Activation of Th2 cells, which results in production of IgE antibodies and activation of eosinophil.**

⏰ **Eosinophil may be more effective at killing helminthes than other leukocytes; because the major basic protein of granules may be more toxic.**

I. **Fungal infections:**

Fungi are eukaryotic organisms that live on dead organic material, like bacteria, most are harmless; some may cause disease in humans which is called mycoses that tend to cause serious infections primarily in individuals with impaired immunity.

Most fungal infections are accidental and originate from an exogenous source by inhalation (*Aspergillus* spp., *Cryptococcus* or endemic mycoses), the gastrointestinal tract for commensals (*Candida* spp.) or reactivation of a latent infection.

Immunology
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Host resistance to infectious diseases:

The main function of immune system is protected by developed various mechanisms to invading pathogens that causes damage and harmful "selfish" relationship with the host.

Different pathogenic agents like bacteria, virus, fungi, parasite, etc. may enable to cause infectious diseases by invasion and establishment of an infection by overcoming onto innate immunity (Surface/mechanical defenses, phagocytic/inflammatory defenses), and adaptive immunity (specific humoral and/or cell-mediated).

immune response factors(as ageneral)

1.Routes and sites of infection,

2.Mechanisms of tissue injury,

3.Timing of immune responses,

4.Regulation of cellular/humoral immunity into infections,

5. Microbial evasion of immune responses,

6. immunizations/vaccines.

For these reasons different

Defense mechanisms

in human may be established against different infections which include:

I. **Bacterial infection:**

Immunity to bacterial infection is achieved by

antibody, **complement, opsonization** and **phagocytosis**.

The relative efficacies of these processes depend onto types of bacterial infection:

A. **Extracellular bacterial infections:**

Bacteria invade the body through mucosal surfaces (e.g. airway, gastrointestinal and genitourinary tracts) or break in the skin and causes disease by two principle mechanisms:

1. Induce inflammation (resulting in tissue destruction at the site of infection).
2. Produce toxins (Endo/Exotoxins) which are components of bacterial cell walls.

Bacterial infection occurs in 4 steps include:

1. Attachment to host cells.

2. Proliferation.

3. Invasion of host tissue.

4. Toxin-induced damage to host cells.

Many strategies used by extracellular bacteria to avoid from immune defenses during infection steps that were showing in table (1):

Infection process	Mechanism of pathogenic resistance
Attachment to host	Inhibit IgA secretion
Proliferation	- Inhibit phagocytic - Induction of apoptosis in macrophages. - Resistance to complement mediated lysis.
Invasion of host tissues	Secretion of elastase that inactivates C3a and C5a.
Toxin-induced damage to host cells.	Secretion of hyaluronidase which enhances bacterial invasiveness.

Immune mechanism to extracellular bacteria is achieved through:

Major mediators of immunity to extracellular bacterial infections are achieved through produce of **antibodies** as a humoral response and ability it's to:

- Provide protective immunity by killing live organisms through:

a. Opsonophagocytosis.

b. Promotes of complement mediated phagocytosis.

c. Neutralizing bacterial toxins to prevent damage and intense damaging inflammatory responses.

d. Blocking attachment to epithelial.

A. **Intracellular bacterial infections:**

Is ability of bacteria to colonize, survive and even replicate within phagocytes where they are inaccessible to circulating antibodies that occurs through:

1. Inhibiting lysosome fusion (e.g. *Mycobacteria* sp.).
2. Resisting oxidative/lysosomal attack (e.g. *Mycobacteria* sp.).
3. Escaping the phagosome following phagocytosis (e.g. *Listeria monocytogenes*).

Immune mechanisms to intracellular bacteria are achieved through:

1. TH1 cells, CTL and NK cells release IFN-γ that activates macrophages, increasing phagocytic activity, killing of phagocytized microbes, and MHC class II expression and antigen presentation.
2. CTL activated by IL-2 (released by TH1 cells) to kill infected macrophages, releasing bacteria that are then phagocytized and killed by activated uninfected macrophages * This type of immune response is called **Delayed type hypersensitive response**.

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Le No 13

Complement System

Complement: is a complex series of serum proteins (over than 30 protein), membrane- bound proteins, and complement receptors that play a major part in amplifying the inflammatory response to destroy and clear foreign antigens. **Complement system** capable of activating and destroying with no specificity a wide variety of bacteria, fungi, protozoans, viruses, tumor cells. Complement system are synthesized in **the liver and small and large intestine**. During fetal life, complement proteins are made from the sixth week.

The complement proteins are as follows:

1. Complement's components designated by letter C (C1–C9)

2. Subcomponents of the 1C (C1q, C1r, and C1s)

3. Fragments of the activated complement such as C2a, C3a, C3b, etc.

4.Factors of the alternative pathway (B, D, and P)

5.Enzymes like convertases

6.Membrane attack complex C5bC6789 (MAC)

7.Complement inhibitors and activators

8.Complement receptors

From a clinical point of view, deficiencies of the complement components are associated with high susceptibility to pyogenic infections, especially *Neisseria meningitidis*. Besides, defects of C1, C2, and C4–C8 may lead to autoimmune disorders like systemic lupus erythematosus (SLE).

The consequences of the complement protein cascade and formation of membrane attack complex (MAC) leads to the lysis of target cells, amplification of inflammation and phagocytosis, and the participation in completing soluble immune complexes.

Pathways of the Complement System

1. **Classical pathway**

2. **Alternative pathway**

3. **Lectin pathway**

1. **The Classical Pathway**

▶ The classical pathway is activated by immune complexes (antibody-antigen complex). However, **not all immunoglobulins are able to activate this pathway.** The immunoglobulin classes that can activate the classical pathway include IgM, IgG, IgG2, and IgG3, but not IgG4, IgA, or IgE.

▶ IgM is the most efficient of the activating immunoglobulins because it has multiple binding sites; thus, it takes only one molecule attached to two adjacent antigenic determinants to initiate the cascade.

▶ In addition to antibodies, a few substances can bind complement directly to initiate the classical cascade. These include C-reactive protein (CRP), several viruses, mycoplasmas, some protozoa, and certain gram-negative bacteria such as *E. coli*. However, most infectious agents can directly activate only the alternative or lectin pathway

▶

Activation Of Classical Pathway
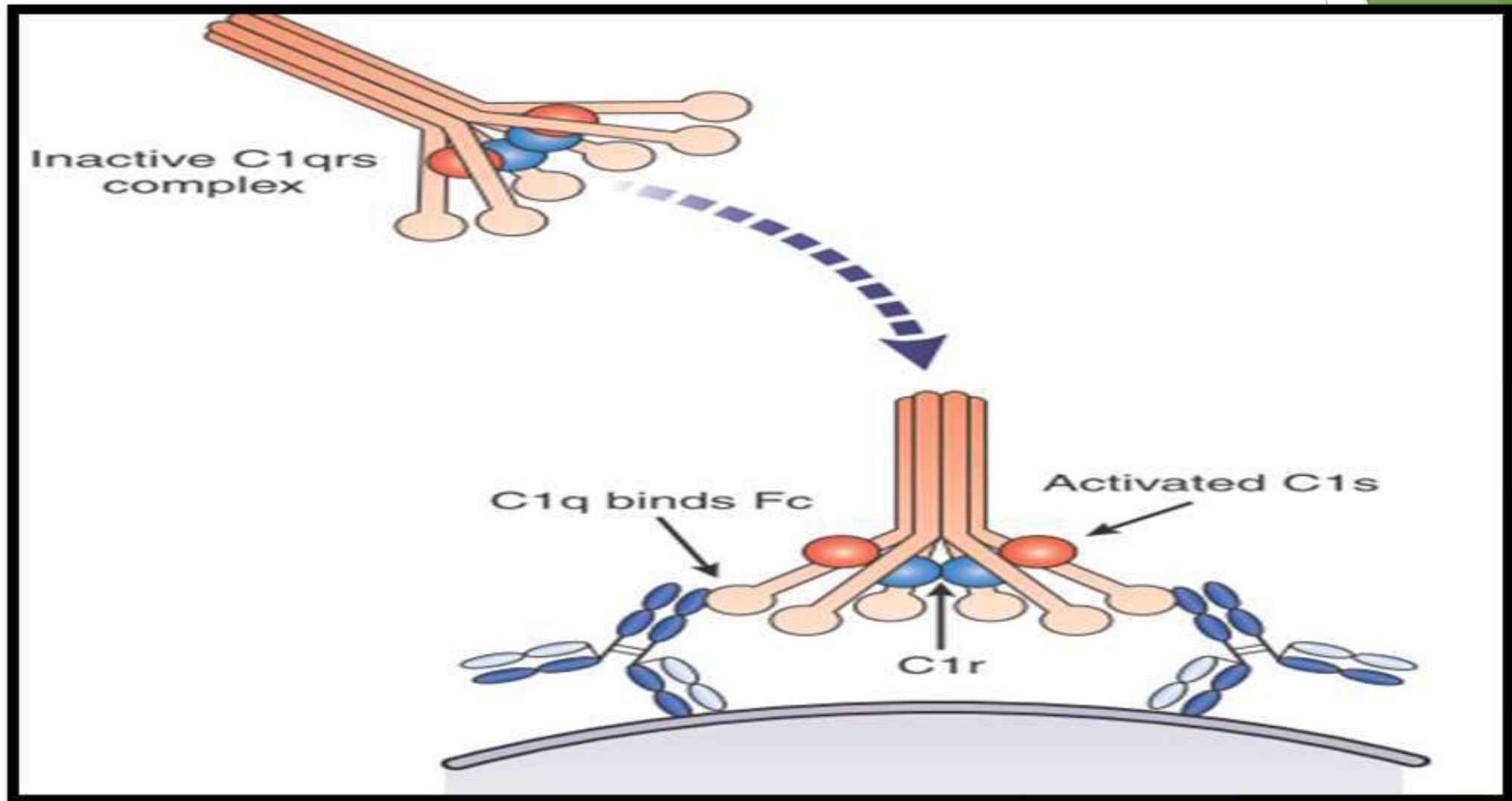
- Conformational change in an immunoglobulin molecule bound to an antigen leads to revelation of the site for binding C1.

- The C1 consists of six subcomponents C1q of a tulip-like structure including a globular head for attaching an antibody, two C1r and two C1s.

- When the antibody attached to a particle of interest (bacterial cell surface or viral envelope) binds to C1q, C1r generates an active enzyme, C1 esterase. Substrates for the enzyme C1s are C2 and C4 that are cleaved into some fragments, C2a/C2b and C4a/C4b.

- C4b and C2a constitute a new enzyme, C4b2a, known as **the classical pathway C3 convertase**, which acts on C3.

- As a rule, during following events, a smaller fragment (−a) will exert powerful biological qualities outside the cascade, whereas a larger fragment (−b) will participate in the formation of new molecules of the complement cascade.

- Next, C3 is cleaved into C3a and C3b. C4b, C2a, and C3b form the enzyme C4b2a3b, known as the classical pathway C5 convertase, which splits C5 into C5a and C5b, a component of the membrane attack complex, C5b6789 (MAC).

- The MAC makes holes in the membrane of a target cell, leading to its osmotic lysis.

CLASSICAL PATHWAY

Antigen:antibody complexes (pathogen surfaces)

↓

C1q, C1r, C1s
C4
C2

→ C3 convertase

C3a, C5a → Peptide mediators of inflammation, phagocyte recruitment

C3b → Binds to complement receptors on phagocytes → Opsonization

Terminal complement components
C5b
C6
C7
C8
C9

→ Membrane-attack complex, lysis of certain pathogens and cells

▶ **Alternative Pathway**

▶ The alternative pathway was originally named for **the protein properdin.** Although properdin has been confirmed to bind and initiate activation, the primary function of properdin is to stabilize the **C3 convertase** formed from activation of other factors.

▶ In addition to properdin, the serum proteins **Factor B and Factor D** are unique to this pathway. C3 is a key component of this pathway.

▶ **Alternative pathway** is initiated by cell-surface constituents that are recognized as foreign to the host, such as LPS. A variety of enzymes (e.g., kallikrein, plasmin, elastase) cleave C3, the most abundant serum complement component, into several smaller fragments. One of these, the continuously present, short-lived, and unstable **C3b fragment, is the major opsonin of the complement system and readily attaches to receptors on cell surfaces.**

1. **C3 is hydrolyzed by water to produce C3b, which binds Factor B and together they attach to target cell surface.**

2. B is cleaved by Factor D into the fragments Ba and Bb. Bb combines with C3b to form C3bBb, an enzyme with C3 initiation convertase activity.

3. More C3 is cleaved by factor D and forming more C3bBb. This enzyme is stabilized by properdin, and it continues to cleave additional C3.

4. If a molecule of C3 remains attached to the C3bBbP enzyme, the convertase now has the capability to cleave C5

5. The C5 convertase thus consists of C3bBbP3b. After C5 is cleaved. C5b inserts into the cell membrane and is the necessary step leading to formation of the membrane attack complex (MAC) and cell lysis.
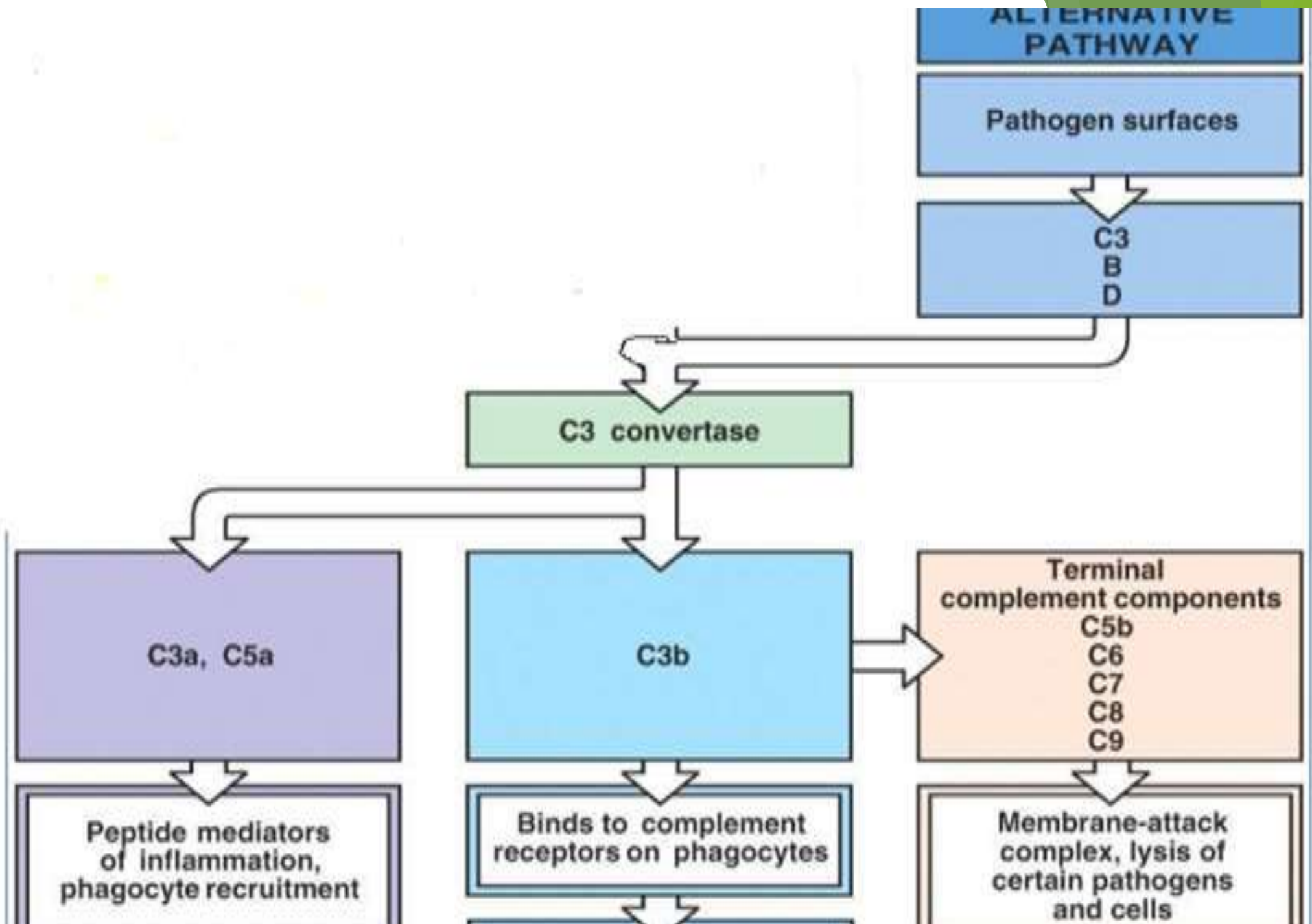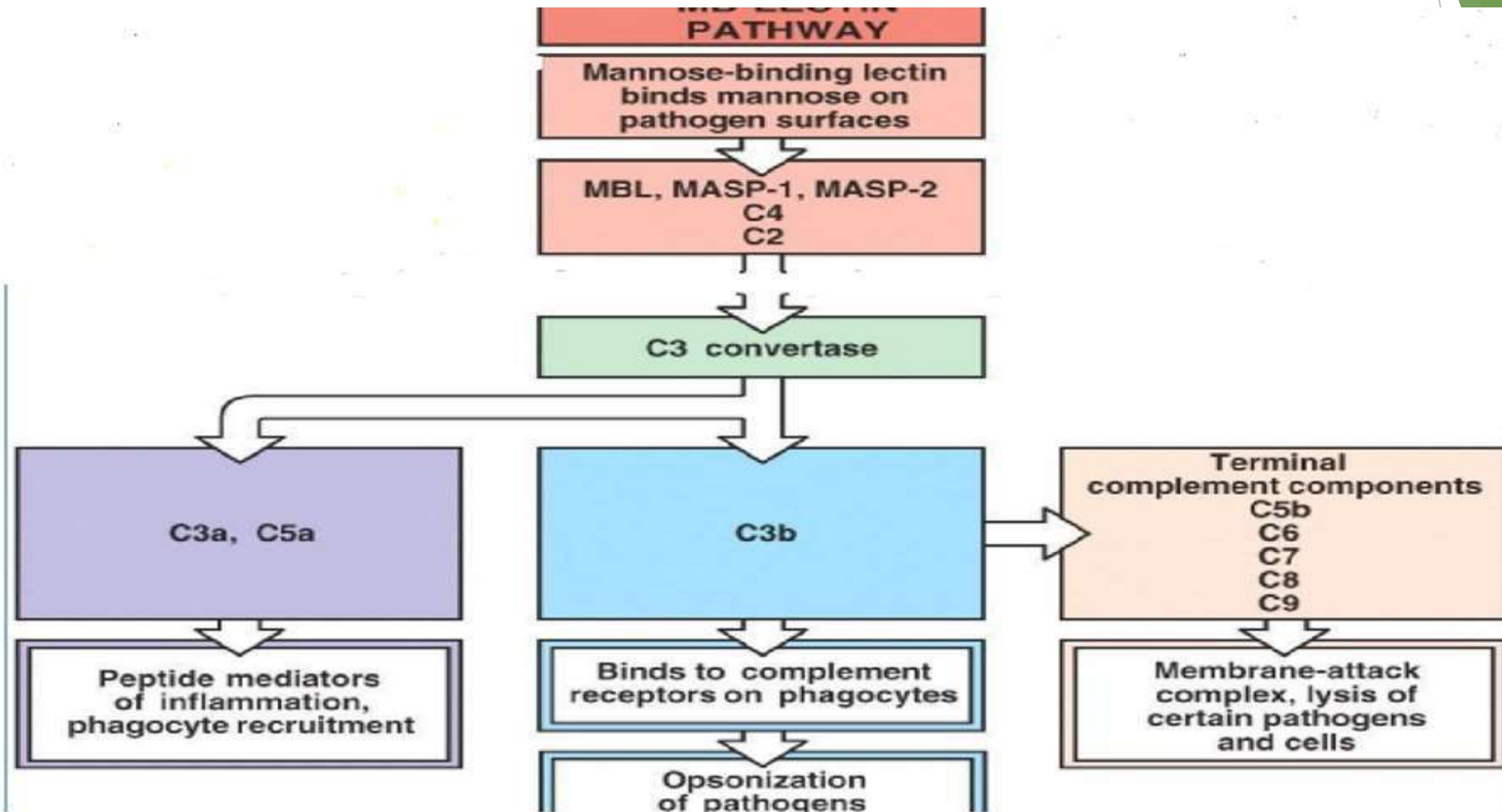
The alternative pathway.
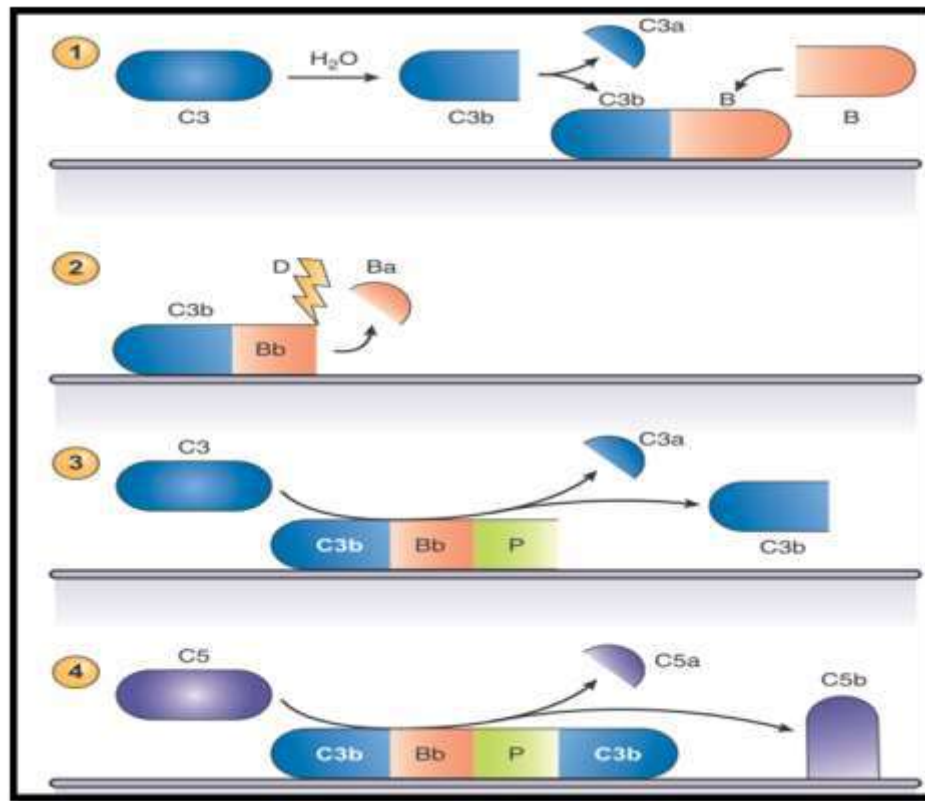
Mannan-binding lectin pathway.

Lectins are proteins that bind to specific carbohydrates. This pathway is activated by binding of mannan-binding lectin (MBL) to mannose-containing residues of glycoproteins on certain microbes (e.g., Listeria spp., Salmonella spp., Candida albicans).

MBL is an acute phase protein, one of a series of serum proteins whose levels can rise rapidly in response to infection, inflammation, or other forms of stress.

1. MBL, once bound to appropriate mannose-containing residues, can interact with MBL-activated serine protease (MASP).

2. Activation of MASP leads to subsequent activation of components C2, C4, and C3.

lectin pathways

Convergence of the classical, alternative, and lectin pathways.
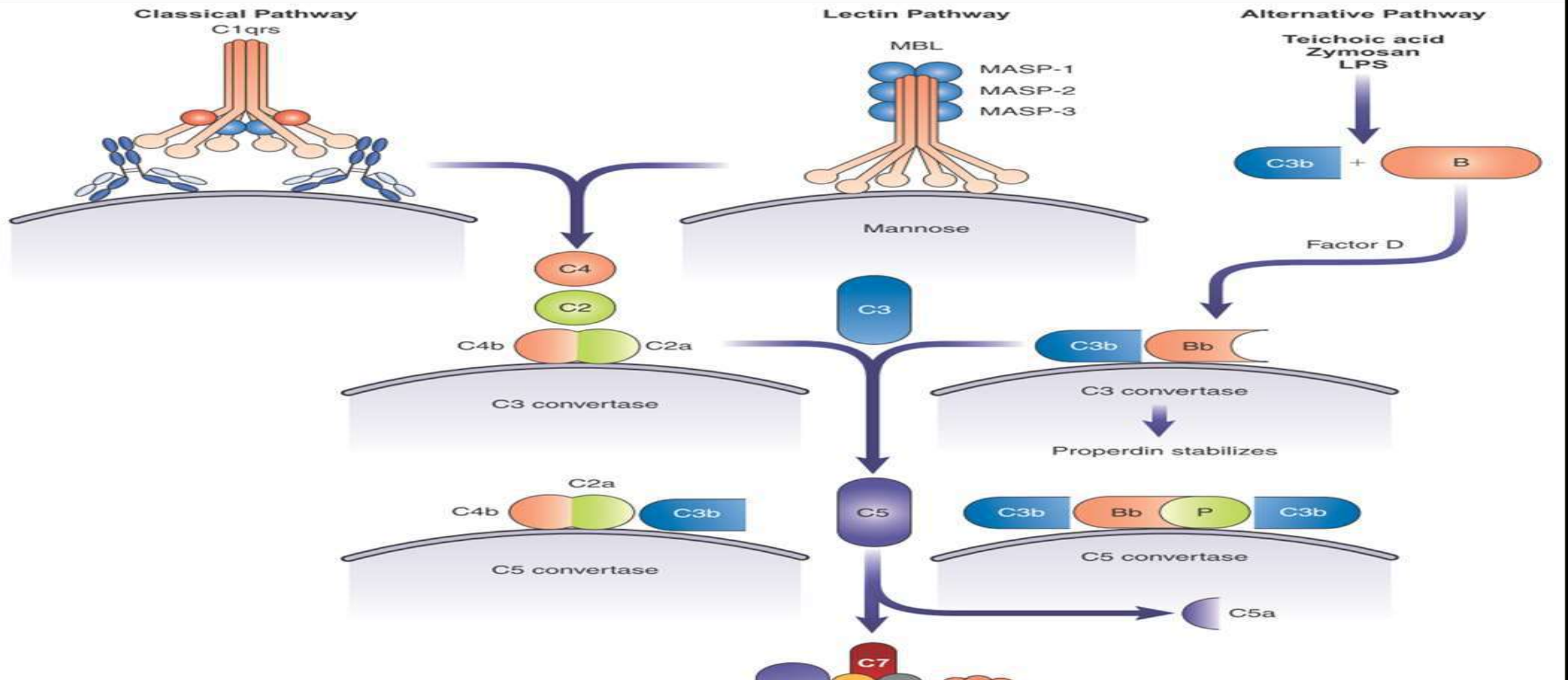
❊ **Anaphylotoxins**. The small fragments (C3a, C4a, C5a) generated by the cleavage of C3 and C5 in the alternative pathway and of C3, C4, and C5 in the MBL pathway act as anaphylotoxins. Anaphylotoxins attract and activate different types of leukocytes. They draw additional cells to the site of infection to help eliminate the microbes. C5a has the most potent effect, followed by C3a and C4a.

Proteins of the Complement System

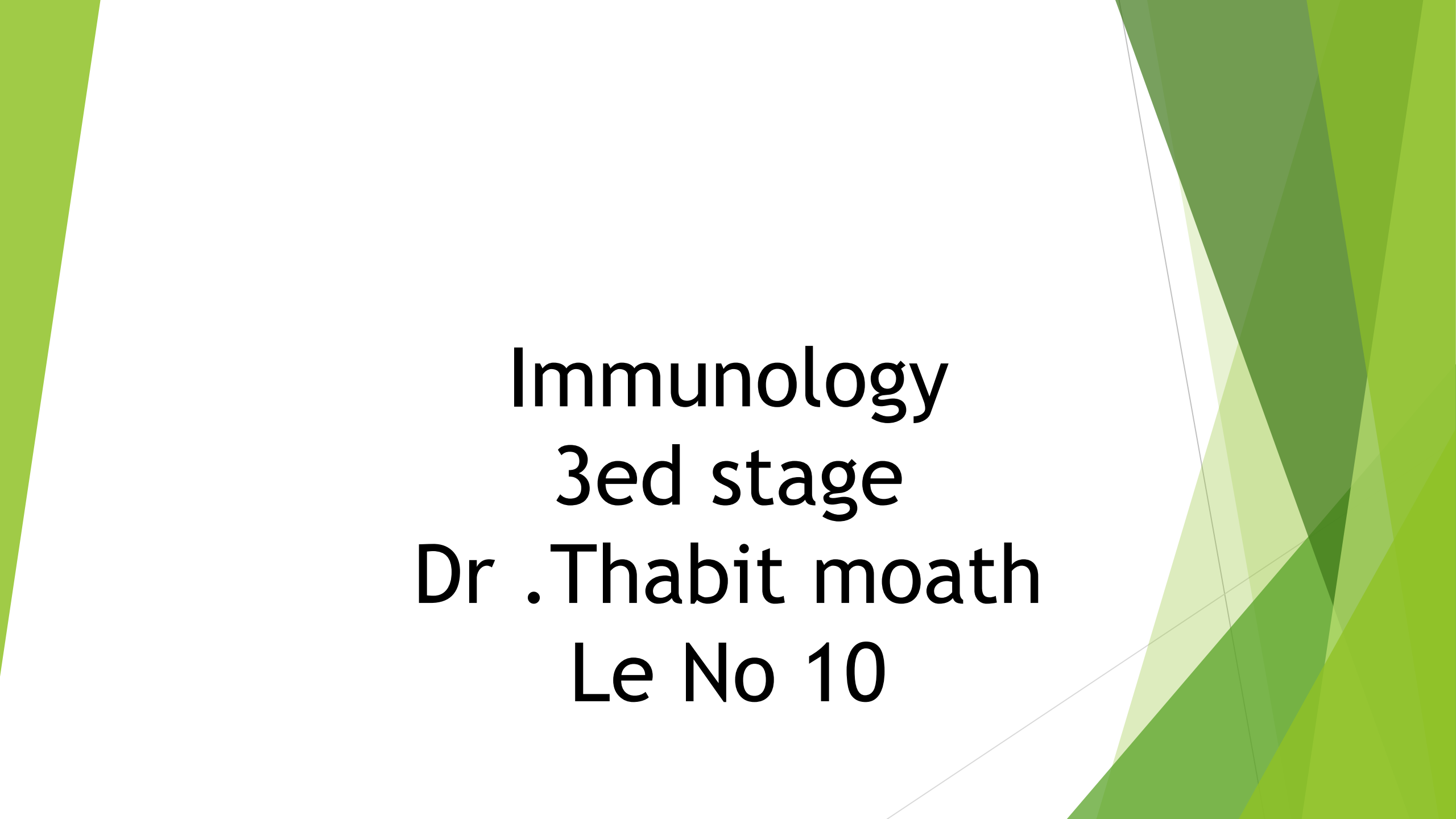| Complement Protein	Function
Classical Pathway	
C1q	Binds to Fc region of IgM and IgG
C1r	Activates C1s
C1s	Cleaves C4 and C2
C4	Part of C3 convertase (C4b(
C2	Binds to C4b—forms C3 convertase
C3	Key intermediate in all pathways
C5	Initiates membrane attack complex
C6	Binds to C5b in MAC
C7	Binds to C5bC6 in MAC
C8	Starts pore formation on membrane
C9	Polymerizes to cause cell lysis
Alternative Pathway	
Factor B	Binds to C3b to form C3 convertase
Factor D	Cleaves Factor B
Properdin	Stabilizes C3bBb—C3 convertase
Lactin pathway	
MBL	Binds to mannose
MASP-2	Cleaves C4 and C2
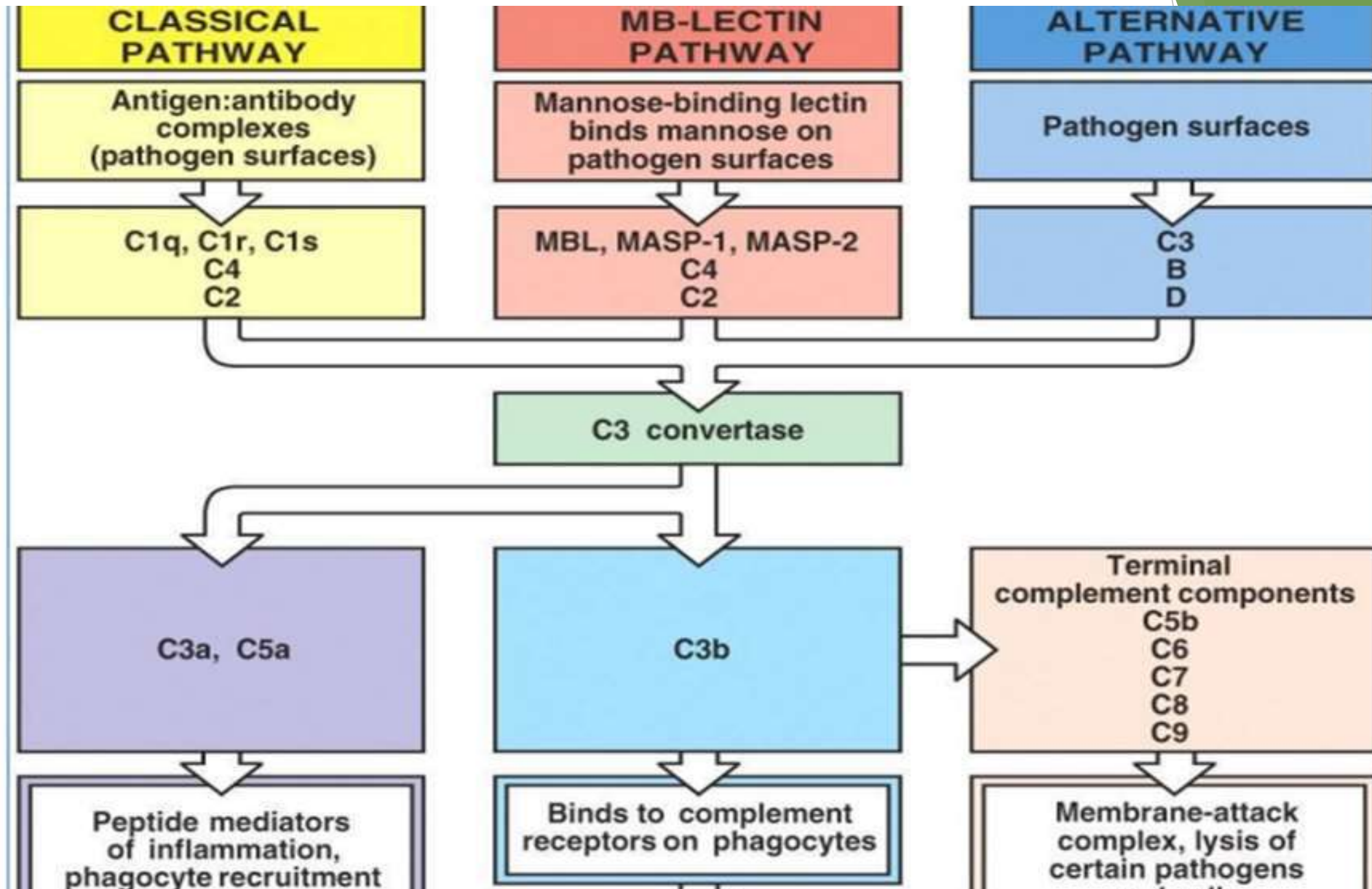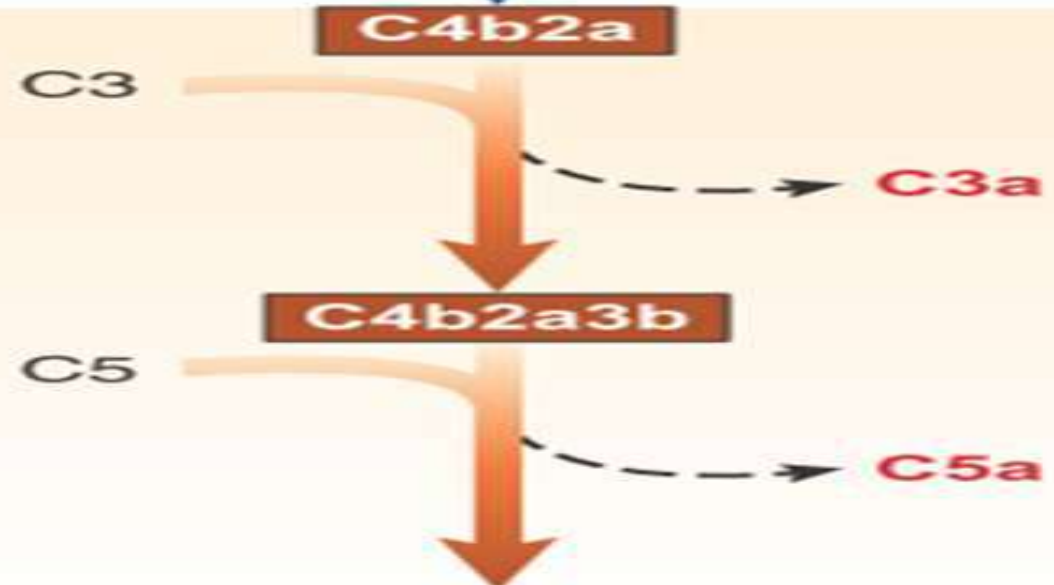
▶ Regarding the complement pathway, which one of the following is the most accurate?

▶ (A) C3 convertase protects normal cells from lysis by complement.

▶ (B) C3a is a decay-accelerating factor and causes the rapid decay and death of bacteria.

▶ (C) In general, gram-positive bacteria are more likely to be killed by complement than gramnegative

▶ bacteria.

▶ (D) The membrane attack complex is formed as a result of activation of the classic pathway but not

▶ by activation of the alternative pathway.

▶ (E) The first time a person is exposed to a microorganism, the alternative pathway of complement

▶ is more likely to be activated than the classic pathway.

▶ 2. Of the following complement components, which one is the most important opsonin?

▶ (A) C1

▶ (B) C3a

▶ (C) C3b

▶ (D) C5a

▶ (E) C5b

▶ 3. Of the following complement components, which one is the most potent in attracting neutrophils to

▶ the site of infection (i.e., acting as a chemokine)?

▶ (A) C1

▶ (B) C2

▶ (C) C3b

▶ (D) C5a

▶ (E) Mannan-binding lectin

- 4. Of the following, which one is the most important function of the complex formed by complement

- components C5b,6,7,8,9?

- (A) To enhance antibody production

- (B) To inhibit immune complex formation

- (C) To opsonize viruses

- (D) To perforate bacterial cell membranes

- (E) To release histamine from mast cells

- 5. A deficiency of which one of the following complement components predisposes to bacteremia

- caused by members of the genus Neisseria?

- (A) C1

- (B) C3b

- (C) C5a

- (D) C5b

- (E) C5b,6,7,8,9

Immunology
3ed stage
Dr .Thabit moath
Le No 10

Monoclonal and polyclonal antibody

Introduction:

Normal serum contains 1016 antibody molecules per milliliter. These antibodies can be collected from experimental animals by conventional antisera production method as show that in figure (1)

PRODUCTION
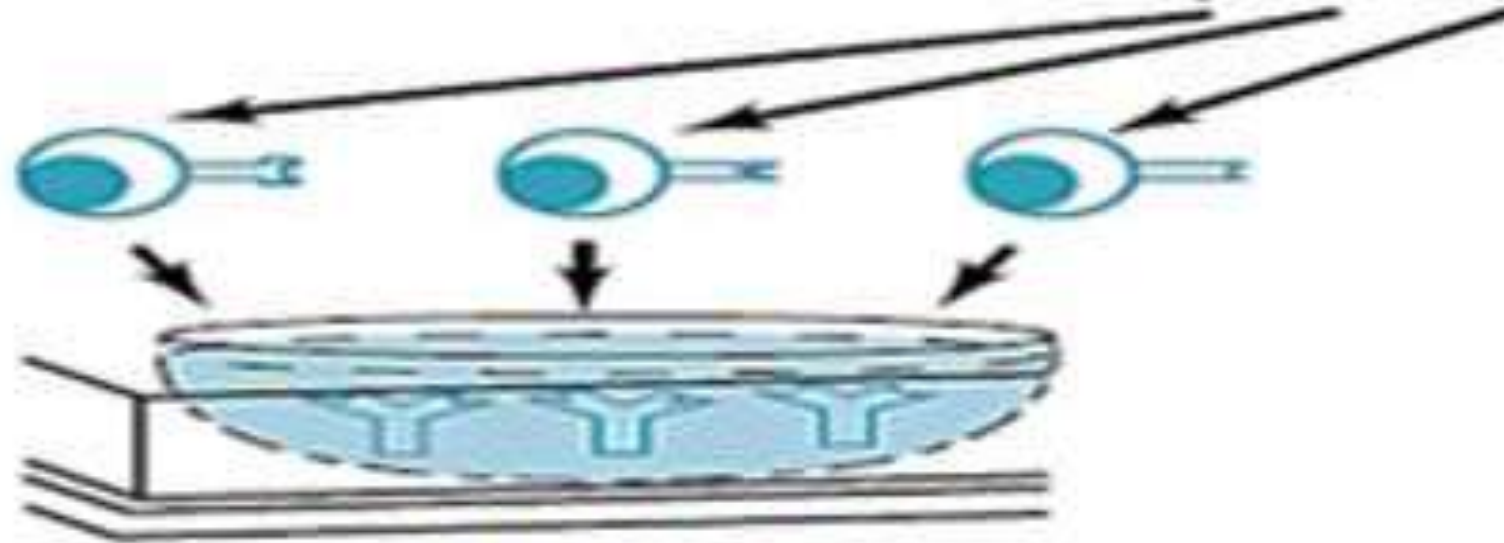
Antigenic determinants

Antigen

Repeated antigen inoculation

Remove spleen

The mouse produces B cells specific for each antigenic determinant.

Spleen cells

Antiserum
(contains a mixture of antibodies specific for each antigenic determinant)

and have been an important tool that used them to identify or label molecules or cells and to separate molecules or cells from mixtures. But these antibodies have been the variability of antisera. When variety antibodies are inducing and producing to different antigens which possess multiple epitopes that resulting serum antibodies are heterogeneous, comprising mixture of antibodies, each specific for one epitope, such antibodies are called polyclonal antibody.
For this reason, recently way of obtaining of pure antibody has defined specificity produced by monoclonal antibodies technique.
The development of monoclonal antibody producing by hybridization techniques is beginning in 1975 when three immunologists (Nielsen, George, and Milstein) were observed that antibody producing B-cells may become cancerous and promote to produce one kind of specific antibody in large amount. In 1984, each of three immunologists has awarded Nobel-Prize for their discovery of a method to prolong the culture life of antibody producing B-cells.
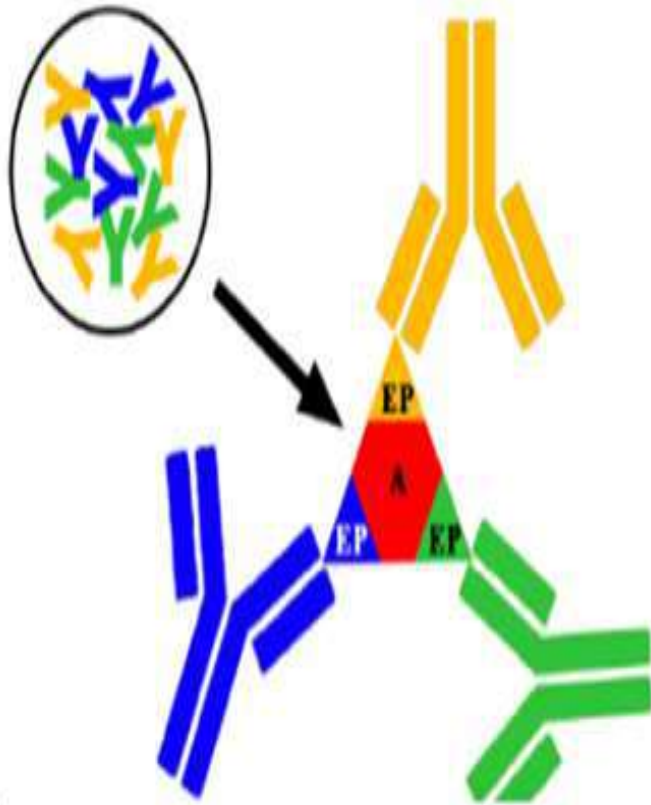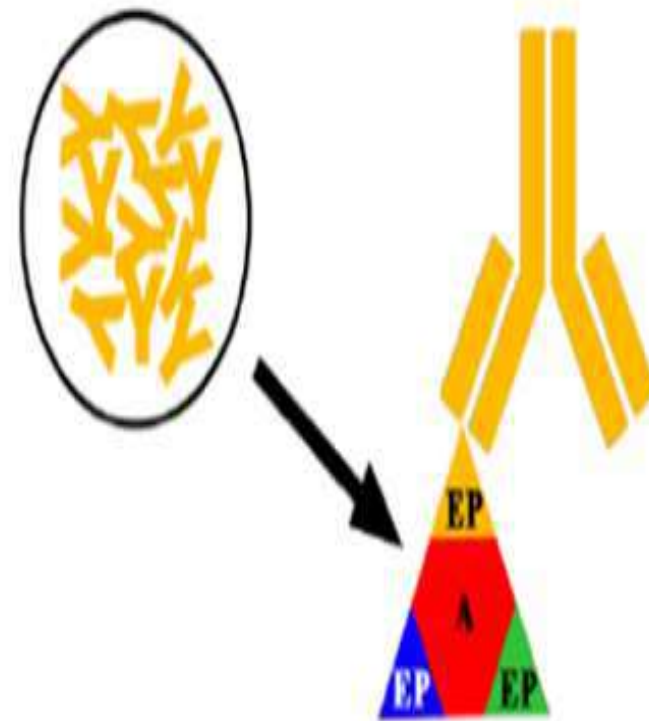
Cells types used in monoclonal antibody technique:
1. Continuous proliferation cells are called myelomas cancer cells (e.g. cancerous plasma cells) which can be isolated and propagated in cell culture.
2. Normal B cells producing antibody
When fused occurred between these cells the combination is termed a Hybridoma, and the hybridoma cells has genetically identical to continuous growth in culture and produces of antibody. The important of hybrid technique is that clones of the antibody secreting in cells culture and can produce amounts of identical antibody molecules; because all these antibody molecules are produced by a single hybridoma clone, they are called Monoclonal antibody

Monoclonal antibody and it is characterized:
1.   Uniform (has a single idiotype).
2.   Highly specific.
3.   Can be readily produced in large quantities.
   Production of monoclonal antibodies:
 This provides pure preparations of antibodies in large quantities:
1.   A mouse is injected with a specific antigen that will induce immune system to produce antibody.
2. Mouse spleen is removed and a prepare suspension from spleen cells include B cells that produce antibodies against injected antigen.
3. Spleen cells are mixed with myeloma cells line and allow cell fusion. 4. Mixture of cells is placed in a selective medium that allow hybrid cells to grow.
5. Selected hybridomas are cultured to produce large number of monoclonal antibodies.

Formation and selection of hybridoma cells

Hybridoma are produced by using polyethylene glycol to fuse spleen B cells from animals that have been immunized with antigen with myeloma cells which can immortal growth. In vitro, genetic information of fuse cells contribution for synthesis specific antibody Cells fusion random occurs and producer gives a complex mixture containing different combinations which includes:

1. Unwanted spleen-spleen cell fusion.
2. Unwanted myeloma-myeloma cell fusion.
3. Desired fusion between spleen-myeloma cells.
4. Single myeloma cells, and single spleen cells.

Selection for only myeloma-spleen cell fusions is accomplished by culturing cell mixture in medium which contain a folic acid that blocks undesired cells

Antigenic determinants

Antigen

Repeated antigen inoculation

Remove spleen

The mouse produces B cells specific for each antigenic determinant.

Spleen cells

FUSION

Myeloma cells

Hybrid-myeloma cells

Grow up individual cells into clones

Clone 1   Clone 2   Clone 3

Figure (2) is show The main procedure of homogenous monocloncal antibody production by B-cell-derived hybridomas can be separated into individual clones and grown indefinitely. One clone gives rise to identical daughter cells, all producing one antibody idiotype (a monoclonal antibody)

which are myeloma–myeloma cells and free myeloma cells and single spleen cells and spleen–spleen cell. Myeloma cells spleen cells now have grow in HAT medium. About 500 hybrids are formed per mouse spleen, and 20 to 30 of them produce specific antibody. The correct antibody producing hybridomas are then identified.

Clinic uses of monoclonal antibodies:
 Monoclonal antibodies are very useful as diagnostics, imaging, and therapeutic agents in clinical medicine by:
1.	Monoclonal antibodies used in vitro as diagnostic agents (e.g. for detecting pregnancy, diagnosing numerous pathogenic microorganisms, measuring levels of various drugs in blood, and matching histocompatibility antigens).
2.	Radiolabeled monoclonal antibodies can be used in vivo for detect of tumor antigens location.
3.	Tumor treatment
4.	 dissolve blood clot
5.	 cleave viral glycoprotein.
6.	It can be used in treatment of auto immune diseases such as rheumatoid arthritis, prostatic arthritis; because anti-TNF antibodies which inhibit induction of an inflammatory reaction as TNF-α which can affect onto blood vessels, and prevent migration of immune cells, also
7.	inhibit inflammation of synovial, cartilage, and bone marrow metabolism

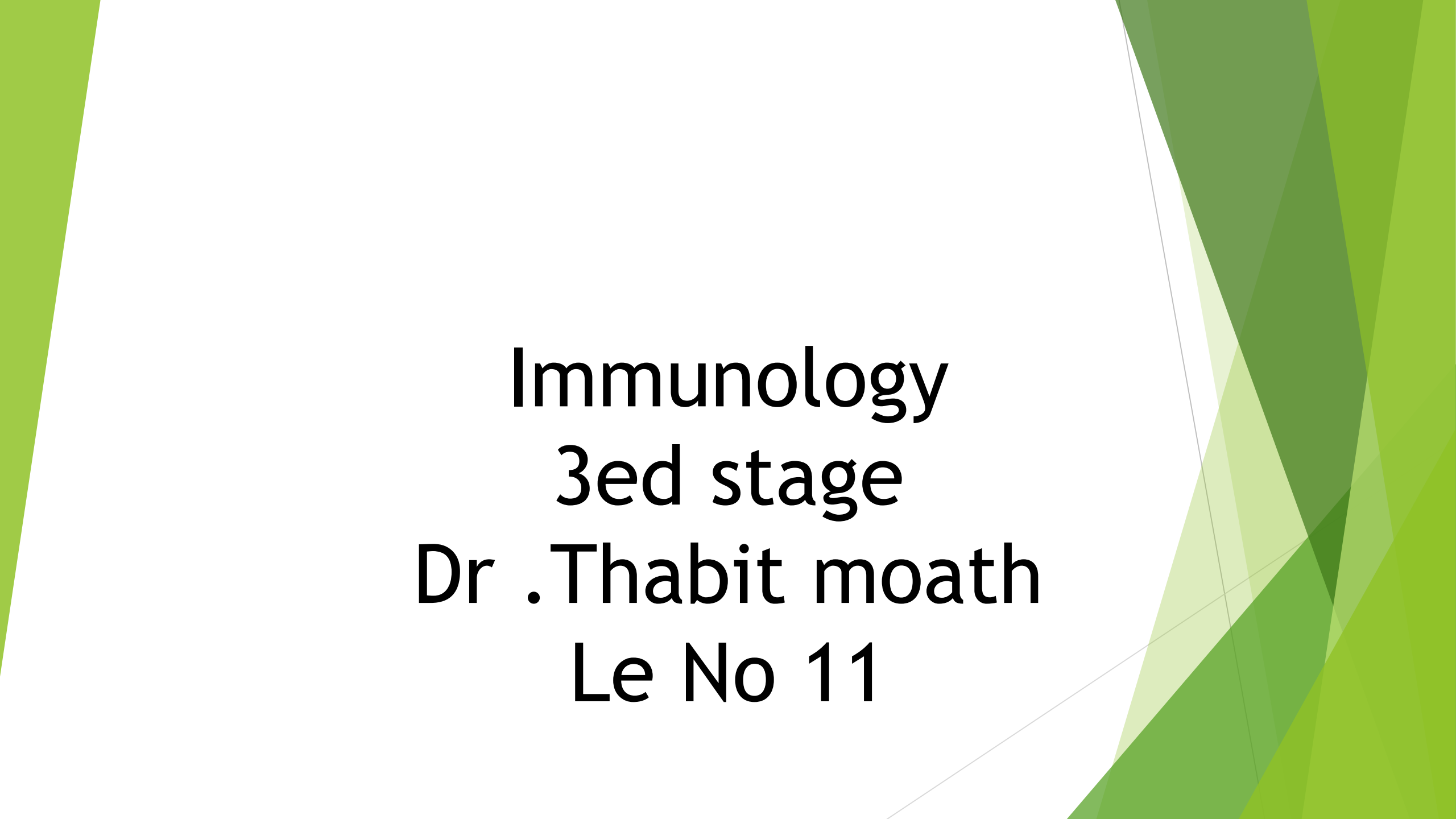| PCA	MCA
at less expense	more expense.
with less technical skill	More technical skill
expect to obtain PAbs within several months of initiating immunizations	generation of hybridomas and subsequent production of MAbs can take up to a year or longer in some cases,
The principal advantages of MAbs are their homogeneity and consistency	Monospecificity evaluating changes in molecular conformation, protein-protein interaction
PAbs are also more stable over a broad pH and salt concentration	whereas MAbs can be highly susceptible to small changes in both
the concentration and purity levels of specific antibody are less in pAbs	the concentration and purity levels of specific antibody are higher in MAbs

Immunology
3ed stage
Dr .Thabit moath
Le No 11

Major Histocompatibility Complex (MHC)

The major histocompatibility complex (MHC), also called the **Human Leukocyte Antigen (HLA) complex**, is a segment of chromosome 6 containing several genes that are critical to immune function. These include genes encoding a variety of enzymes and structural molecules needed for the activation and function of B and T cells. The encoded molecules fall into three groups or classes known as MHC (or HLA) class I, II, and III molecules. MHC class III molecules include complement components C4, Bf, and C2. MHC class I and II molecules serve entirely different functions.
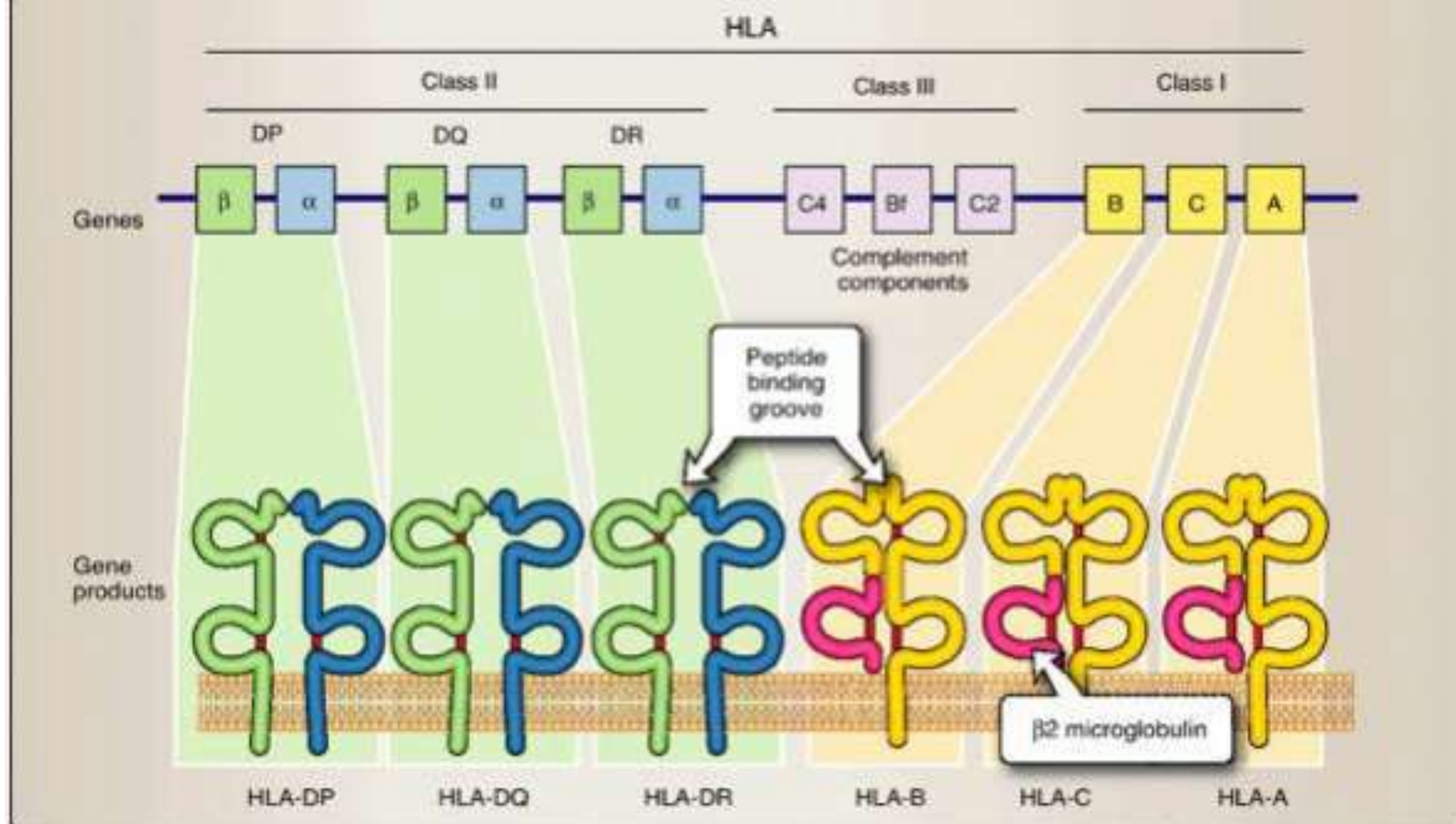
Genetic and protein organization of MHC class I, II, and III. Located on chromosome 6, HLA (human leukocyte antigen) genes are arranged as shown. They are grouped into Class I, Class II, and Class III based on structural and functional characteristics.

A. MHC class I molecules

MHC class I molecules are one of two primary classes of major histocompatibility complex (MHC) molecules (the other being MHC class II) and **are found on the cell surface of all nucleated cells in the bodies of vertebrates**. They also occur on platelets, but not on red blood cells.

✳ Their function is to display peptide fragments of proteins from within the cell to cytotoxic T cells; this will trigger an immediate response from the immune system against a particular non-self-antigen displayed with the help of an MHC class I protein. Because MHC class I molecules present peptides derived from cytosolic proteins, the pathway of MHC class I presentation is often called cytosolic or endogenous pathway.

✳ class I MHC itself can serve as an inhibitory ligand for natural killer cells (NKs). Reduction in the normal levels of surface class I MHC, a mechanism employed by some viruses and certain tumors to evade Cytotoxic T lymphocytes (CTL) responses, activates NK cell killing.

✳ In humans, the HLAs corresponding to MHC class I are HLA-A, HLA-B, and HLA-C.

α_2 α_1

α_3 β-microglobulin

Schematic representation of MHC class I molecule, consisting of three α-domains and one β2-microglobulin molecule. The peptide-binding groove is situated between domains α1 and α2.
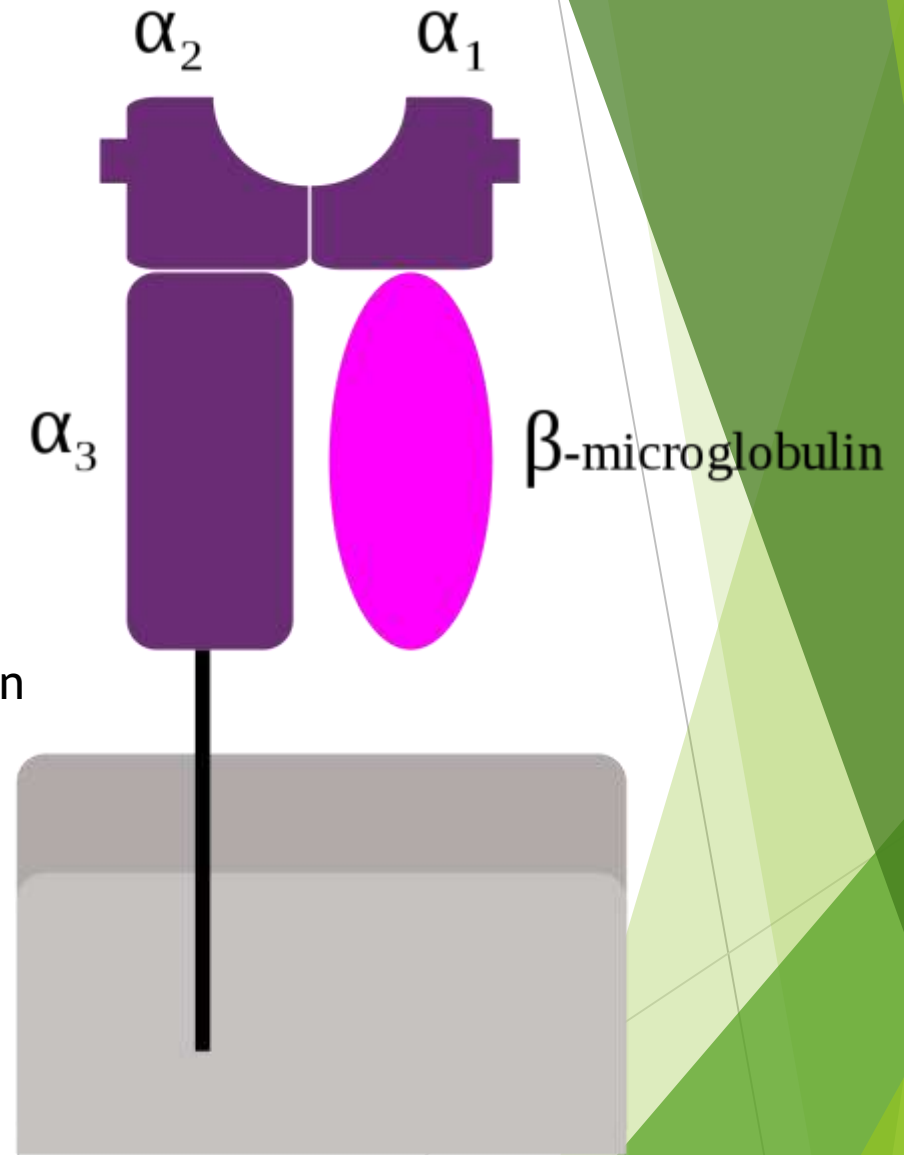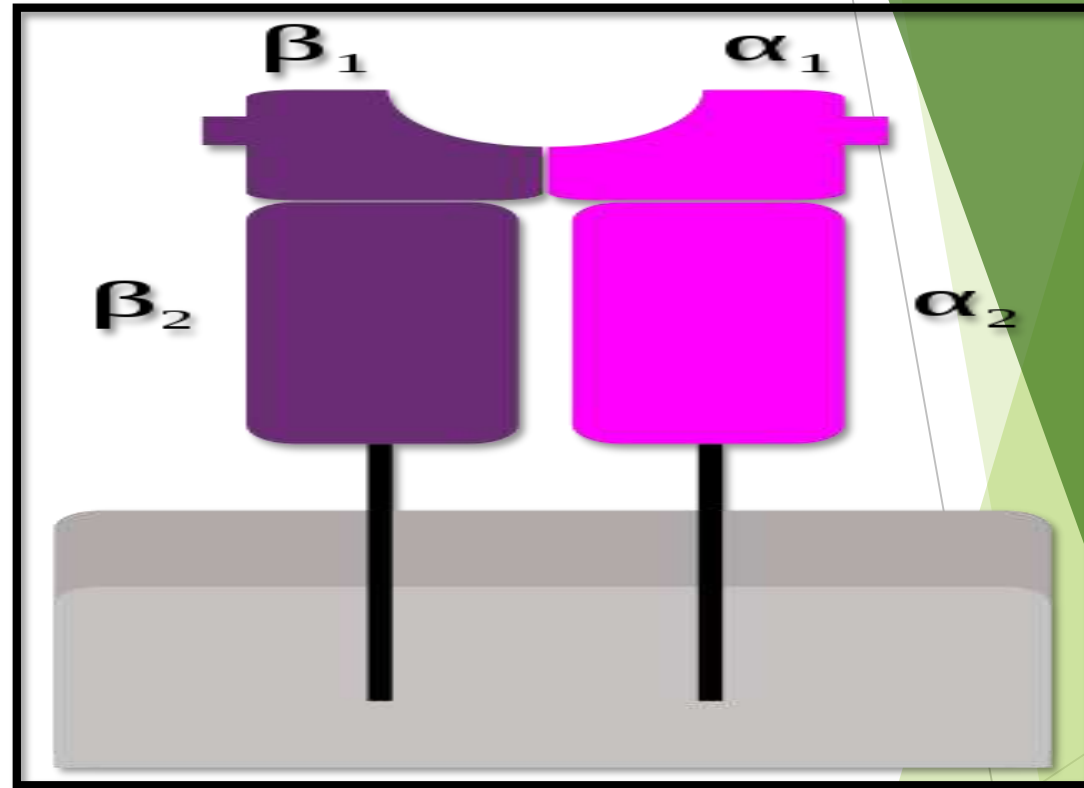
B.   MHC class II molecules

MHC Class II molecules are a class of major histocompatibility complex (MHC) molecules normally found only on professional antigen-presenting cells such as dendritic cells, mononuclear phagocytes, some endothelial cells, thymic epithelial cells, and B cells. These cells are important in initiating immune responses.The antigens presented by class II peptides are derived from extracellular proteins (not cytosolic as in MHC class I).

Loading of a MHC class II molecule occurs by phagocytosis, extracellular proteins are endocytosed, digested in lysosomes, and the resulting epitopic peptide fragments are loaded onto MHC class II molecules prior to their migration to the cell surface. In humans, the MHC class II protein complex is encoded by the human leukocyte antigen gene complex (HLA). HLAs corresponding to MHC class II are HLA-DP, HLA-DQ, and HLA-DR.

❋ **<u>Importance and function</u>**

❖ Having MHC class II molecules present proper peptides that are bound stably is essential for overall immune function. Because class II MHC is loaded with extracellular proteins, it is mainly concerned with presentation of extracellular pathogens (e.g. bacteria that might be infecting a wound or the blood).

❖ Class II molecules interact mainly with immune cells, like the T helper cell (CD4+). The peptide presented regulates how T cells respond to an infection.

❖ Stable peptide binding is essential to prevent detachment and degradation of a peptide, which could occur without secure attachment to the MHC molecule. This would prevent T cell recognition of the antigen, T cell recruitment, and a proper immune response.

❖ The triggered appropriate immune response may include localized inflammation and swelling due to recruitment of phagocytes or may lead to a full-force antibody immune response due to activation of B cells.

Schematic representation of MHC class II molecule, consisting of two α-domains and two β-domains. The peptide-binding groove is situated between domains α1 and β1.

Antigen Presentation

Phagocytes are cleanup their surrounding environment by phagocytosis and macropinocytosis. Ingested proteins are enzymatically degraded, and some of the resulting peptide fragments are loaded into MHC class II (forming pMHC class II) molecules in a process called **antigen presentation**. Some pathogens avoid phagocytotic and macropinocytic mechanisms altogether or infect cells that do not express MHC class II molecules. Such antigens are broken down, and their peptide fragments are loaded into MHC class I (forming pMHC class I) molecules.

1. **Presentation by MHC class II**

2. Class II MHC binds invariant chain to block binding of endogenous antigen.

3. MHC complex goes through Golgi complex.

4. Invariant chain is degraded, leaving CLIP(CLIP: class II invariant chain peptide) fragment.

5. Exogenous antigen taken in and degraded and routed to intracellular vesicle.

6. CLIP fragment exchanged for antigenic peptide.

7. Class II MHC antigenic peptide is transported to cell surface.

8. Class II MHC peptide complex binds to CD4+ T cell.
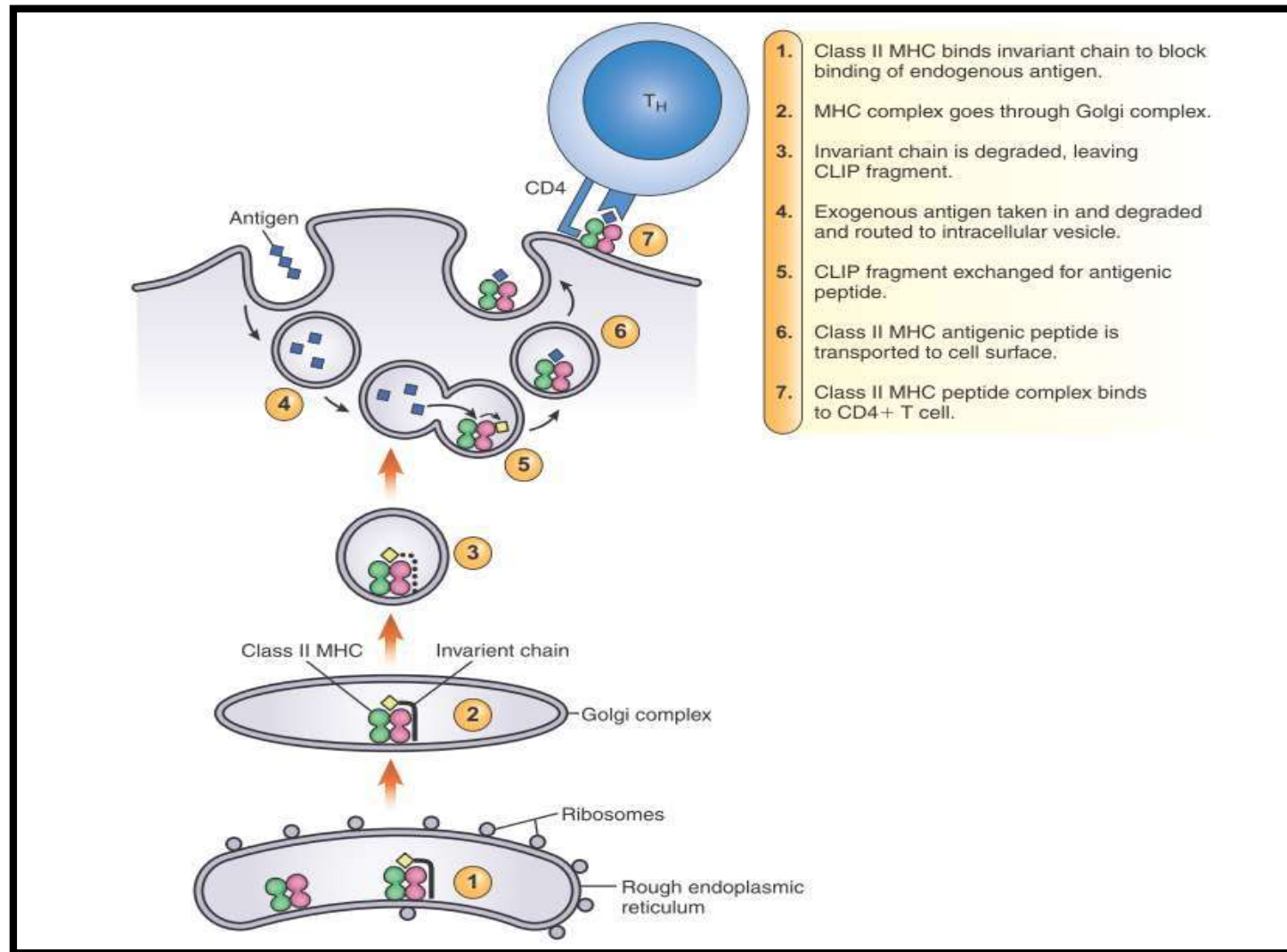
Antigen-processing pathway for exogenous antigen.

1. Presentation by MHC class I

Not all antigens enter cells by phagocytosis or macropinocytosis. Some pathogens avoid phagocytes and endocytic vesicles entirely. Intracellular microbes and viruses bind to cell membranes and directly enter the cytoplasm of the host cell. These pathogens are processed differently.

Nucleated cells normally degrade and recycle cytoplasmic proteins. Both self and non-self-cytoplasmic proteins targeted for destruction are covalently tagged with ubiquitin, a highly conserved protein. Binding of one or more ubiquitin molecules to a protein selects it for destruction by the proteasome, a large proteolytic enzyme complex within the cytoplasm.

1. Endogenous antigen within cytosol is degraded by proteasome.

2. Peptides transported into endoplasmic reticulum by TAP.

3. Alpha chain of class I MHC binds β_2- microglobulin.

4. Alpha chain of class I MHC binds peptide.

5. Peptide-class I MHC transported to Golgi complex and then to cell surface .

6. Class I MHC peptide binds to CD8+ T cell.

1. Endogenous antigen within cytosol is degraded by proteasome.
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5. Peptide-class I MHC transported to Golgi complex and then to cell surface.
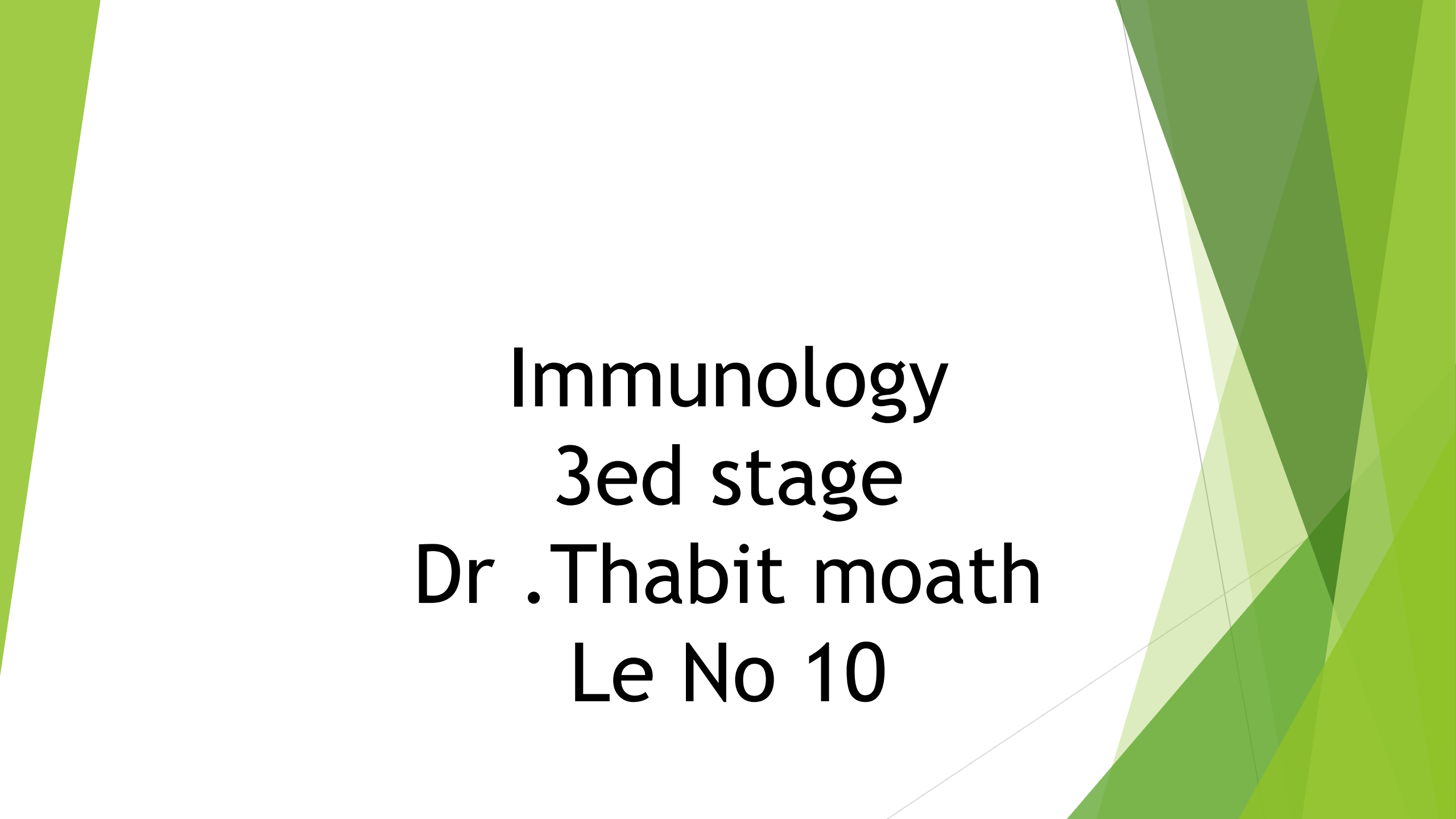6. Class I MHC peptide binds to CD8+ T cell.

CD8

T

Class I MHC — β_2-microglobulin
Golgi complex

Ribosomes
Rough endoplasmic reticulum

Protein
Tap
Proteosome
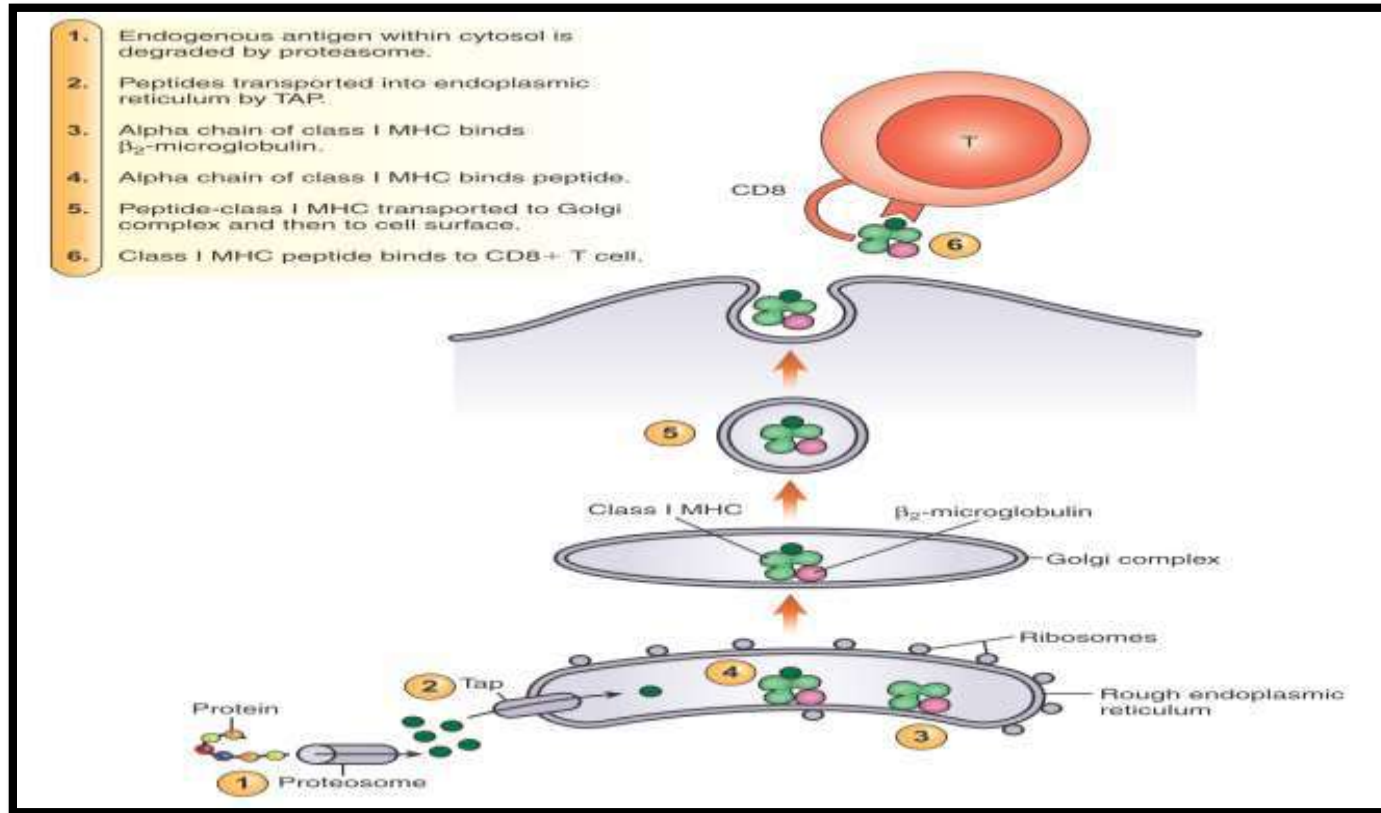
▶ Antigen-processing pathway for endogenous antigens on MHC class I .

A Comparison of Class I and Class II MHC Molecules

	Class I MHC Molecules	Class II MHC Molecules
Cellular Distribution	All nucleated cells	B cells, monocytes,macrophages, dendritic cells, thymic epithelial cells
Structure	One α chain and β_2-microglobulin	An α chain and a β chain
Classes	A, B, C	DP, DQ, DR
Size of Peptides Bound	8 to 11 amino acids	13 to 18 amino acids
Nature of Peptide Binding Cleft	Closed at both ends	Open at both ends
Interaction with T Cells	Presents endogenous antigen to CD8+ T cells	Presents exogenous antigen to CD4+ T cells

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Le No 10

Cytokines and Chemokine

❖ Cytokines

Cytokines are small secreted proteins released by cells have a specific effect on the interactions and communications between cells. Cytokine is a general name; other names include **lymphokine** (cytokines made by lymphocytes), **monokine** (cytokines made by monocytes), **chemokine** (cytokines with chemotactic activities), and **interleukin** (cytokines made by one leukocyte and acting on other leukocytes).

Cytokines may act on the cells that secrete them (autocrine action), on nearby cells (paracrine action), or in some instances on distant cells (endocrine action). **Cytokines** are specialized regulatory molecules, which mainly act on cells in a short distance manner. They all are low-molecular-weight proteins, peptides, or glycoproteins.

Some cytokine groups may function in both a synergic and an antagonistic manner and in both a pro-inflammatory and an anti-inflammatory way. They may affect numerous processes in the immune system including reactions of the innate and adaptive immune responses.

There are some classifications of cytokines. The Classifications of cytokines are based upon: **identical or shared biological activities** and **shared biochemical properties of cytokines are problematic and complex**. Families of cytokines share sequence similarity and display homology in their mutual receptor systems.

The classical nomenclature of cytokines includes:

1. Interleukins (ILs)

2. Colony-stimulating factors (CSFs)

3. Interferons (IFNs)

4. Tumor necrosis factors (TNFs)

5. Transforming growth factors (TGFs)

1. Interleukins (ILs)

Interleukins (ILs) are a group of cytokines (secreted proteins and signal molecules) that were first seen to be expressed by white blood cells (leukocytes). They are identical in 15–25% of their amino. The human genome encodes more than 50 interleukins and related proteins.

The majority of interleukins are synthesized by **helper CD4$^+$ T lymphocytes**, as well as through **monocytes**, **macrophages**, and **endothelial cells**. They promote the development and differentiation of T and B lymphocytes, and hematopoietic cells.

IL-1 and IL-6 may be secreted by **macrophages** in response to "patterns" to result in toxic shock syndrome. IL-1 is divided into IL1α and IL1β. IL-1 and IL-6 may act at the systemic level like a hormone and takes part in the **"acute phase" inflammation** and in the maintenance of the blood-brain barrier.

IL-2 is secreted by **T cells**, **upregulates T and B-cell growth in the adaptive immune responses** and **activates NK cells as cells of the innate immunity**. **IL15** act like IL-2

IL-4 is produced by **type 2 helper T (Th2) cells**, **mast cells**, and **B cells**. It is the keystone cytokine of Th2 and IgE antibody synthesis. Analogous to IL21, IL4 plays a role in the advanced B-cell-mediated immune response. Also **IL13** which act like IL4

IL-5 refers to cytokines of the Th2 profile and upregulates the eosinophil generation, maturation, and activation.

IL7 is a hemopoietic growth factor secreted by **stromal and other cells in the bone marrow**. IL7 upregulates the differentiation of multipotent stem cells into lymphoid stem cells, which are very important for both B lymphopoiesis and T lymphopoiesis.

IL8 (CXCL8) refers to chemokines. IL8 is responsible for the migration of neutrophils toward the site of infection, activation of phagocytosis, and angiogenesis.

IL10 is a keystone anti-inflammatory and immunosuppressive cytokine, but it belongs to the Th2 profile. IL10 is also produced by **natural T regulatory (nTreg) cells**, mast cells, and B cells, which down-regulates type 1 helper T (Th1) cells, and the expression of HLA molecules and co-stimulatory molecules required for the adaptive immune responses.

IL12 is produced by **antigen-presenting cells** and **NK cells** to stimulate the Th1 formation during T-cell-mediated responses. IL12 up-regulates the activity of cytotoxic CD8+T cells and NK cells due to the production of IFNγ and TNFα by these cells.

IL17 is a **keystone cytokine** of type 17 helper T cells and produced by some CD4^{+}T cells under the influence of IL23. IL17 is a pro-inflammatory cytokine involved in the chronic inflammation with the participation of neutrophils and other cells and in autoimmune disorders. IL17 acts synergistically with TNFα, TNFβ, IL1, and IL6.

1. Colony-Stimulating Factors (CSFs)

Granulocyte-macrophage colony-stimulating factor (GM-CSF) is secreted by a many types of cells and stimulates stem cells in the bone marrow to produce granulocytes and monocytes. GM-CSF may function at the systemic level and affect mature cells of the immune system.

1. **<u>Interferons (IFNs)</u>**

<u>Interferons</u> (IFNs) are host-encoded secreted proteins, often join in multiple immune interplays and perform both the **induction and regulation of innate and adaptive** <u>antiviral mechanisms</u> when viruses infect the host. Once viral infections occur, the expression of IFN (IFN-α & IFN-β) <u>will function as a essential innate antiviral defense responses</u>.

<u>IFNα</u> and **<u>IFNβ</u>** is **an pro-inflammatory and anti-inflammatory cytokine**, secreted by **fibroblasts and other cell types**. They act as a <u>stimulator of NK</u> cell and <u>macrophage activity</u>. They play an important role in defense against **viral infections by directly inhibits viral replication.**

<u>IFNγ</u> is <u>produced by lymphocytes upon their activation and shows wide immunoregulatory qualities in the immune processes.</u>

1. **Tumor Necrosis Factors (TNFs)**

TNFs is a small protein used by the immune system for cell signaling

TNFα is produced by macrophages and a variety of cell types. TNFα acts synergistically with TNFβ, IL1, IL6, and IFNγ. It is a potent pro-inflammatory cytokine and even endogenous toxin, an endogenous pyrogen, regulator of **adaptive immune** responses and lyse particular tumor cell lines.

clinical point: there are many clinical conditions associated with cytokine effects, which may be both positive and negative. Over secretion of cytokines can trigger **dangerous auto-inflammatory** syndromes known as a **cytokine storm** as well as **toxic shock syndrome**. The specific activities of some cytokines have been the basis for **therapeutical** intervention, such the treatment of malfunctions of hemopoiesis and tumor therapy. Current concepts use the support of chemo- and radiotherapy, bone marrow transplantation, and approaches to immune enhancement therapy

In general cytokines can be dividing into two groups:

1. **pro-inflammatory cytokines** are secreted from Th1cells, CD4$^+$ cells, macrophages, and dendritic cells. They are characterized by production of IL-1, IL-2, IL-12, IL-17, IL-18, IFN-γ, and TNF-α. **The key pro-inflammatory cytokines are IL-1, IL-6, and TNF-α.** They are essential for directing cell mediated response and play a critical role in modulating the immune system. Pro-inflammatory cytokines regulate growth, cell activation, differentiation, and homing of immune cells to the sites of infection to control and eradicate the intracellular pathogens.

2. **The anti-inflammatory cytokines** are a series of immune-regulatory molecules that control **and inhibit the pro-inflammatory cytokine response**. Their physiologic role in inflammation and pathologic role in systemic inflammatory states are increasingly recognized. Major anti-inflammatory cytokines include IL-1 receptor antagonist, IL-4, IL-10, IL-11, and IL-13. INF-α, IL-6, and (TGF)-β are categorized as either anti-inflammatory or pro-inflammatory cytokines, under various conditions.

❖ Chemokine

Chemokine (chemo-attractants) are specialized cytokines, which drive the directed **migration of immune system's cells for homing and/or inflammation**. Chemokine induce the **recruitment** of well-<u>defined cell types and leukocyte subsets</u>. Chemokine have been structurally classified into four main subfamilies depending on the location of cysteine (C) between other amino acids (X) in the chemokine's protein molecule sequence:

(1)C   (2) CC   (3)CXC   (4) CX3C

For example, CXC chemokine can attract neutrophils but not macrophages, while **CC chemokine preferentially induce the migration of macrophages**. To date, about 40 chemokine have been identified in humans. They mainly act on **neutrophils**, **monocytes/macrophages**, **lymphocytes**, and **eosinophil** and play an **essential role in host defense mechanisms**. In addition, chemokines <u>regulate the lymphoid organ development</u>, the functioning of the nervous system, and may stimulate tumor cell metastasis.

Chemokine may be also functionally divided into two groups, **homeostatic** and **<u>inflammatory</u>**.

1. **<u>Homeostatic chemokine</u>**: are produced in **certain tissues and cells** <u>and are responsible homing leukocytes in particular organs or tissues. The homeostatic chemokine has many functions involved in the organogenesis, migration of progenitor cells, cell development, carcinogenesis and metastasis of cancer cells.</u>

2. **<u>Inflammatory chemokines</u>** <u>are constituted under pathological conditions due to pro- inflammatory stimuli from sites of infection, injury, or tissue damage and take an active part in the inflammatory response attracting a wide variety of cells in both the innate and adaptive immunity. Under the influence of inflammatory chemokine, cells will extravasation from the blood vessel and follow the gradient to pro-inflammatory stimuli. This group of chemokines is also recruited in wound healing.</u>

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Le No 9

Immune Responses: Primary and Secondary

Immune responses to antigens may be categorized as primary or secondary responses:

Primary Immune

Occurs when an antigen comes in contact with the immune system for the first time. Primary Immune Response which initiated against antigenic **stimulus is slow, sluggish, short live with a long lag phase and low antibody titer that do not persist for long time ,antibody formed are 1gM.** Depending on the nature of the antigen and the site of entry this response can take up to 14 days to resolve and leads to the generation of memory cells with a high specificity for the inducing antigen. The humoral response, mediated by B cells with the help of T cells, produces high-affinity and antigen-specific antibodies.

This is in contrast with the CD8 T-cell response which leads to the generation of large numbers of antigen-specific cells that are capable of directly killing infected cells. Antigen specific CD4 T cells, which provide help to B cells in the form of cytokines and other stimulatory factors, can also be expanded upon antigenic stimulation. The antibody classes start with 1gM followed by IgG and described as Antigen specific response.

Secondary Immune

Occurs when the second time (3rd, 4th, etc.) the person is exposed to the same antigen. At this point immunological memory has been established and the immune system can start making antibodies. The secondary response of both B and T cells is observed following subsequent encounter with the same antigen and is more rapid leading to the activation of previously generated memory cells
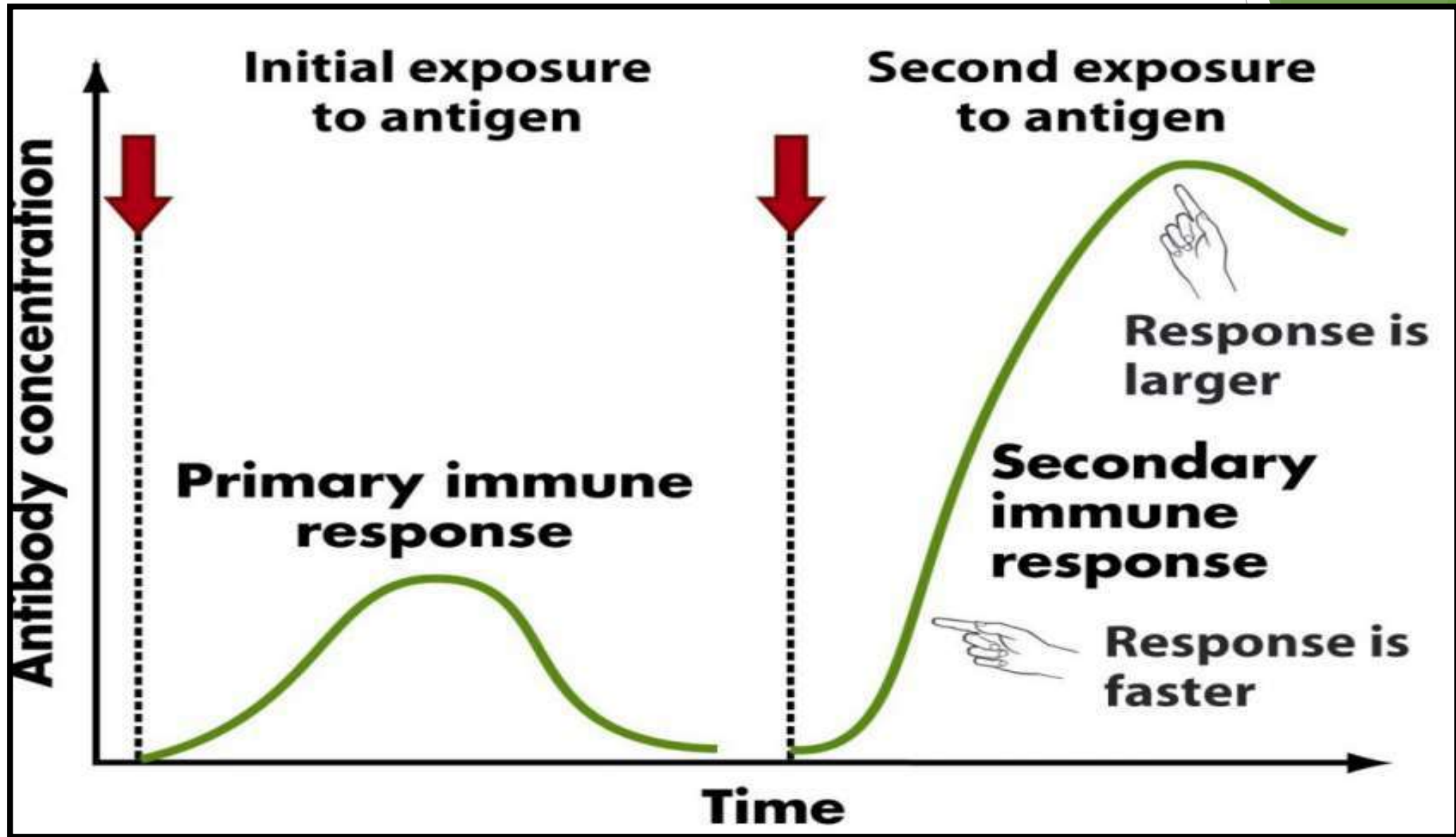
follow factor influencing Antibody production:

1. Age

2. Nutritional status

3. Route of administration

4. Size and Number of doses

5. Multiple antigens

6. Adjuvant

7. Immunosuppressive agent

Cells Involved in Primary and Secondary Immune Response

1. **Plasma Cells**

Fully differentiated B-lymphocyte which produces a single type of antibody.

1. **Memory B Cells**

B cell sub-type that are formed within germinal centers following primary infection and are important in generating an accelerated and more robust antibody-mediated immune response in the case of re-infection (also known as a secondary immune response).

Primary Immunodeficiency (PID)

PID disorders are inherited conditions, some PIDs are diagnosed during infancy or childhood, many are diagnosed later in life.

➤ **Causes**:

- Single-gene mutations

- Unknown genetic susceptibility combined with environmental factors.

➤ **Example**

- B cell immunodeficiencies (adaptive) – B cells are one of two key cell types of the adaptive immune system. Their main role is to produce antibodies, which are proteins that attach to microbes, making it easier for other immune cells to detect and kill them.

Secondary Immunodeficiency (SID)

SIDs are more common than PIDs and are the result of a primary illness, such as HIV, or other external factor such as malnutrition or some drug regimens .Most SIDs can be resolved by treating the primary condition.

➢ **Example:**

•Malnutrition: Protein-calorie malnutrition is the biggest global cause of SIDs which can affect up to 50% of the population in some communities in the developing world.

Compare Between Primary and Secondary Immune Response

| Character	Primary Response	Secondary Response
Lag Phase	Longer, (4-7 days) sometimes as long as weeks or months	Shorter (1-4 days) due to the presence of memory cell.
Antibody Level	Reaches peak in 7 to 10 days.	Reaches peak in 3 to 5 days.
Antibody Production	Mainly IgM, small amount of IgG are also produced .	Mainly IgG, sometimes small amount of IgM, IgA and in the case of allergy IgE are produced.
Amount of ab production	Depends on nature of antigen . Usually produced in low amount .	Usually 100-1000 times more antibodies are produced.
Ab -Ag affinity	Affinity of antibody is lower for its antigen	Antibodies have greater affinity for antigen.
Appearance	Appears mainly in the lymph nodes and spleen .	Mainly in the bone marrow, followed by spleen and lymph nodes
Antigen presentation	Mainly non-B cells, e.g. dendritic cells	B lymphocytes increasingly important

Class switching

The ability of antibody-producing cells to switch the class of antibody produced without affecting its specificity involves recombination events in the DNA coding for immunoglobulin. For example, changes in the class of antibody expressed, from IgM to the IgA1 subclass, are the direct effect of the enzymatic removal of the intervening sequences between the constant regions of IgM and IgA1 from the immunoglobulin heavy chain DNA. As a consequence, the genes coding for the constant heavy chain sequences of IgA1 become contiguous with those coding for the antibody-combining site.

Likewise, the expression of IgE is dependent on the removal of the DNA sequence coding for the constant heavy chain regions of IgM and IgD, all IgG subclasses and IgA1. As class switching occurs by deletion of intervening heavy chain gene sequences, the process is normally irreversible.

Class switching, like affinity selection also occurs in **the light zone area of germinal centers of secondary lymphoid tissue,** where close interactions between B cells and other cells take place. A key interaction is between a molecule on B cells known as CD40 and its ligand (CD40L or gp39) on activated T cells. This interaction results in B -cell proliferation and, in addition to

being crucial to isotype switching, is also important in affinity maturation and B -cell memory development.

The antibody class finally expressed is influenced by the cytokines produced locally, mainly by activated T cells .Thus in humans interferon g (IFNg) favours the production of antibody of the IgG1 subclass while interleukin-4 predisposes to the development of **IgG4 and IgE** .Cytokines influence class switching by inducing the selective transcription of germline transcripts upstream of the relevant switch regions in the heavy chain gene locus.
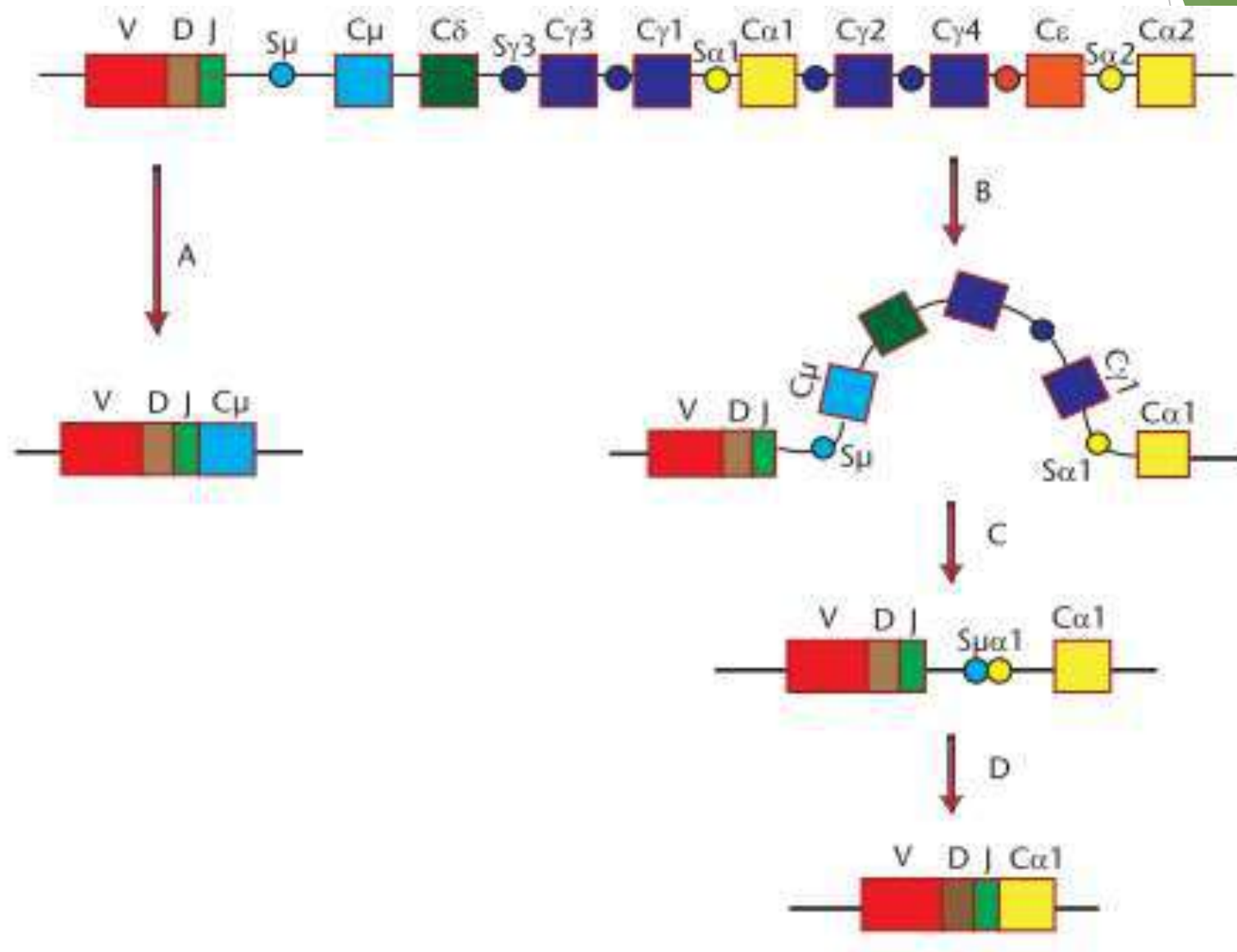
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Le No 8

1. Immunoglobulin M (IgM)

IgM is known as a **macroglobulin** because it has a has a molecular weight of approximately 900,000d. The half-life of IgM is about 6 days, much shorter than that of IgG. It accounts for between 5 %and 10% of all serum immunoglobulins.

IgM consists of about 576 amino acids and includes one more constant domain than is found on the γ chain. The **pentamer form** is found in **serum**, whereas the **monomer** form occurs on the surface of **B cells.**

The five monomeric units are held together by a J or joining chain, which is a glycoprotein made in plasma cells that contains several cysteine residues. These serve as linkage points for disulfide bonds between two adjacent monomers. The J chain also facilitates secretion at mucosal surfaces. One J chain is present per pentamer.

IgM thus configured assumes a **star-like** shape with 10 functional binding sites. The **Fab** arms can bend out of the plane to bind two or more separate antigens or multivalent antigens. The high valency of IgM antibodies helps to overcome the fact that they tend to have a low affinity for antigen.

- Because of its large size, IgM is found mainly in the intravascular pool and not in other body fluids or tissues.

- It cannot cross the placenta.

- **IgM** is known as the primary response antibody; it is the first to appear after antigenic stimulation and the first to appear in the maturing infant.

- It is synthesized only as long as antigen remains present because there are no memory cells for IgM. So that, it can be used to diagnose an acute infection, as its presence indicates a primary exposure to antigen.
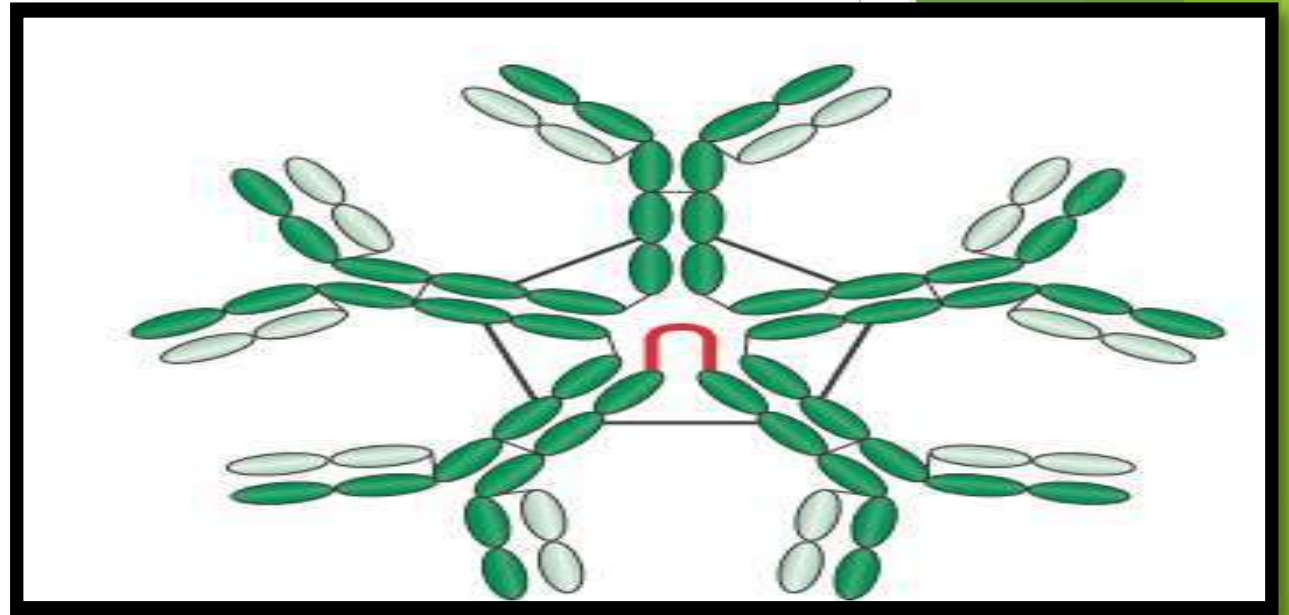
✤ **The functions of IgM include:**

1. Complement fixation

2. Agglutination

3. Opsonization

4. Toxin neutralization.

❖ IgM is the **most efficient** of all immunoglobulins at triggering the classical complement pathway because a single molecule can initiate the reaction as a result of its multiple binding sites. The larger number of binding sites also makes IgM more efficient at agglutination reactions, especially with multivalent antigens. Thus, IgM forms a potent defense against many bacterial diseases.

❖ IgM also serves as a surface receptor for antigen.
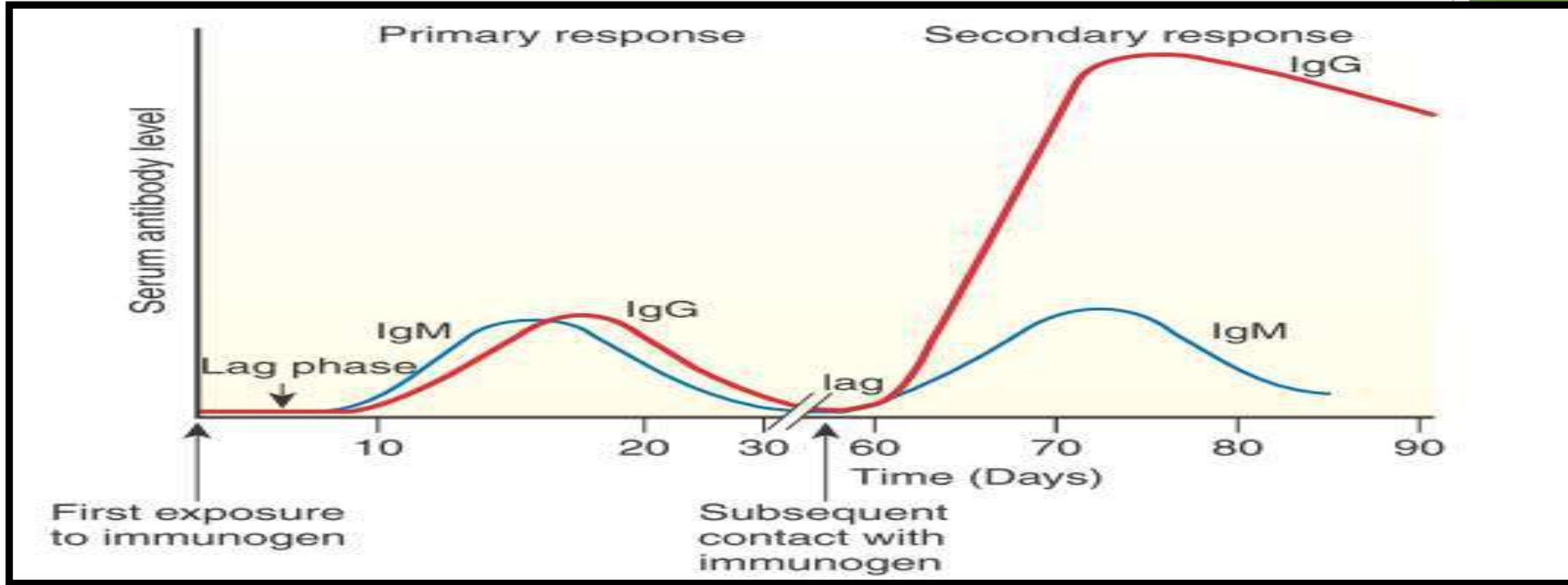
difference between the primary response, which is predominantly IgM, and the secondary response, which is mainly IgG. The primary response is characterized by a long lag phase, a slow increase in antibody, and a short-lived response. The second or anamnestic response is distinguished by a shortened lag period, a much more rapid rise in antibody, and higher serum levels for a longer period of time. The secondary response is the result of the larger number of antigen-specific memory T and B cells generated during the primary response.

1. Immunoglobulin A (IgA)

In the serum, IgA represents 10% to 15% of all circulating immunoglobulin. It appears as a monomer. **The H chain, called the α chain**, consists of about 472 amino acids. These amino acids comprise one variable and three constant regions.

➢ **Classes of IgA:**

➔ There are **two** subclasses, designated **IgA1** and **IgA2**.

➔ They differ in content by 22 amino acids, of which are located in the hinge region and are deleted in IgA2. **The lack of this region appears to make IgA2 more resistant to some bacterial proteinases that are able to cleave IgA1**.

➔ **IgA2** is the predominant form in **secretions at mucosal surfaces**, whereas **IgA1 is mainly found in serum**.

➔ **IgA2** is found as a dimer along the respiratory, urogenital, and intestinal mucosa; it also appears in breast milk, colostrum, saliva, tears, and sweat. Because mucosal surfaces are a major point of entry for pathogens, **IgA2** serves to keep antigens from penetrating farther into the body

The IgA dimer consists of two monomers held together by a J chain. The J chain is essential for the polymerization and secretion of IgA. Secretory IgA is synthesized in plasma cells found mainly in mucosal-associated lymphoid tissue and is released in dimeric form. IgA is synthesized at a much greater rate than that of IgG-approximately 3 grams per day in the average adult-but because it is mainly in secretory form, the serum concentration is much lower.

❖ **The main function of secretory IgA:**

1. Patrol mucosal surfaces and act as a first line of defense.

2. It plays an important role in neutralizing toxins produced by microorganisms.

3. Helps to prevent bacterial and viral adherence to mucosal surfaces.

4. The major role of serum IgA is as an anti-inflammatory agent.

Complexes of IgA and antigen are easily trapped in mucus and then eliminated by the ciliated epithelial cells of the respiratory or intestinal tract. This prevents pathogens from colonizing the mucosal epithelium. Because IgA is found in breast milk, breastfeeding helps to maintain the health of newborns by passively transferring antibodies and greatly decreasing infant death from both respiratory and gastrointestinal infections.

IgA is not capable of fixing complement by the classical pathway, although aggregation of immune complexes may trigger the alternate complement pathway.

1. Immunoglobulin D (IgD)

IgD was not discovered until 1965, when it was found in a patient with multiple myeloma, a cancer of the plasma cells.

➢ It is extremely rare in the serum, representing less than 0.001% of total immunoglobulins. It is synthesized at a low level and has a half-life of only 1 to 3 days. The molecule migrates as a fast γ protein.

➢ **The delta (δ) H chain** appears to have an extended hinge region consisting of 58 amino acids.

➢ Most of the IgD is found on the surface of immune-competent but unstimulated B lymphocytes.

➢ It is the second type of immunoglobulin to appear (IgM being the first) and it may play a role in B-cell activation

➢ The high level of surface expression and its intrinsic flexibility make it an ideal early responder to antigen. Those cells bearing only IgM receptors appear incapable of an IgG response, whereas those with both IgM and IgD receptors are capable of responding to T-cell help and switching to synthesis of IgG, IgA, or IgE.

➢ Thus, IgD may play a role in regulating B-cell maturation and differentiation.

• IgD does not appear to serve a protective function because it does not bind complement,

• It does not bind to neutrophils or macrophages

Because of its unusually long hinge region, IgD is more susceptible to proteolysis than other immunoglobulins. This may be the main reason for its short half-life.

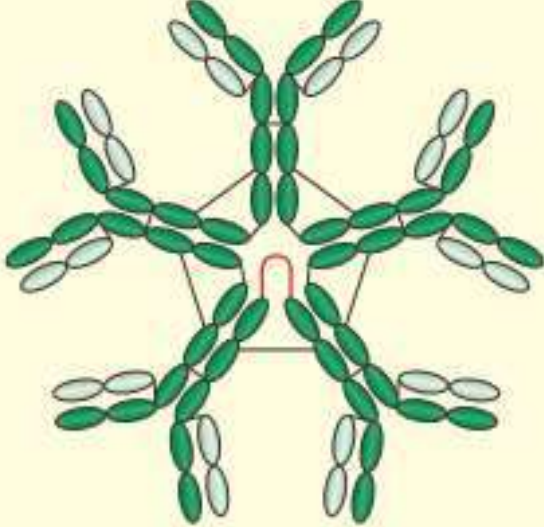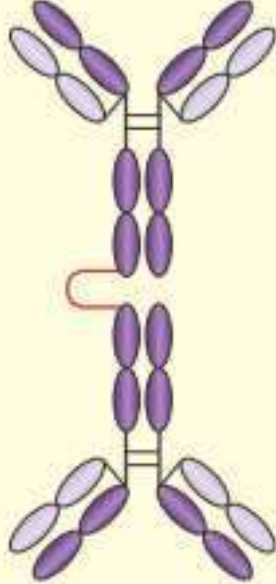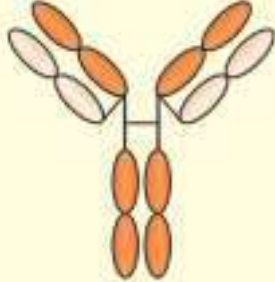**In the secreted form in the serum**:
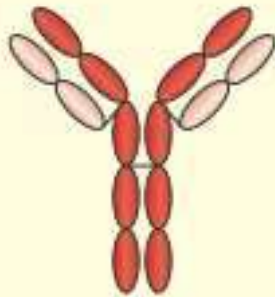
1. Immunoglobulin E (IgE)

IgE is best known for its very low concentration in serum and the fact that it has the ability to activate mast cells and basophils. It is the least abundant immunoglobulin in the serum, accounting for only 0.0005% of total serum immunoglobulins.

The **epsilon (ε)** or **H chain** is composed of around 550 amino acids that are distributed over one variable and four constant domains. A single disulfide bond joins each ε chain to an L chain and two disulfide bonds link the H chains to one another.

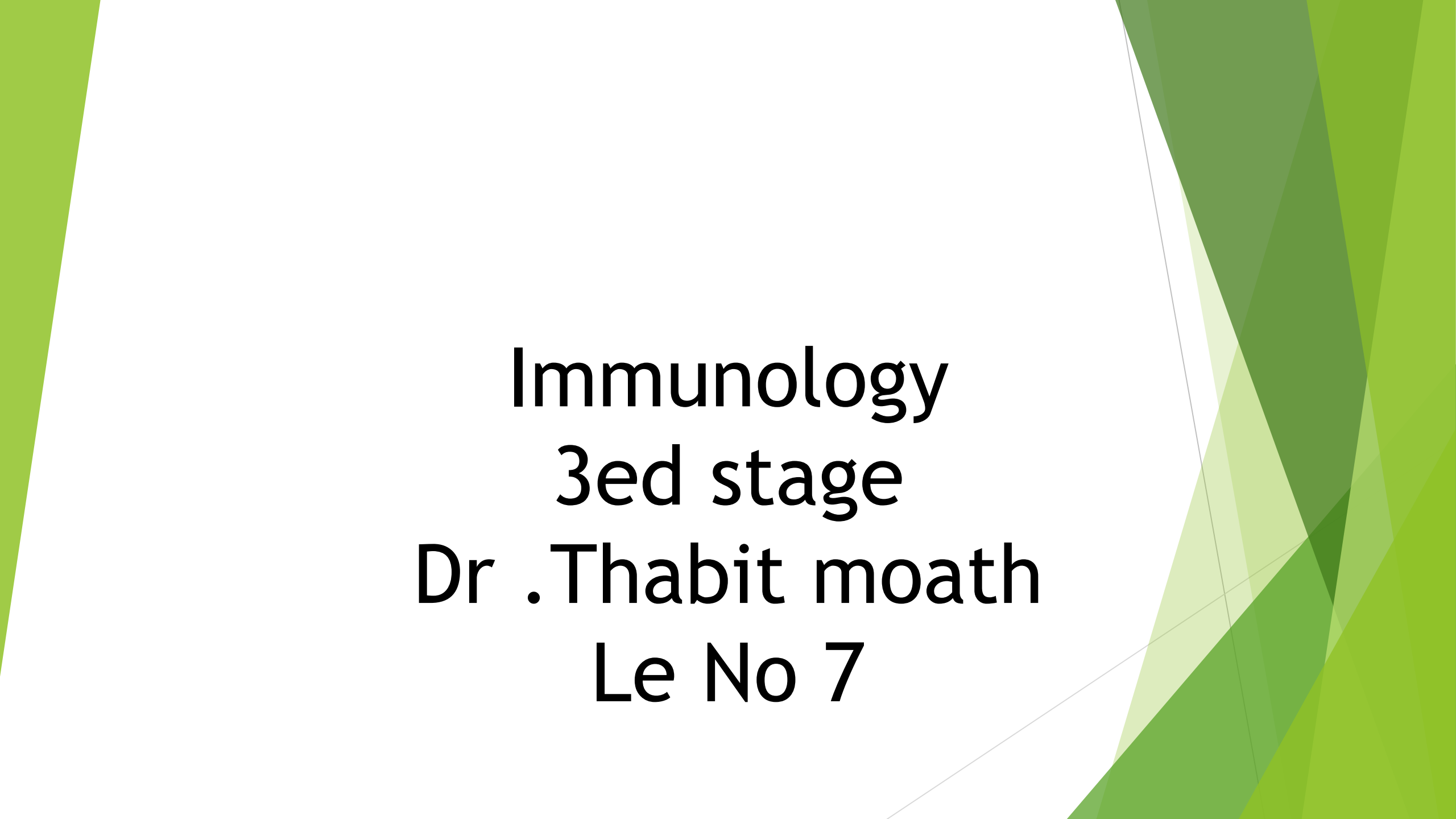- **IgE** is the most heat-labile of all immunoglobulins; heating to 56°C for between 30 minutes and 3 hours results in conformational changes and loss of ability to bind to target cells☞

- **IgE** does not participate in typical immunoglobulin reactions such as complement fixation, agglutination, or opsonization ☞

- It is incapable of crossing the placenta.

- Shortly after synthesis it attaches to basophils, Langerhans cells, eosinophils, and tissue mast cells by means of specific surface proteins, termed high-affinity FC ε RI receptors, which are found exclusively on these cells.

- Plasma cells that produce IgE are located primarily in the lungs and in the skin.

- Mast cells are also found mainly in the skin and in the lining of the respiratory and alimentary tracts. These cell may have several hundred thousand receptors, each capable of binding an IgE molecule. When two adjacent IgE molecules on a mast cell bind specific antigen, a cascade of cellular events is initiated that results in degranulation of the mast cells with release of vasoactive amines such as histamine and heparin. Release of these mediators induces what is known as a **type I immediate hypersensitivity** or **allergic reaction**.

- Typical reactions include hay fever, asthma, vomiting and diarrhea, hives, and life-threatening anaphylactic shock. Recently developed anti-IgE antibody that targets free IgE has been used as therapy for allergies and asthma.

- IgE appears to be a irritation antibody; however, it may serve a protective role by triggering an acute inflammatory reaction that recruits neutrophils and eosinophils to the area to help destroy invading antigens that have penetrated IgA defenses.

IgG	IgM	IgA	IgD	IgE
Most abundant in serum	Primary response antibody	Monomer and dimer	Present on B cells	Binds to mast cells
Able to cross placenta	Pentamer with 10 antibody-combining sites	Protects mucosal surfaces	Role in B-cell activation	Triggers allergic response
Increases with second exposure	Indicates acute infection	Has secretory component	Identifies mature B cells	Role in response to parasites

Feature	IgM	IgG	IgA	IgE	IgD
Form	Pentameric	Monomeric	Monomeric (serum IgA)or dimeric (secretory IgA)	Monomeric	Monomeric
Heavy chain	μ	γ	α	ε	δ
H chain subclasses	None	1 γ, 2 γ, 3 γ, 4 γ	1 α , 2 α	None	None
Accessory chain	J	-	Secretory component (SC)	-	-
Subisotypes (subclasses)		1 (65%), 2(20%) 3 (10%) 4 (5%)	1 2	-	-
Half-life (days)	6	23	5	1-3	2-3
Serum concentration in healthy adults (g/L)	0.6-2.0	8-16	0.7-3.0	0.003	0.04
Placental transfer	-	+	-	-	-
complement fixation	yes	yes	no	no	no

Immunology
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Le No 7

Antibodies

❖ **Introduction:-**

Antibodies are protective circulating glycoproteins that are produced in vertebrates in response to exposure to **antigens**. When B lymphocytes are stimulated by antigen and undergo differentiation, the end product is an **antibody**, also known as an **immunoglobulin**. Antibodies are extremely diverse and specific in their ability to recognize foreign molecular structures. Because these protein were discovered as serum molecules that provided protection against diphtheria toxin, they were initially called **antitoxins**. When it was appreciated that similar proteins could be generated against many substances, not just microbial toxins, they were given the general name **antibodies**.

Immunoglobulin is considered to be the main humoral element of the adaptive immune response. They play an essential role in antigen recognition and in biological activities related to the immune response such as opsonization and complement activation. They are divided into five major classes on the basis of a part of the molecule called the **heavy chain**. These classes are designated as **IgG**, **IgM**, **IgA**, **IgD**, and **IgE** , and the heavy chains are γ, μ, α, δ, and ε.

✳ **Characteristics of Antibodies:-**

1. **Affinity** is the quality of an immunoglobulin molecule to be bound to antigen strongly on the base of close agreement of their specificities.

2. **Avidity** is the same quality based on polyvalence of the antigen-binding sites.

3. **Cross-reactivity** is the ability of one antibody to bind to different antigenic epitopes.

4. **Isotype** is a diversity of antibodies inside a species. In humans, there are IgM, IgD, IgG, IgA, and IgE isotypes or classes.

5. **Allotype** is an individual antibody diversity based on the inheritance of different alleles.

6. **Idiotype** is a clonal antibody diversity.
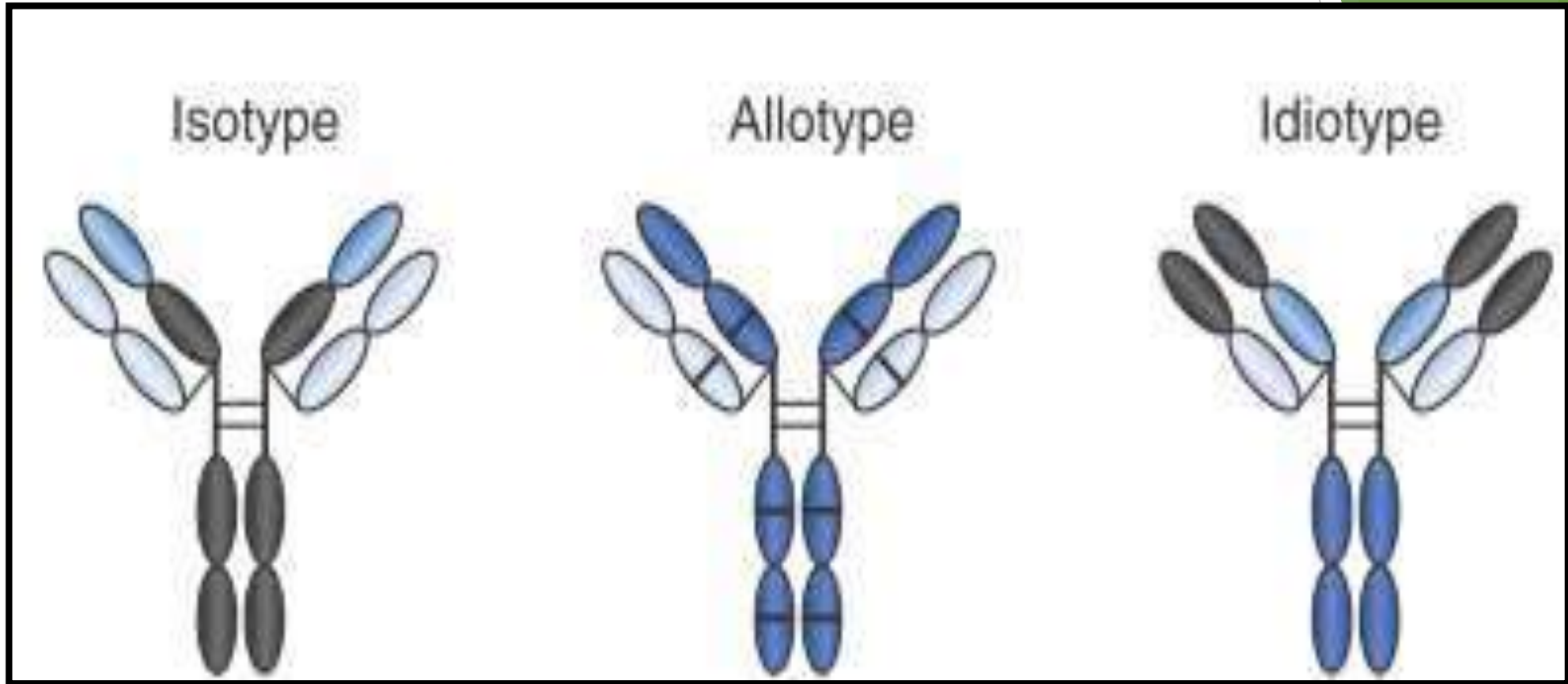
Antibody variations (shown in black). (A) Isotype-the H chain that is unique to each immunoglobulin class. (B) Allotype-genetic variations in the constant regions. (C) Idiotype-variations in variable regions that give individual antibody molecules specificity.

Variable domains (V)

Antigenbinding site

Fab

NH₂

VH

VL

S. S.

CH1

CL

κ, λ

Light chain (L)

CH2

Heavy chain (H)

μ,γ,α,ε,δ

FC {

CH3

Constant domains (C)

COOH
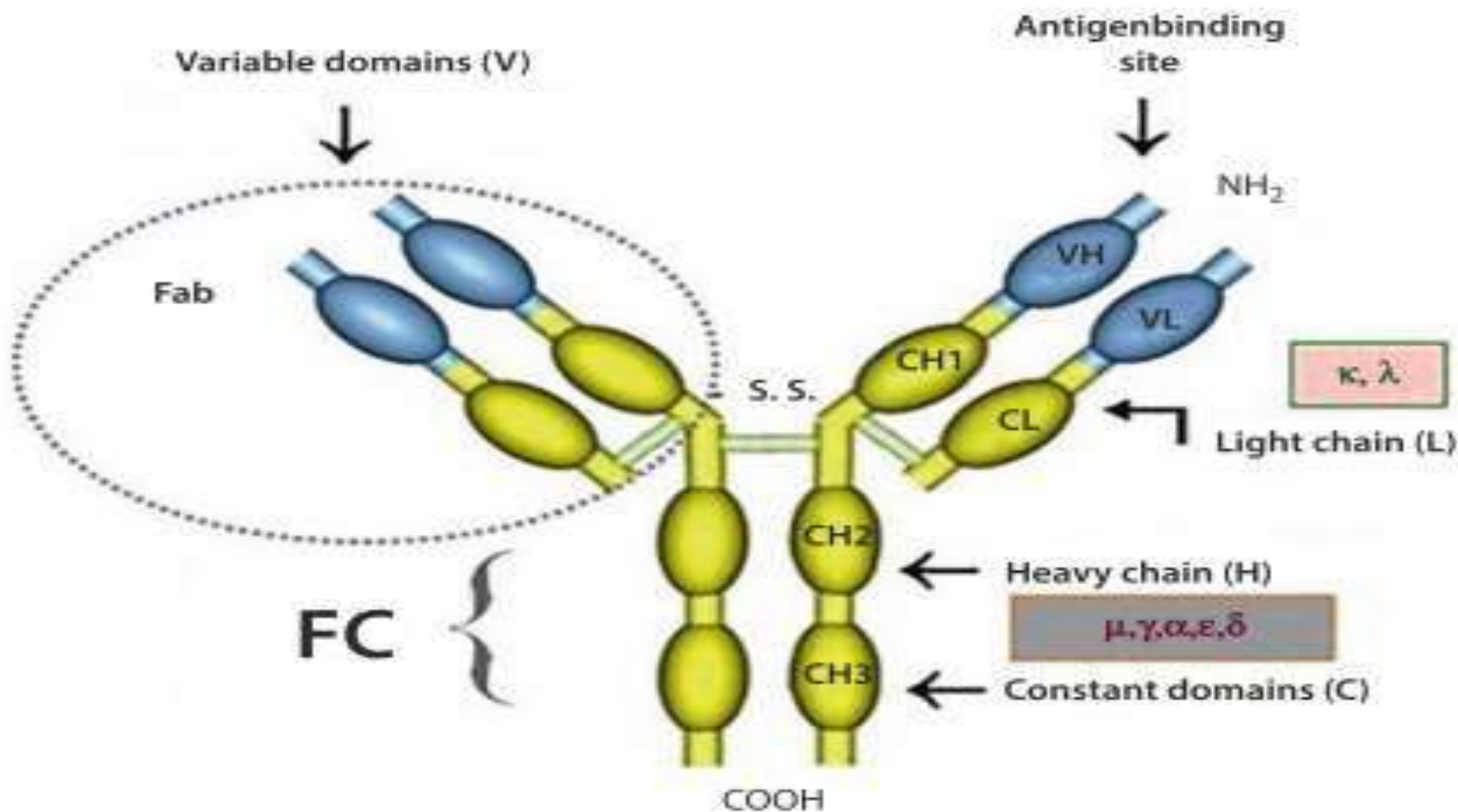
❋ **Structure of Immunoglobulins:-**

All immunoglobulin molecules are made up of a **basic four chain tetra peptide unit** that consists of two large chains called **Heavy or H chains** and two smaller chains called **Light or L chains**. Each chain has a **single amino-terminal variable region (V)** that participate in antigen recognition and **one or more carboxy-terminal constant regions**. The variable region is unique to each specific antibody. These chains are held together by non-covalent forces and disulfide inter-chain bridges.

Both the light chains and heavy chains contain a series of repeating homologous structural units, each about 110 amino acid residues in length, that fold independently in a globular motif that is called an **Ig domain**

❖ **The constant region**:- located at the **carboxy-terminal end** of the molecule and named **the Fc (fragment crystallize) region**, had no antigen-binding ability and is now known to represent the carboxy-terminal halves of two H chains that are held together by **S–S bonding**. **The Fc fragment** is important in effector functions of immunoglobulin molecules, which include **opsonization**, **complement fixation** and **binding to effector cells** such as neutrophils, basophils, eosinophils, and mast cells. **The variable region**:- is at the amino-terminal end, called the **Fab fragment (fragment antigen binding)** this determines the specificity of that molecule for a particular antigen.
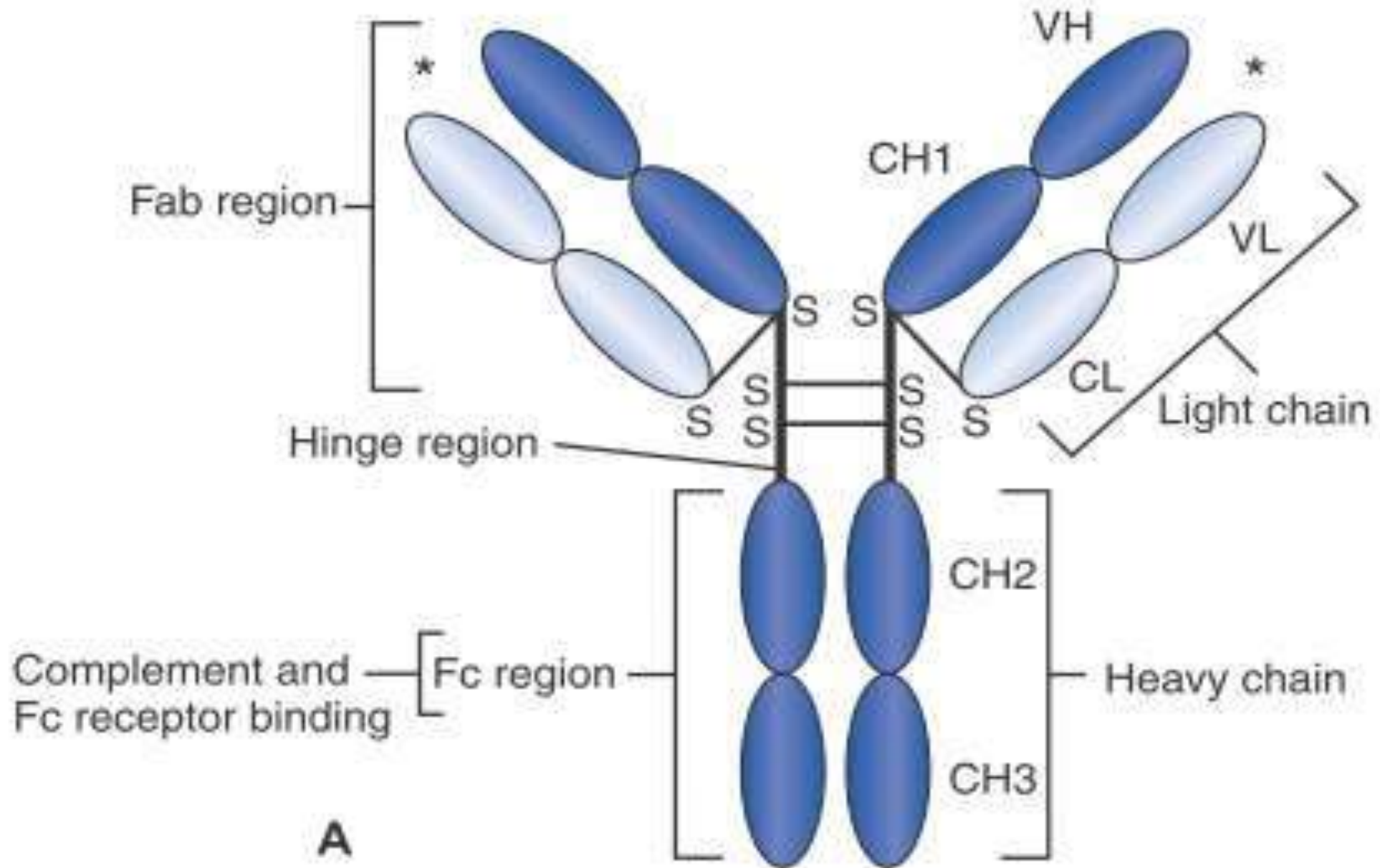
❖ **Light Chain:-**

    L chains, which were designated **kappa (κ) chains and lambda (λ) chains**. Each contained between 200 and 220 amino acids; from position number 111 onward (the amino terminus is position number 1), it was discovered that each type had essentially the same sequence. This region was called the **constant region** and the amino-terminal end was called the **variable region.**

❖ **Heavy Chain:-**

The first approximately 110 amino acids at the amino-terminal end constitute the variable domain and the remaining amino acids can typically be divided up into three or more constant regions with very similar sequences, designated CH1, CH2, and CH3. Constant regions of the H chain are unique to each class and give each immunoglobulin type its name.

- IgG has γ H chain

- IgM has μ chain

- IgA has α chain

- IgD has δ chain

- IgE has ε chain

- These five different types of heavy chains are called isotypes.

- Minor variations in a particular type of heavy chain are called allotypes.

The variable portion of the light and heavy chains unique to a particular immunoglobulin molecule is known as the Idiotype

✳ **Hinge Region**

The segment of H chain located between the CH1 and CH2 regions is known as **the hinge region**. It has a high content of proline and hydrophobic residues; the high proline content allows for flexibility. This ability to bend lets the two antigen-binding sites operate independently and engage in an angular motion relative to each other and to the Fc stem.

Types of Immunoglobulin:

1. Immunoglobulin G (IgG):-

IgG is the predominant immunoglobulin in humans, comprising approximately 70% to 75% of the total serum immunoglobulins. IgG has the longest half-life of any immunoglobulin class, approximately 23 days .There are **four major subclasses** with the following distribution: **IgG1**, 66%; **IgG2**, 23%; **IgG3**, 7%; and **IgG4**, 4%. These subclasses differ mainly in the number and position of the disulfide bridges between the **γ chains**.

Variability in the hinge region affects the ability to reach for antigen and the ability to initiate important biological functions such as complement activation. **IgG3** has the <u>largest</u> hinge region and the largest number of inter chain disulfide bonds; therefore, it is the most efficient at binding complement, followed by **IgG1**, **IgG2** and **IgG4** have <u>shorter</u> hinge segments.

All subclasses have the ability to cross the placenta except IgG2.

❖ **Major functions of IgG include the following :**

1. Providing immunity for the newborn because IgG is the only antibody that can cross the placena

2. Fixing complement

3. Coating antigen for enhanced phagocytosis (opsonization)

4. Neutralizing toxins and viruses

5. Participating in agglutination and precipitation reactions

All subclasses are able to participate in the secondary immune response, an enhanced and quicker response to antigen, although their appearance depends on the triggering antigen. **IgG1and IgG3** are induced in response to **protein antigens**, whereas **IgG2 and IgG4** are associated with **polysaccharide antigens**.
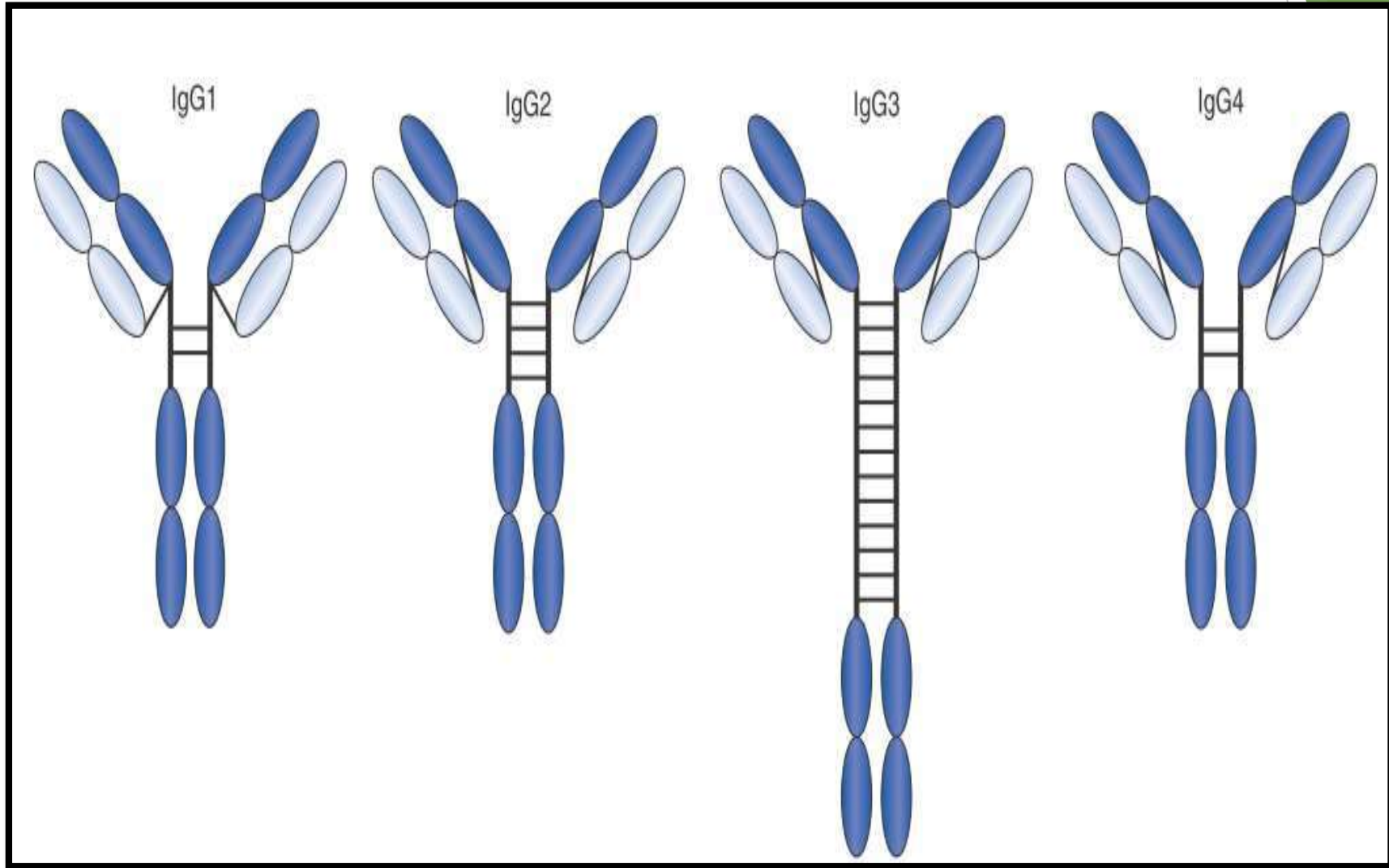
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Le No 6

✳ <u>Types of antigen on the basis of source</u> Antigens are categorized into broad classes based on their origin.

Based on the Source Antigens, they are classified into:

1. **Exogenous (entering from outside) 2Endogenous (generated within cells) 3Auto antigen4Tumor antigen 5Native antigen**

1. <u>**Exogenous Antigens**</u>

antigens entered the body **from the outside**, for example by <u>inhalation</u>, <u>ingestion</u>, or <u>injection.</u> Exogenous antigens are the most common kinds of antigens, and includes <u>pollen</u> or <u>foods</u> that may <u>cause allergies</u>, as well as the molecular components of <u>bacteria and other pathogens</u> that could cause an <u>infection</u>.

1. <u>**Endogenous Antigens**</u>

antigens are that have been generated within **previously-normal cells** as a result of **normal cell metabolism** or because of <u>viral or intracellular</u> <u>bacterial infection</u> (which both change cells from the inside in order to reproduce). The fragments are then presented on the surface of the infected cells in the complex **<u>with MHC class I molecules</u>**.

<u>Some antigens can be exogenous in origin but later can turn endogenous. E.g. Intracellular viruses.</u>

1. **Autoantigens**

Auto-antigens are normal "self" protein or complex of proteins or nucleic acid that is attacked by the host's immune system, causing an autoimmune disease. These antigens should, under normal conditions, not be the target of the immune system, but due to mainly **genetic** and **environmental factors**, the normal immunological tolerance for such an antigen has been lost.

1. **Tumor Antigens (Neoantigens)**

antigens result from a tumor-specific mutation during malignant transformation of normal cells into cancer cells. Despite expressing this antigen, many tumors have developed ways to evade antigen recognition and immune system killing.
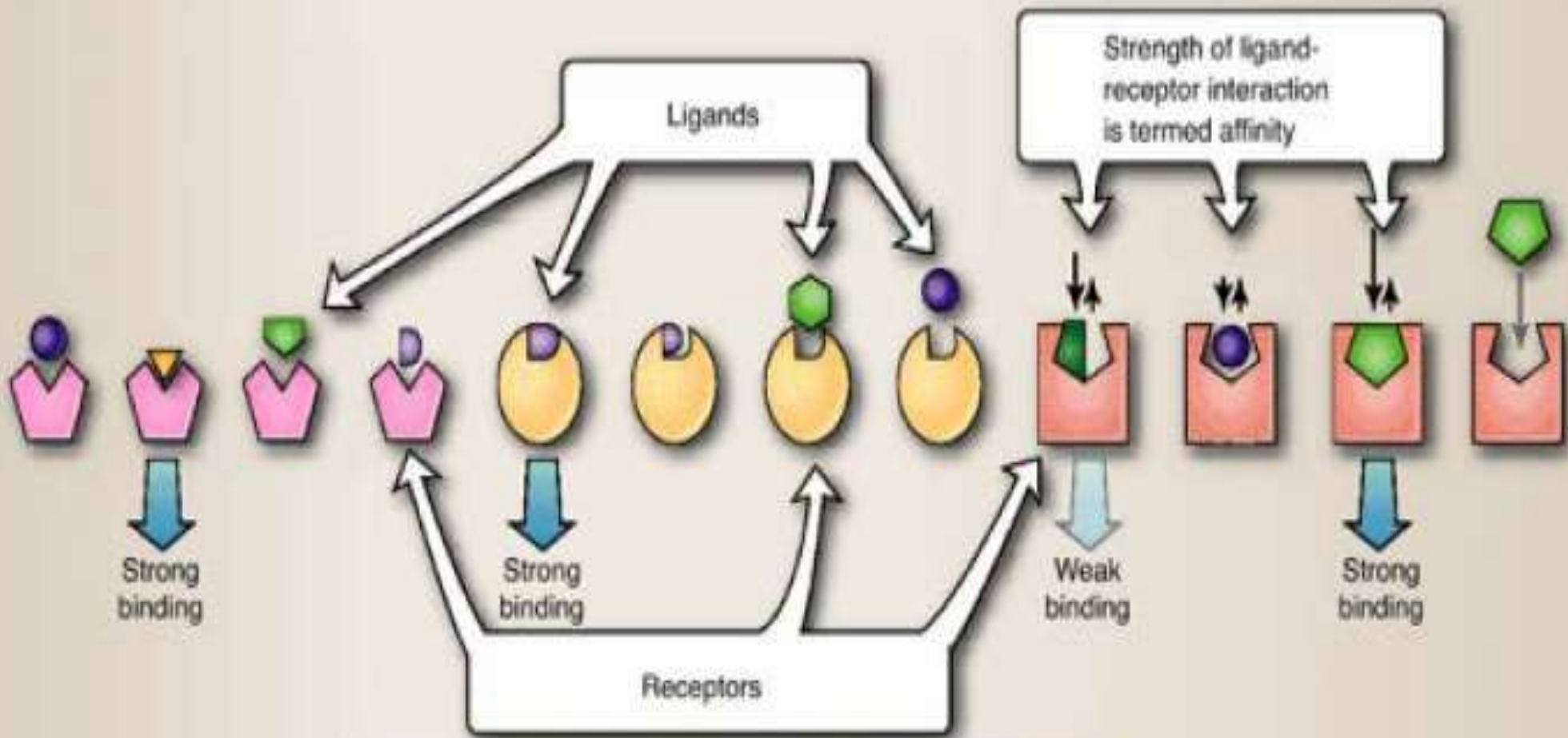
1. **Native Antigens**

A native antigen is an antigen that is not yet processed by an APC to smaller parts. T cells cannot bind native antigens, but require that they be digested and processed by APCs, **whereas B cells can be activated by native ones without prior processing**.

✳ **Classification of antigens On the basis of immune response**

Immune responses are initiated by the interaction between a **receptor** and a **ligand** (a molecule that interacts with a receptor). These interactions are what trigger the activation of leukocytes or white blood cells and the shapes of the ligand and its receptor are **critical** and **specific**. The effectiveness of interaction often increases with the affinity or strength of interaction between ligand and receptor.

Several factors influence the binding of a ligand to a cell-surface receptor:

1. The shape and charge affect binding **affinity**

2. The collective affinities where multiple receptors may be involved (**avidity**).

3. The intracellular signals that are triggered

4. The presence of other receptors that may also influence the action of immune response.

Ligands

Strength of ligand-receptor interaction is termed affinity

Strong binding

Strong binding

Weak binding

Strong binding

Receptors

Receptors must bind soluble or membrane-bound molecules of appropriate shape

Different lymphocytes, each with a unique set of receptors, may recognize different epitopes on the same antigen. Some receptors (**e.g., those of B cells**) can recognize their specific epitopes whether they are **part of free soluble molecules**, **surface-bound molecules**, or even degraded (**proteolytic) fragments of antigens**. Other receptors (e**.g., T cell receptors**) can bind only to epitopes that are on small fragments affixed to specialized host cell surface molecules that display them to the T cells. Depending upon the nature of the immune responses they trigger, antigens/epitopes are divided into three broad functional types: **immunogens**, **haptens**, and **tolerogens**.

A. Immunogens (complete Antigens) **An immunogen is an antigen** or any substance that may be specifically bound by components of the immune system (antibody, lymphocytes).

Characteristic of immunogens:

1. Have antigenic properties by their ability to generate an immune response by themselves.
2. High molecular weight (more than 10,000 d)
3. May be proteins or polysaccharides

A. Haptens (incomplete antigens)

Haptens are small, normally non-immunogenic, molecules, usually of non-biologic origin .Haptens are antigens and can bind to immune receptors but cannot by themselves induce a specific immune response and hence are not immunogenic. When a molecule of haptens are coupled to **carrier proteins** they become accessible to immune system and **function as an immunogen**, and the immune responses may **be generated against both the hapten and the epitopes** on the **immunogen**.

They generally have low molecular weight (Less than 10,000 Daltons) and are usually non-protein substances. E.g. **polysaccharide "C" of β-haemolytic streptococci**.

Carrier molecule is a non-antigenic component and helps in provoking the immune response.

Hapten molecules + Carrier molecule → Complete antigen
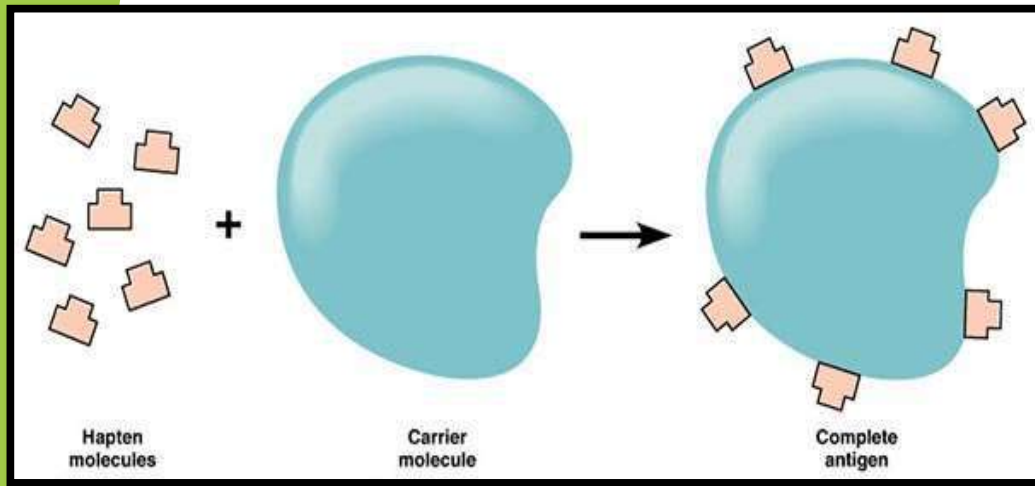
Determinants of Antigenicity

The whole antigen does not activat immune response and only a small part of it induces B and T cell response.

The small area of chemical grouping on the antigen molecule that determines specific immune response and reacts specifically with antibody is called an ***antigenic determinant.***

Immunogenicity: the **ability** of immunogen **to stimulate** humoral and/or cell mediated immunity response.

✤ **<u>Characteristics of Immunogenicity</u>**

1. **Macromolecular size:**

Usually an immunogen must have a large molecular weight of at least 10000 daltons

Examples: tetanus toxoid, egg albumin, thyroglobulin are highly antigenic.

Insulin (5700) are either non-antigenic or weakly antigenic.

1. **Foreignness:**

All immunogens share is foreignness.

1. **Chemical composition and molecular complexity**

Immunogenicity is also determined by a substance's chemical composition and molecular complexity.

Proteins and **polysaccharides** are the most effective immunogens.

Proteins are powerful immunogens because they are made up of a variety of units known as amino acids. The particular sequential arrangement of amino acids, the primary structure, determines the secondary structure, which is the relative orientation of amino acids within the chain.

Carbohydrates are less immunogenic than protein because they are **smaller t**han proteins and have a limited number of sugars available to create their structures. Other carbohydrates that are important immunogens are the capsular polysaccharides of bacteria such as **Streptococcus pneumoniae.**

Pure nucleic acids and **lipids** are not immunogenic by themselves, although a response can be generated when they are attached to a **suitable carrier molecule**. This is the case for autoantibodies to DNA that are formed in **systemic lupus erythematosus (SLE)**. These autoantibodies are actually stimulated by a DNA-protein complex rather than by DNA itself.

1. **The ability to be processed and presented with MHC molecules**.

For a substance to elicit an immune response it must be subject to antigen processing, which involves enzymatic digestion to create small peptides or pieces that can be complexed to MHC molecules to present to responsive lymphocytes. If a macromolecule cannot be degraded and presented with MHC molecules, then it would be a poor immunogen.

1. **Physical Form**

- In general particulate antigens are more immunogenic than soluble ones.
- (abnormal shape) antigens are more immunogenic than the native form.

1. **Antigen Specificity**

Antigen Specificity depends on the specific actives sites on the antigenic molecules (Antigenic determinants).

1. **Auto-specificity**

The autologous or self-antigens are ordinarily not immunogenic, but under certain circumstances lens protein, thyroglobulin and others may act as *autoantigens*.

1. **Genetic Factors**

2. **Age**

1. **Degradability:**

Antigens that are easily phagocytosed are generally more immunogenic. This is because for most antigens (T-dependant antigens) the development of an immune response requires that the antigen be phagocytized, processed and presented to helper T cells by an antigen presenting cell (APC).

1. **Dose of the antigen**

The dose of administration of an immunogen can influence its immunogenicity. There is a dose of antigen above or below which the immune response will not be optimal.

1. **Route of Administration**

Generally the subcutaneous route is better than the intravenous or intragastric routes. The route of antigen administration can also alter the nature of the response. Antigen administered intravenously is carried first to the spleen, whereas antigen administered subcutaneously moves first to local lymph nodes.

❋ Types of Antigenic Determinants

The epitopes of protein antigens are divided into two categories based on their structures and interaction with the paratope.

1. **Conformational epitope** is composed of discontinuous sections of the antigen's amino acid sequence. These epitopes interact with the paratope based on the 3-D surface features and tertiary structure (overall shape) of the antigen. Most epitopes are conformational.

2. **Linear epitopes** interact with the paratope based on their primary structure (shape of the protein's components). A linear epitope is formed by a continuous sequence of amino acids from the antigen, which creates a "line" of sorts that builds the protein structure.

Epitopes recognized by B cells may differ from those recognized by T cells. Surface antibody on B cells may react with both linear and conformational epitopes present on the surface of an immunogen. Anything that is capable of cross-linking surface immunoglobulin molecules is able to trigger B-cell activation. The immunogen does not necessarily have to be degraded first. However, for T cells to be able to recognize an immunogen it must first be degraded into small peptides by an antigen-presenting cell (APC). Then the peptides form a complex with MHC proteins and are carried to the surface of the APC.
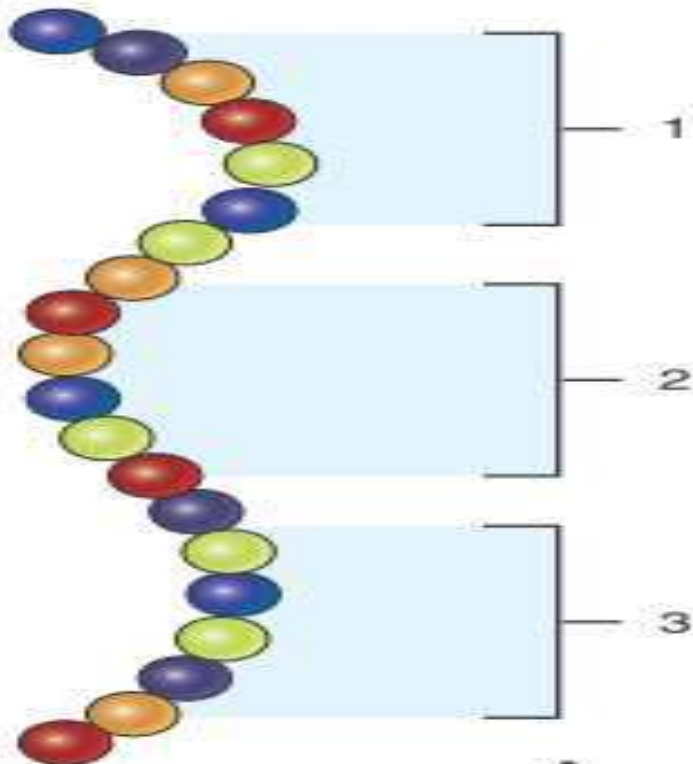
Linear epitopes   A
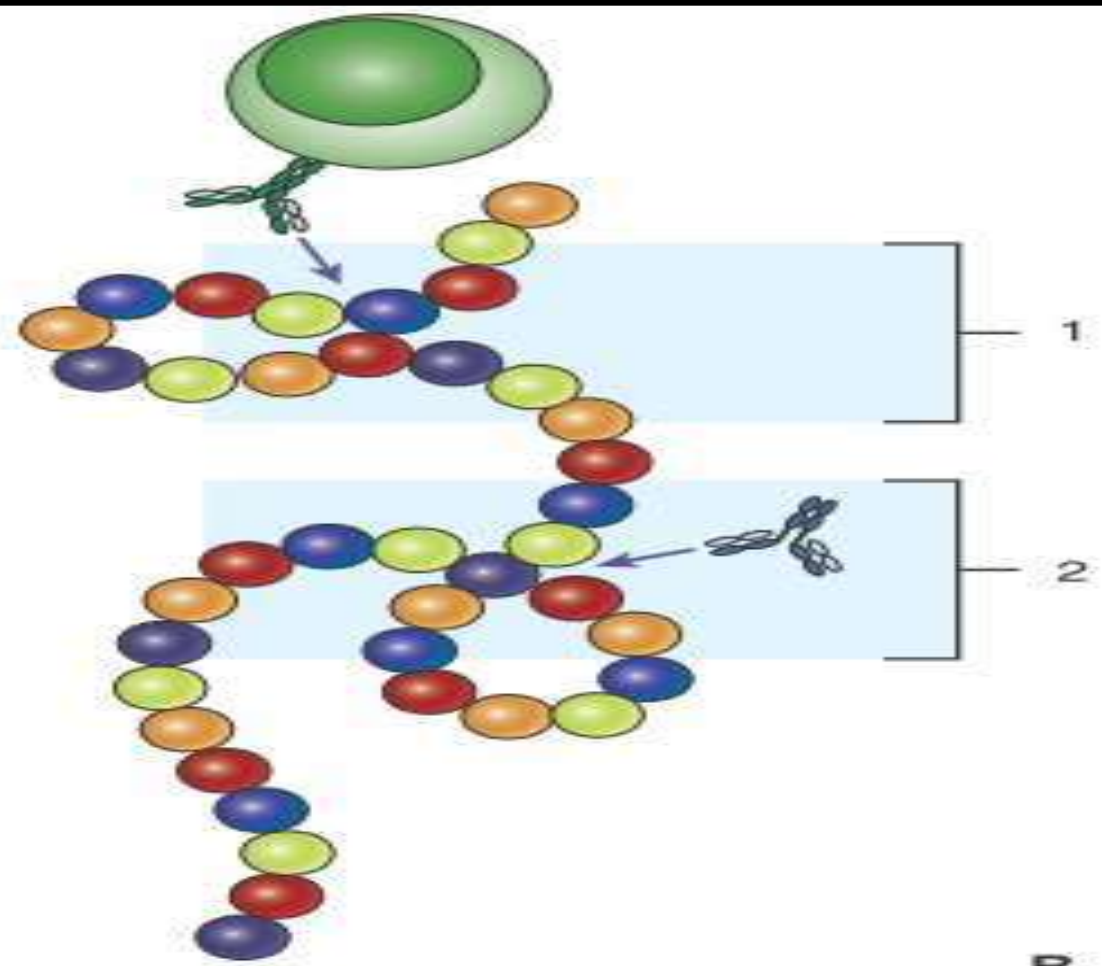
Conformational epitopes   B

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1. Inflammation

When pathogens breach the outer barriers of innate immunity, both cellular and humoral mechanisms are involved in a complex, highly coordinated process known as **inflammation**. **Inflammation** can be defined as the body's overall reaction to injury or invasion by an infectious agent.

The four cardinal signs or clinical symptoms of inflammation are **redness** (erythema), **swelling** (edema), **heat**, and **pain**.

Major events that occur rapidly after tissue injury are:

1. **Increased blood supply to the affected area**. Dilation of the blood vessels caused by the release of chemical mediators such as histamine from injured mast cells brings additional blood flow to the affected area, resulting in redness and heat.

2. **Increased capillary permeability** caused by contraction of the endothelial cells lining the vessels. The increased permeability of the vessels allows fluids in the plasma to leak into the tissues, resulting in the swelling and pain associated with inflammation.

3. **Migration of WBCs,** mainly neutrophils, from the capillaries to the surrounding tissue in a process called **diapedesis**. Soluble mediators, which include acute-phase reactants, **chemokines**, and **cytokines**, act as chemo-attractants to initiate and control the response.

4. **Migration of macrophages** to the injured area. Migration of macrophages and dendritic cells from surrounding tissue

5. Acute-phase reactants stimulate phagocytosis of microorganisms. Macrophages, neutrophils, and dendritic cells all attempt to clear the area through phagocytosis, in most cases, the healing process is completed with a return of normal tissue structure .

The acute inflammatory response acts to combat the early stages of infection and also begins a process that repairs tissue damage. However, when the inflammatory process becomes prolonged, it is said to be chronic. The failure to remove microorganisms or injured tissue may result in continued tissue damage and loss of function.
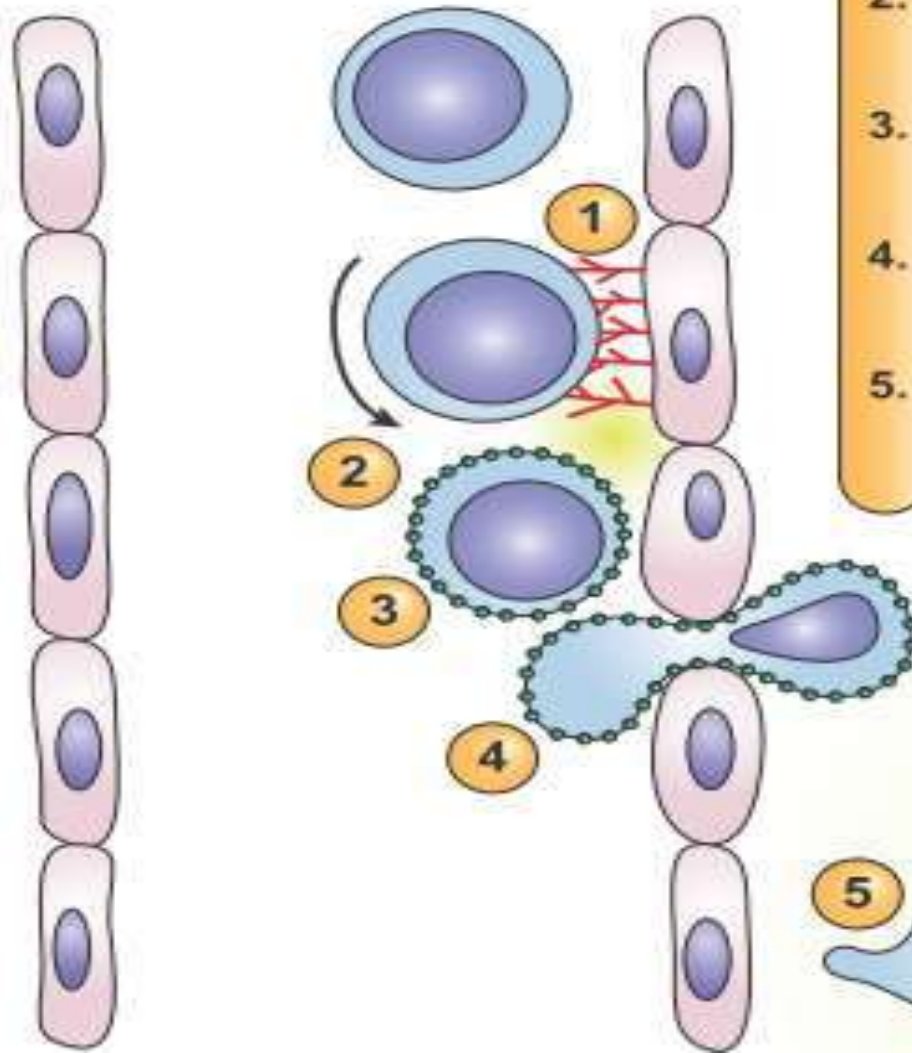
1. Resident macrophages and mast cells at the site of infection release chemokines that cause vasodilation and induce selectins.

2. Selectins loosely bind circulating leukocytes and cause them to roll along the vascular wall.

3. Chemokine-induced integrins on the leukocytes bind firmly to the endothelial cells and

4. Integrins enable the leukocytes to crawl between endothelial cells (diapedesis).

5. Leukocytes then follow the chemokine concentration gradient to the site of infection (chemotaxis).

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chronic inflammation response:

chronic inflammation are characterized   by :

1-   which lasts at least two weeks.

 2--   a slow process

3-   formation of new connective tissue.

4-   causes permanent tissue damage.
5-   without acute inflammation.

persistence of bacteria can stimulate chronic inflammation. For example, *mycobacterium tuberculosis*, have cell walls with a very high lipid and wax content, making them resistant to phagocytosis and intracellular killing. also, these bacteria and some protozoa such as Leishmania, can survive within macrophages. If macrophages fail to protect the host from tissue damage, the body attempts to wall off and isolate infected site by crowded of infiltrated lymphocytes and macrophages and forming **a granuloma.**

granuloma (Latin, granulum, a small particle; Greek, oma , to form), Granulomas are formed when neutrophils and macrophages are unable to destroy the microorganism during inflammation, such as Infections by some microbial pathogens, helminth parasites and large antibody-antigen complexes (as in rheumatoid arthritis) causes granuloma formation. (Granuloma: Term applied to nodular inflammatory lesions containing phagocytic cells
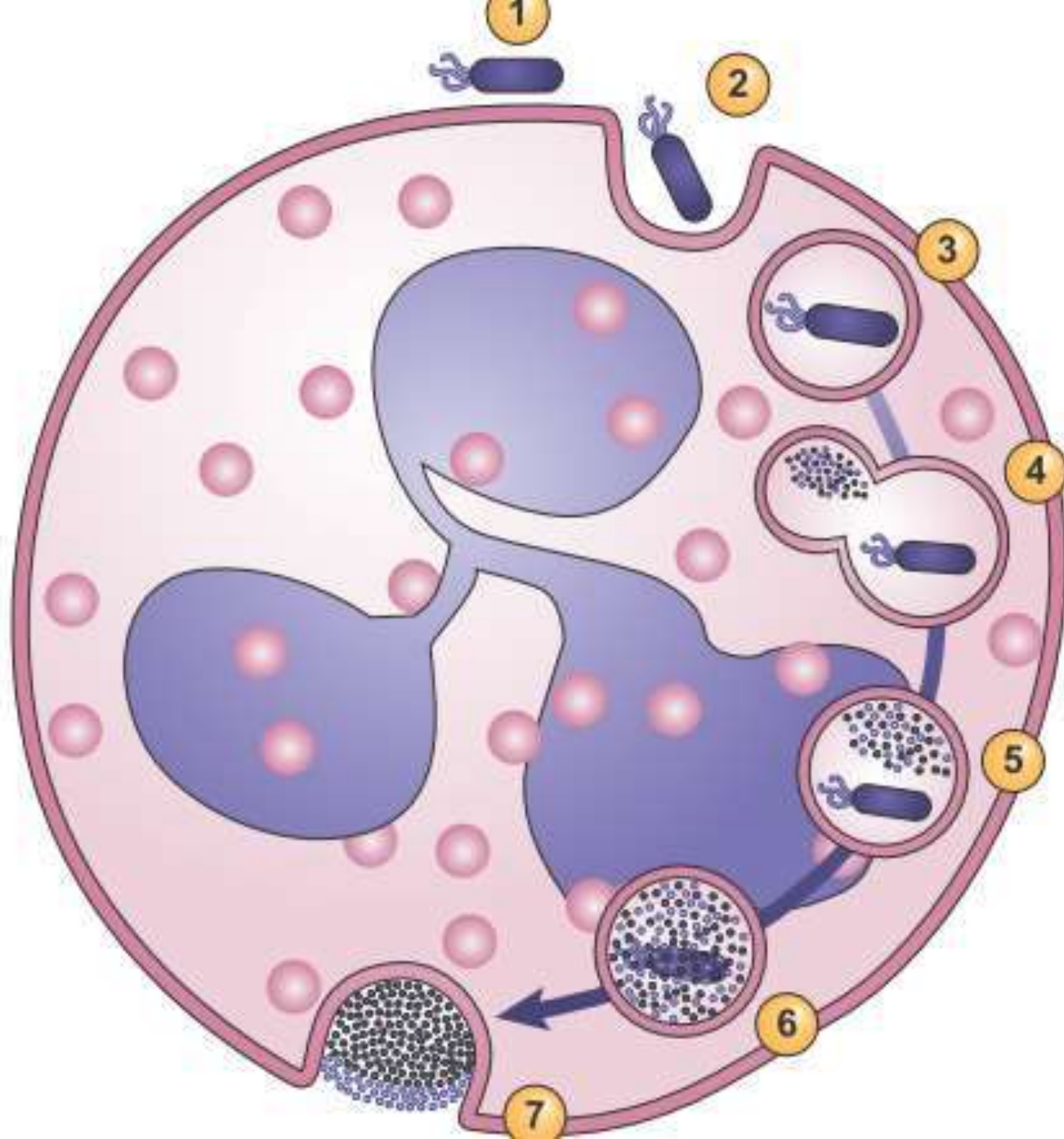
1. Adherence: physical contact between the phagocytic cell and the microorganism occurs, aided by opsonins.

2. Engulfment: outflowing of cytoplasm to surround the microorganism.

3. Formation of phagosome: microorganism is completely surrounded by a part of the cell membrane.

4. Granule contact: lysosomal granules contact and fuse with the phagosome.

5. Formation of the phagolysosome: contents of the lysosome are emptied into this membrane-bound space.

6. Digestion of the microorganism by hydrolytic enzymes.

7. Excretion: contents of phagolysosome are expelled to the outside by exocytosis.

1. Phagocytosis

The main purpose of the inflammatory response is to attract cells to the site of infection and remove foreign cells or pathogens by means of **phagocytosis**. Although the acute-phase reactants enhance the process of phagocytosis, it is the cellular elements of the internal defense system that play the major role. The cells that are most active in phagocytosis are neutrophils, monocytes, macrophages, and dendritic cells.

Once the WBCs are attracted to the area, the actual process of phagocytosis consists of 7*main steps:*

1. **Adherence**: physical contact between the phagocytic cell and the microorganism occurs, aided by opsonin's.

2. **Engulfment**: outflowing of cytoplasm to surround the microorganism.

3. **Formation of phagosome**: microorganism is completely surrounded by a part of the cell membrane.

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Opsonins, a term derived from the Greek word meaning "to prepare for eating." **Opsonins** are serum proteins that attach to a foreign cell or pathogen and help prepare it for phagocytosis. Opsonins may act by neutralizing the surface charge on the foreign particle, making it easier for the cells to approach one another.

Phagosome: After attachment to a foreign cell or pathogen has occurred, the cell membrane invaginates and pseudopodia (outflowing of cytoplasm) surround the pathogen. The pseudopodia fuse to completely enclose the pathogen, forming a structure known as a Phagosome

❋ **Antigen**: is a molecule that initiates the production of an antibody and causes an immune response. Antigens may be originated from "non-self," "former self," and even "self."

❋ **Auto-antigen**: Any antigen that stimulates auto-antibodies in the organism that produced it. These are "self" antigens that are involved in autoimmune disease pathogenesis.

❋ **Epitope:** is immunologically **active regions of antigen** that binds to antigen-specific membrane receptors on lymphocytes or to secreted antibodies. It is also called **antigenic determinants** or it is a molecular surface feature of an antigen that can be bound by an antibody.

❋ **Paratope:** is the molecular surface feature of an antibody that binds to an epitope.

❋ **Adjuvants**: are substances that are non-immunogenic alone but enhance the immunogenicity of any added immunogen.

❋ **Molecular patterns** **are low-molecular substances** evoking the reactions of innate immunity with no memory. There are pathogen-associated molecular patterns (PAMPs), allergen- associated molecular patterns (AAMPs), damage-associated molecular patterns (DAMPs), and tumor-associated molecular patterns (TAMPs).

✳ **Molecular Structure of Antigens**

▶ At the molecular level, an antigen is characterized by its ability to be "bound" at the antigen-binding site of an antibody. Antibodies tend to discriminate between the specific molecular structures presented on the surface of the antigen. **Antigens** are usually either proteins, peptides, or polysaccharides. This includes parts (coats, capsules, cell walls, flagella, fimbrae, and toxins) of bacteria, viruses, and other microorganisms. Lipids and nucleic acids are antigenic only when combined with proteins and polysaccharides. **For example**, the combination of lipids and polysaccharides are lipopolysaccharides (LPS).

▶ Cells present their immunogenic-antigens to the immune system via a major histocompatibility (MHC) molecule. Depending on the antigen presented and the type of the histocompatibility molecule, several types of immune cells can become activated due to an antigen.

▶ By using the "lock and key" metaphor, the antigen itself can be seen as a string of keys-any epitope being a "key" – each of which can match a different lock.

✷ <u>Types of antigen on the basis of source</u>

► Antigens are categorized into broad classes based on their origin. So many different molecules can function as an antigen in the body, and there is considerable diversity even within these categories. These are the main classes of antigens that are involved in immune system activation. Their diversity is analogous to the immense diversity of the diseases that the immune system works to overcome.

► **Based on the Source Antigens, they are classified into:**

   1.Exogenous (entering from outside)

1. Endogenous (generated within cells)

2. Auto antigen

3. Tumor antigen

4. Native antigen

1. **Exogenous Antigens**

▶ Exogenous antigens are antigens that have entered the body from the outside, for example by inhalation, ingestion, or injection. Exogenous antigens are the most common kinds of antigens, and includes pollen or foods that may cause allergies, as well as the molecular components of bacteria and other pathogens that could cause an infection.

1. **<u>Endogenous Antigens</u>**

▶ Endogenous antigens are that have been generated within previously-normal cells as a result of normal cell metabolism or because of viral or intracellular bacterial infection (which both change cells from the inside in order to reproduce). The fragments are then presented on the surface of the infected cells in the complex with MHC class I molecules.
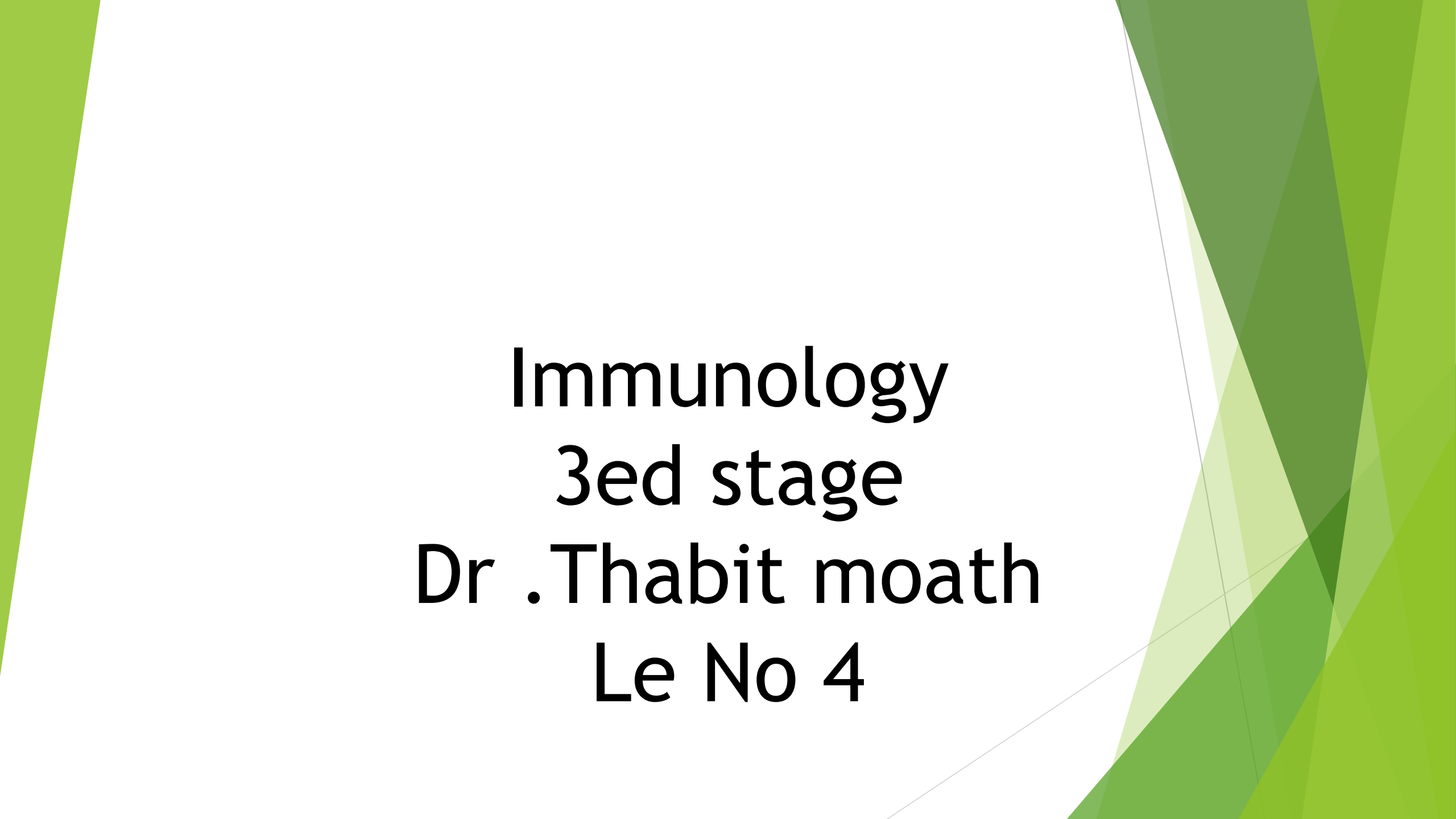
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▶ Auto-antigens are normal "self" protein or complex of proteins or nucleic acid that is attacked by the host's immune system, causing an autoimmune disease. These antigens should, under normal conditions, not be the target of the immune system, but due to mainly genetic and environmental factors, the normal immunological tolerance for such an antigen has been lost.

1. **<u>Tumor Antigens (Neoantigens)</u>**

▶ These antigens are presented by MHC I or MHC II molecules on the surface of tumor cells. These antigens result from a tumor-specific mutation during malignant transformation of normal cells into cancer cells. Despite expressing this antigen, many tumors have developed ways to evade antigen recognition and immune system killing.

1. **<u>Native Antigens</u>**

▶ A native antigen is an antigen that is not yet processed by an APC to smaller parts. T cells cannot bind native antigens, but require that they be digested and processed by APCs, whereas B cells can be activated by native ones without prior processing.

❋ Some antigens can be exogenous in origin but later can turn endogenous. E.g. Intracellular viruses.
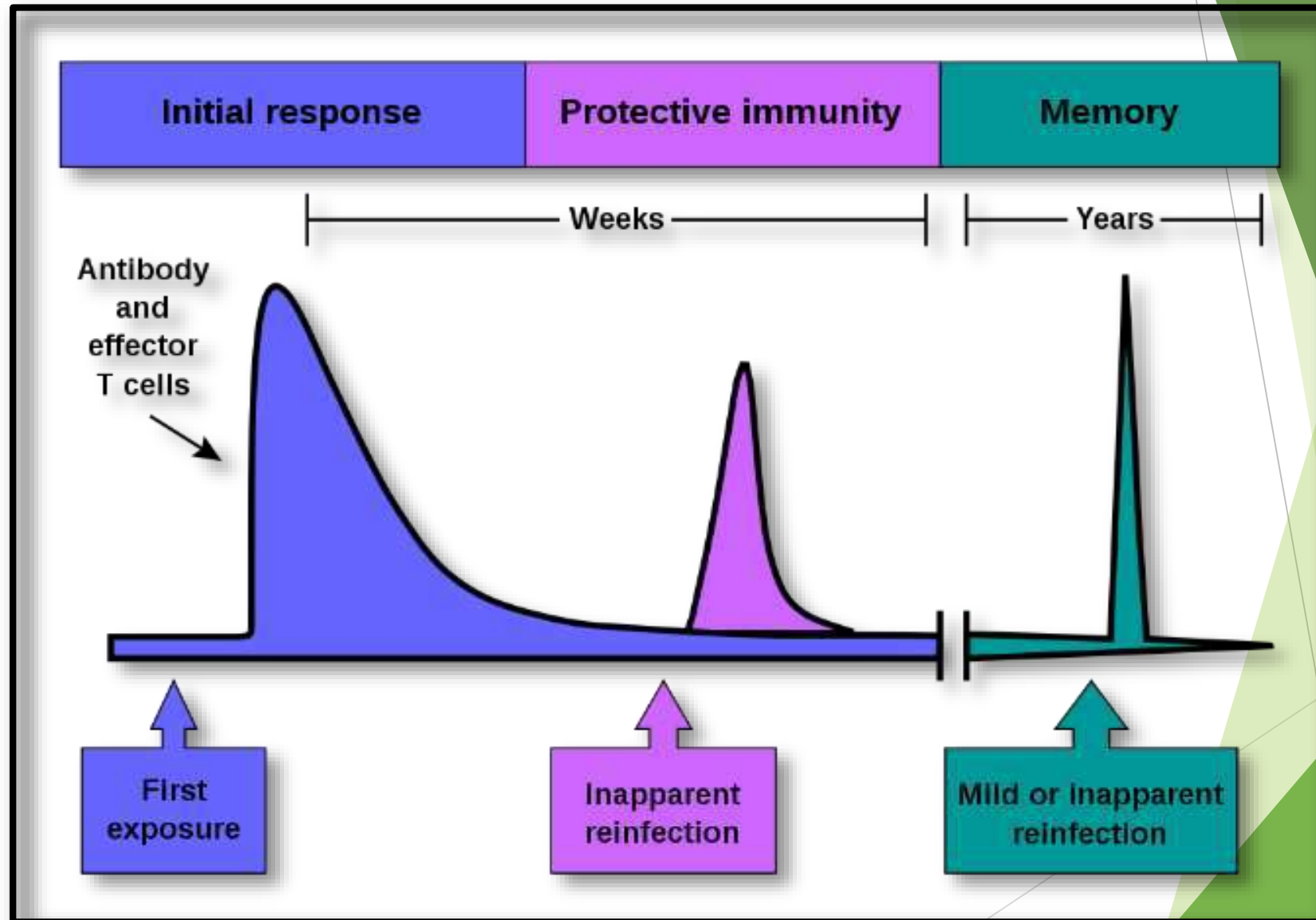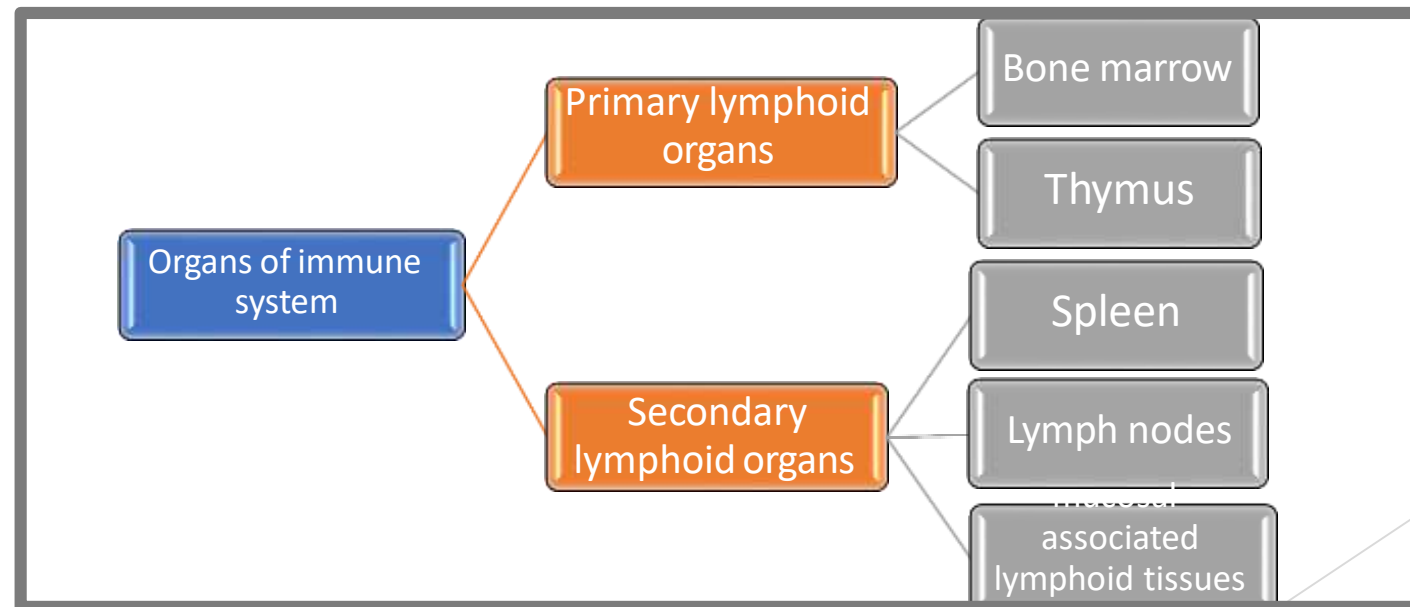
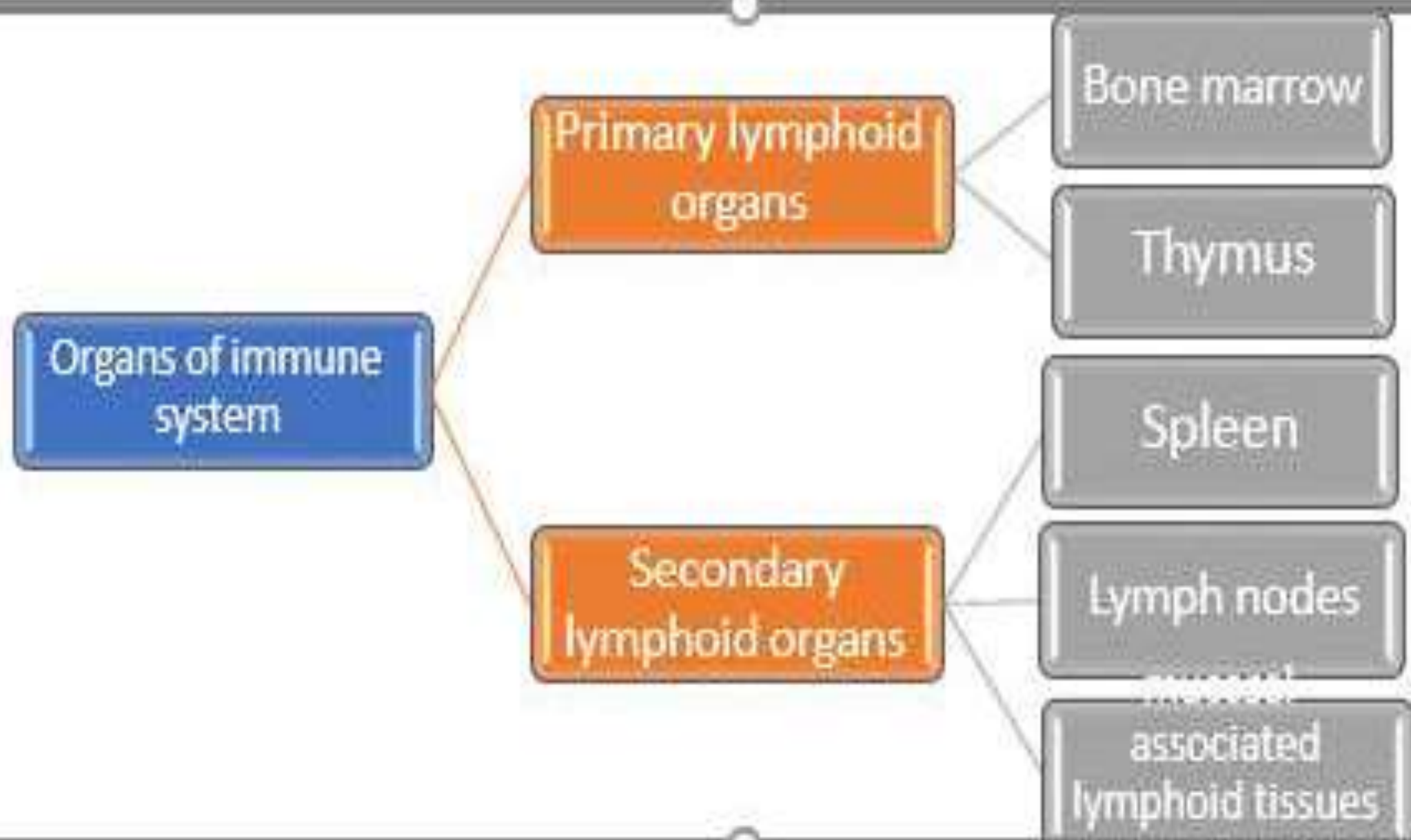**Immunologic Memory**

Immunological memory is responsible for the adaptive component of the immune system, special **T and B cells** so-called memory T and B cells. Immunological memory is the basis of vaccination.

Organs of immune system

Primary lymphoid organs
- Bone marrow
- Thymus

Secondary lymphoid organs
- Spleen
- Lymph nodes
- Mucosal associated lymphoid tissues

Lymphoid system:

Some stem cells are migrate to primary lymphoid organs which are includes

Thymus and Bone marrow to continue of proliferate and differentiate of

them.

•Thymus is a smallest gland situates in front of the heart and behind the

•sternum, it's receiving progenitor cells that leaved bone marrow for

•proliferation, differentiation, and form differentiated cell known as thymus-

• derived T cell.

•Bone marrow is the source of progenitor cells; it also plays a role in

• differentiate of progenitor cells into B-lymphocytes.

•Mature of T and B lymphocytes leave their differentiated site and migrate

• to peripheral or secondary lymphoid organs which are include:

1 3-Lymph nodes: its act like lymphoid filters in the lymphatic system.

2Spleen: its act like a lymphatic filter with in the blood vascular tree, it's an important **site**

3 of antibody production in response to **intravenous particulate antigen**

e.g. bacteria.
Mucosa-associated lymphoid tissue (MALT): is a collection cells in sub-

mucosal of different tissue. MALT is composed from

a) of gut-associated lymphoid tissues (GALT) which is lining the intestinal tract

b)   includes lymphoid tissue in the intestines (payer's patches) and the liver,

c)bronchus associated lymphoid tissue (BALT) lining the respiratory tract, and
d)lymphoid tissue lining the genitourinary tract.

5- Tonsils: are nodular aggregates of lymphoid tissues, their function is to detect
 and respond to pathogens in the respiratory secretion.
*Blood: is an important lymphoid organ and immunologic effector tissue.

The function of the secondary lymphoid organs is to

maximize encounters between lymphocytes and foreign

 substances.
Maturation of T and B lymphocyte:

The maturation of lymphocyte cells include recognized and

binds of each specialized receptors which are located on the

 surface of T and B lymphocytes.

●Pre- thymocytes make contact by receptors with specialized epithelial cell, dendritic cells, and macrophages in the thymus that provides an opportunity for the selection and differentiate of T cells. Cytokines such as interleukin 1, 2, 6, and 7 also thymic hormones are play role of maturity during the selections process.

●B cells contact with stromal cells in the bone marrow and cytokines is important for differentiation of B cells such as interleukin 1, 6, and 7 in this process
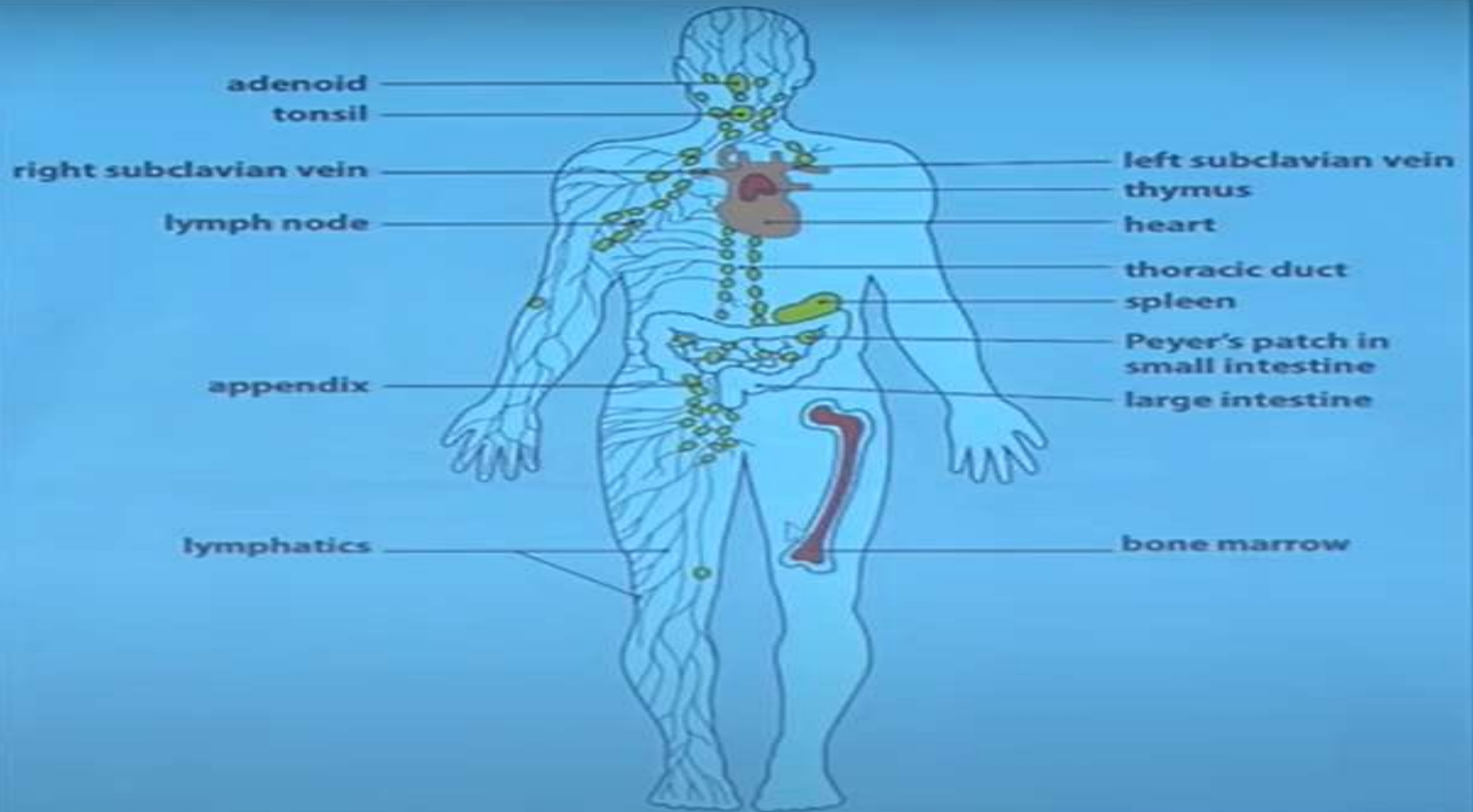
Figure 1.18 The Immune System, 3ed. (© Garland Science 2009)

The lymph node

lymphoid follicle (mostly B cells)

afferent lymphatic vessel

medullary sinus

artery

vein

T-cell area

efferent lymphatic vessel

germinal center

marginal sinus

lymphoid follicle

germinal center

T-cell area

medullary sinus
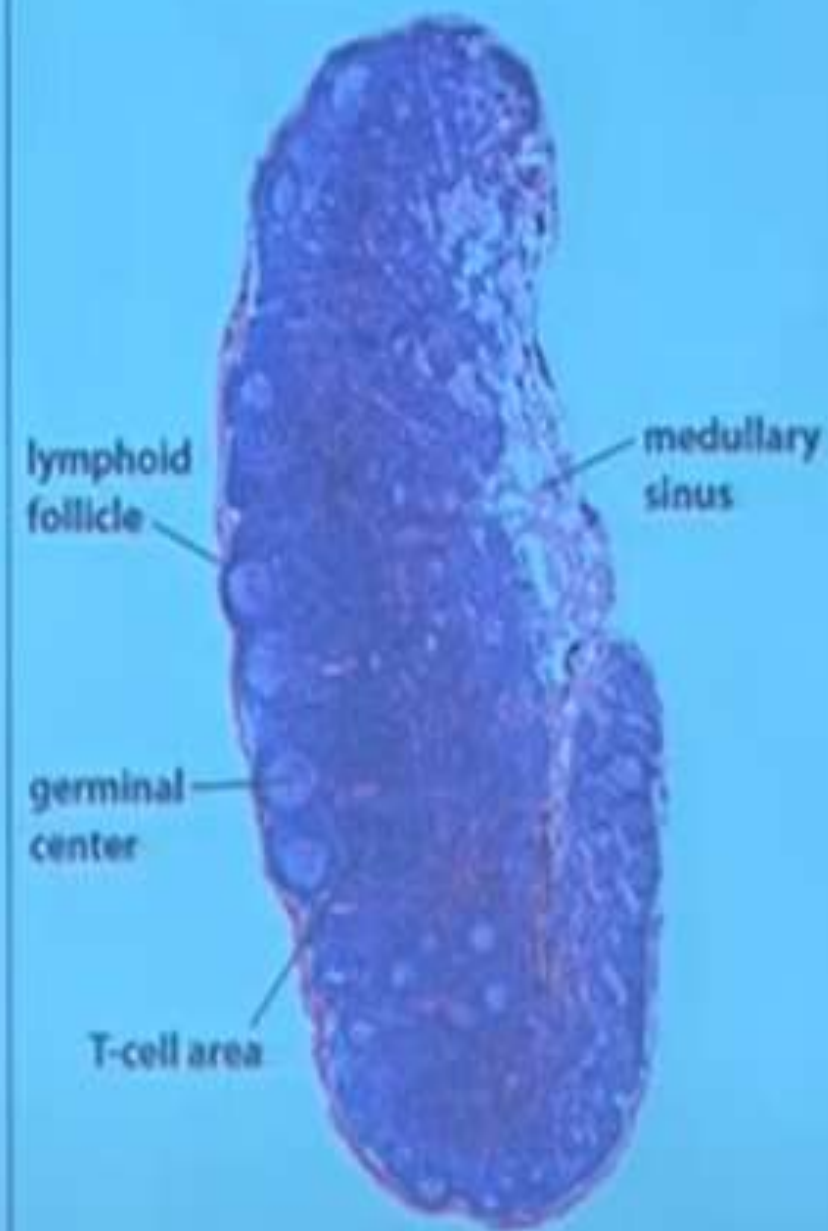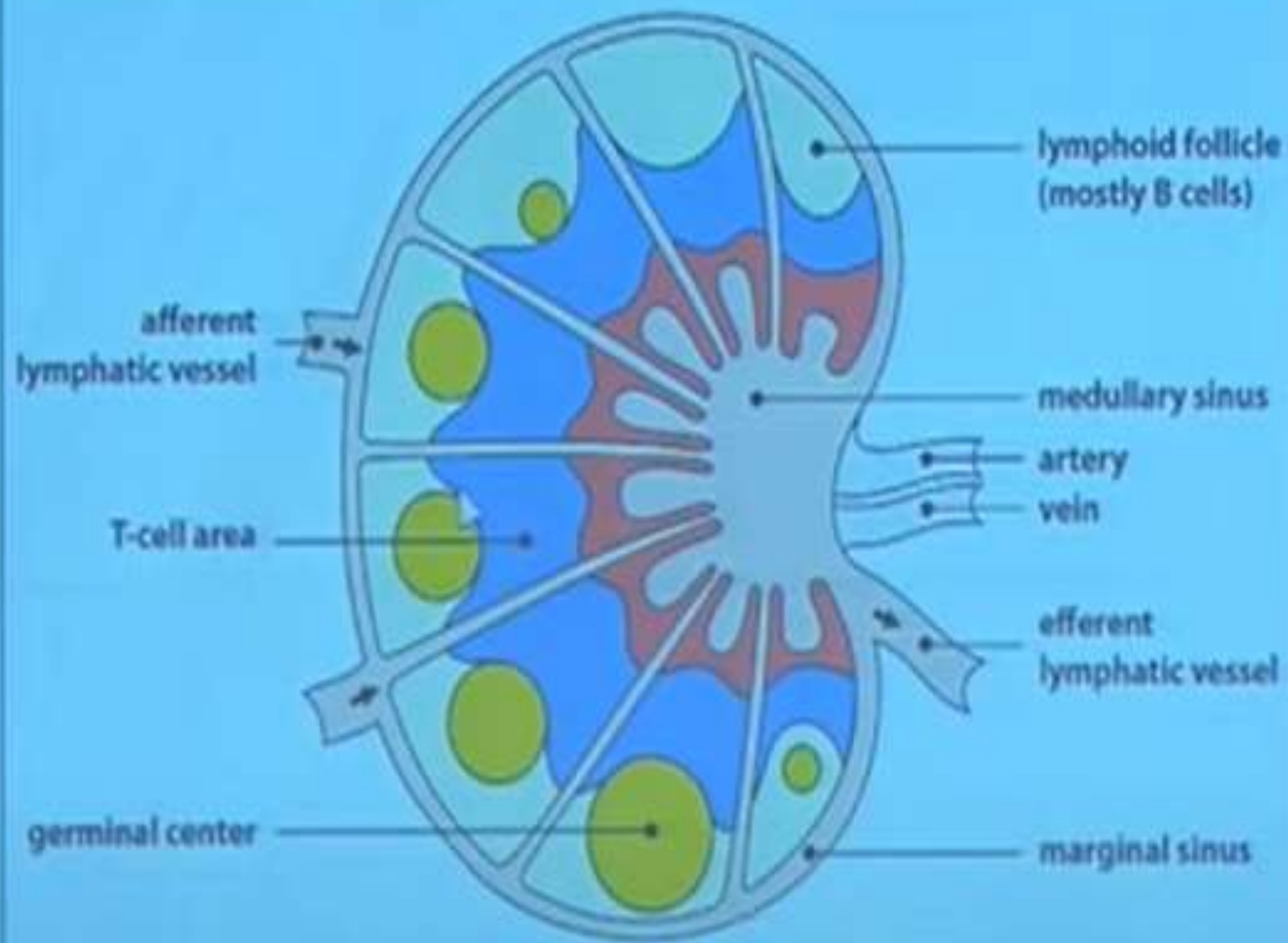
Gut-associated lymphoid tissue
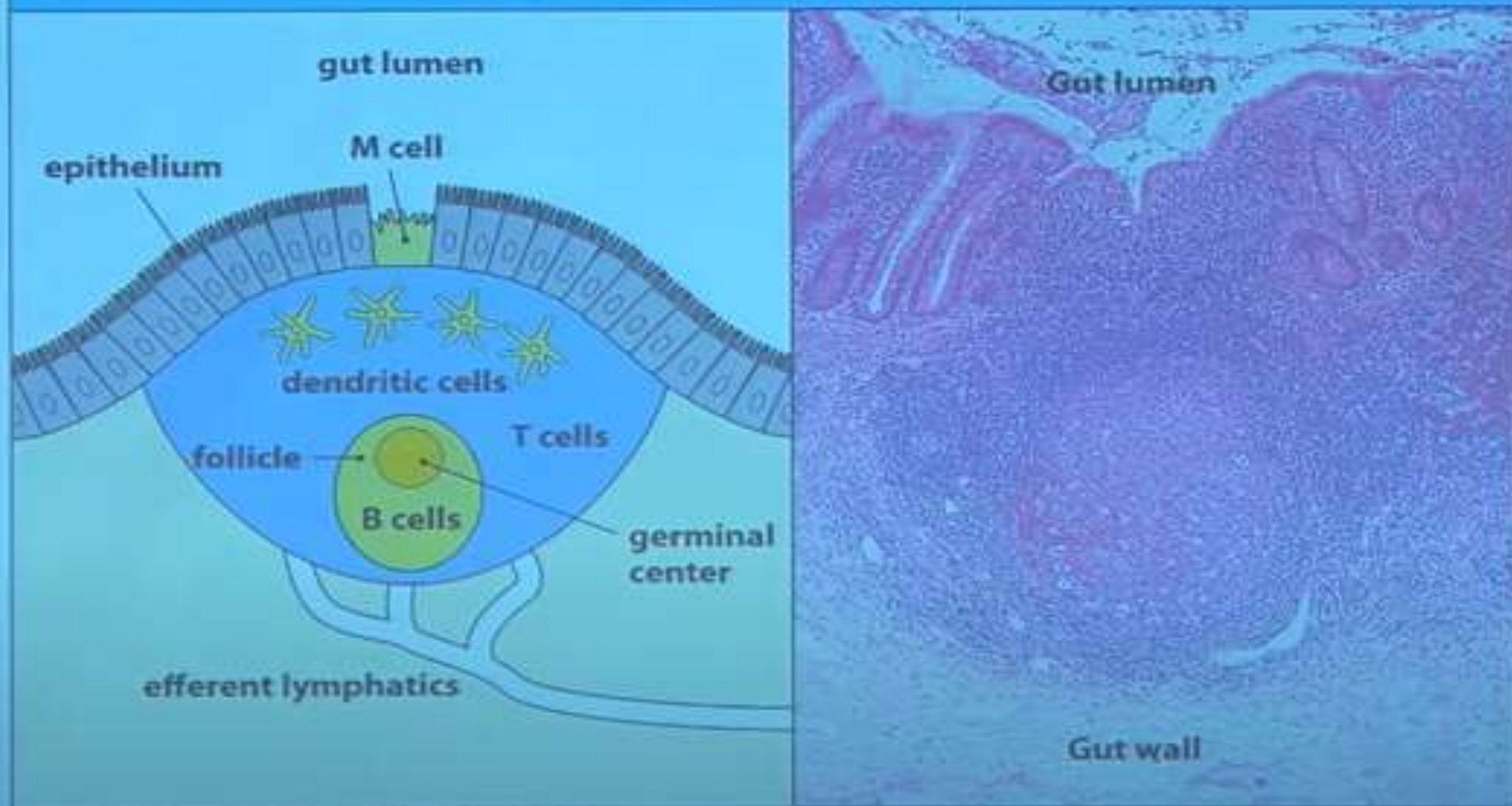
Figure 1.25 The Immune System, 3ed (© Garland Science 2009)

Immunology
3ed stage
Dr .Thabit moath
Le No 3

Phagocytosis

1. Activation of Phagocytic cells and Chemotaxis
2. Recognition of invading microbes
3. Ingestion and formation of phagosomes
4. Formation of phagolysome
5. Microbial killing and formation of residual bodies
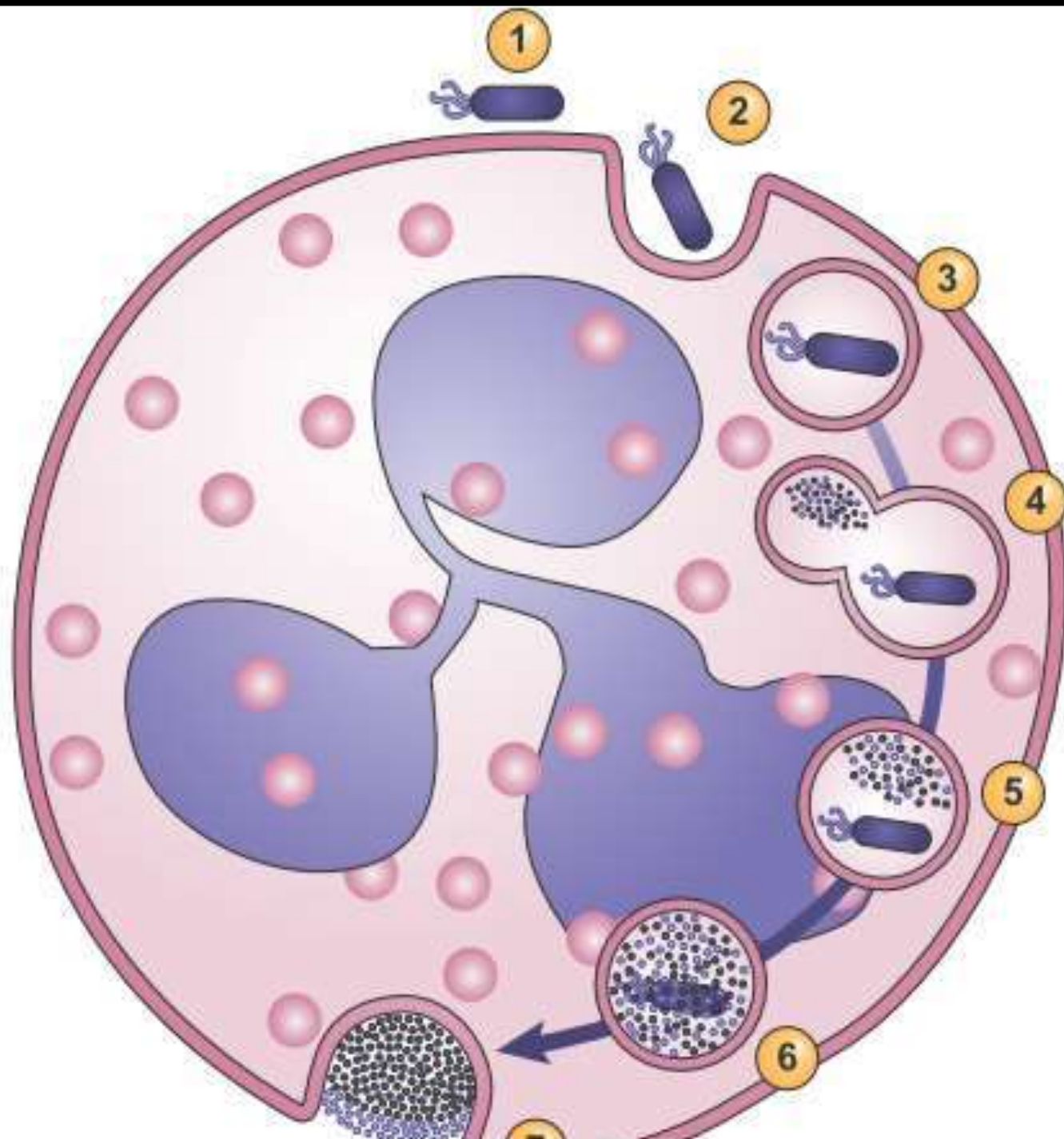
1. Adherence: physical contact between the phagocytic cell and the microorganism occurs, aided by opsonins.

2. Engulfment: outflowing of cytoplasm to surround the microorganism.

3. Formation of phagosome: microorganism is completely surrounded by a part of the cell membrane.

4. Granule contact: lysosomal granules contact and fuse with the phagosome.

5. Formation of the phagolysosome: contents of the lysosome are emptied into this membrane-bound space.

6. Digestion of the microorganism by hydrolytic enzymes.

7. Excretion: contents of phagolysosome are expelled to the outside by exocytosis.

Cells of the Adaptive Immune System:

The cells that differentiate along with **Lymphoid lineage cells** (lymphocytic pathways) are known as **lymphocytes** that are the key cell involved in the adaptive immune response. Lymphocytes may differentiate along one of several different pathways based on specific functions and the proteins on their cell surfaces into:

1. T cell

2. B cell

3. Natural killer (NK) cells.

In the peripheral blood of adults, approximately 10% to 20% of lymphocytes are B cells, 61% to 80% are T cells, and 10 %to 15% are NK cells.

1. B Cells (derived from bone marrow)

B cells, also known as B lymphocytes, are a type of white blood cell of the lymphocyte subtype. They function in the humoral immunity component of the adaptive immune system by Immunoglobulin production. Additionally, B cells present antigens (they are also classified as professional antigen-presenting cells (APCs)) and secrete cytokines. In mammals, B cells mature in the bone marrow, which is at the core of most bones.

B cells, unlike the other two classes of lymphocytes, T cells and natural killer cells, express B cell receptors (BCRs) on their cell membrane. BCRs allow the B cell to bind to a specific antigen, against which it will initiate an antibody response.

1. T Cells (derived from thymus)

T cells are so named because they differentiate in the thymus .Lymphocyte precursors called **Thymocytes** enter the thymus from the bone marrow through the bloodstream. As they mature, the T cells express unique surface markers that allow them to recognize foreign antigens bound to cell membrane proteins called **MHC molecules**.

Three main subtypes of T cells can be distinguished according to their unique functions: **Helper**, **Cytotoxic**, and **regulatory T cells**. The subtypes can be identified by the presence of the CD4+ and CD8+. T cells bearing the **CD4$^+$** receptor are mainly either helper or regulatory cells, whereas the **CD8$^+$** population consists of cytotoxic T cells.

1. **T helper cells (Th cells), CD4+ T cells**: are a type of T cell that play an important role in the immune system, particularly in the adaptive immune system. As their name suggests, they "help" the activity of other immune cells by releasing **cytokines**, small protein mediators that alter the behavior of target cells that express receptors for those cytokines. They are generally considered essential in B cell antibody class switching, breaking cross-tolerance in dendritic cells, in the activation and growth of cytotoxic T cells, and in maximizing bactericidal activity of phagocytes such as macrophages and neutrophils

1. **A cytotoxic T cell (also known as TC, cytotoxic T lymphocyte, CTL, T-killer cell, cytolytic T cell, CD8+ T-cell or killer T cell**) is a T lymphocyte that kills cancer cells, cells that are infected (particularly with viruses), or cells that are damaged in other ways. Most cytotoxic T cells express **T-cell receptors (TCRs)** that can recognize a specific antigen. An antigen is a molecule capable of stimulating an immune response and is often produced by **cancer cells** or **viruses**. Antigens inside a cell are bound to **class I MHC** molecules, and brought to the surface of the cell by the class I MHC molecule, where they can be recognized by the T cell. If the **TCR is specific for that antigen**, it binds to the complex of the class I MHC molecule and the antigen, and the T cell destroys the cell.

► **The role of T cells is:**

1. Produce cytokines that contribute to immunity by **stimulating B** cells to produce antibodies

2. Assisting in **killing tumor cells** or **infected target cells**

3. **Helping to regulate** both the <u>innate</u> and <u>adaptive</u> immune response.

► This process is known as **Cell Mediated Immunity**.

► **Cell-mediated immunity** is an immune response that **does not involve antibodies**. Rather, cell-mediated immunity is the activation of <u>phagocytes</u>, **antigen-specific cytotoxic T-lymphocytes**, and the release of various cytokines in response to an antigen.

❋ CD= cluster of differentiation

1.Natural Killer (NK) Cells

Natural killer cells, also known as NK cells or **large granular lymphocytes (LGL)**, are a type of **cytotoxic lymphocyte** critical to the <u>innate</u> immune system. The role of NK cells is analogous to that of cytotoxic T cells in the **adaptive** immune response. NK cells provide rapid responses to **virus-infected cells**, acting at around 3 days after infection, and **respond to tumor formation**.

1. NK cells look like large granular lymphocytes and are found in blood, liver and secondary lymphoid organs particularly the spleen and mucosal associated lymphoid tissue (MALT).
2. can kill target cells, even in the absence of antibody or antigenic stimulation.
3. they do not need prior activation
4. but have the relevant recognition molecules on their surfaces already. Non-specific agents, such as mitogens
5. IFN-γ and IL-12, can activate them further.
6. NK cells IS part of the early host response to viral infection
   NK cells express two types of surface receptor.

1. Expression of MHC class I proteins by most normal cells prevents NK cells from killing healthy cells. Interference
2. NK-mediated killing either directly (secretion of granzymes or perforin), by FcRIII and ADCC or by secretion of IFN-γ and TNF-α.
3. they show little specificity and they have no memory.
4. The range of their potential targets is broad.
5. Animals and rare patients with deficient NK cell function have an increased incidence of certain tumours and viral infections.
6. The human immunodeficiency X-linked lymphoproliferative syndrome is an example in which EBV driven tumours are associated with absent NKT cells

Antigen-presenting cells (APCs) are a heterogeneous group of immune cells that mediate the cellular immune response by processing and presenting antigens for recognition by certain lymphocytes such as T cells. Classical APCs include dendritic cells, macrophages, and B cells.

Enumerate phagocytosis process
Activation of Phagocytic cells and Chemotaxis
Recognition of invading microbes
Ingestion and formation of phagosomes
Formation of phagolysome
Microbial killing and formation of residual bodies
Enumerate mast cell enzymes &
 bio chemical medieator

acid phosphatase, alkaline phosphatase, and protease, as well as histamine

- Mast cells are located in the thymus

- Mast cells are larger than basophils with a small round nucleus and more granules. like basophils, they have a long-life span of between 9-18 months.

- Dendritic cells are so named because they are rounded shape

- Dendritic cells capturing an antigen in the tissue by attachment.

- Dendritic cells, however, are considered the **less effective APC** in the body, as well as the most potent phagocytic cell.

- Toll-like receptors (TLRs) are expressed on the:
1. Membranes of macrophages, neutrophils, dendritic cells, lymphocytes, epitheliocytes, platelets, splenocytes, etc.

2. Present in the endosomes of the cells.

- Toll-like receptors (TLRs) are expressed on the Bells
- Toll-like receptors (TLRs) activated T cells
- Which are **Cells of the Adaptive Immune System**
1. T cell
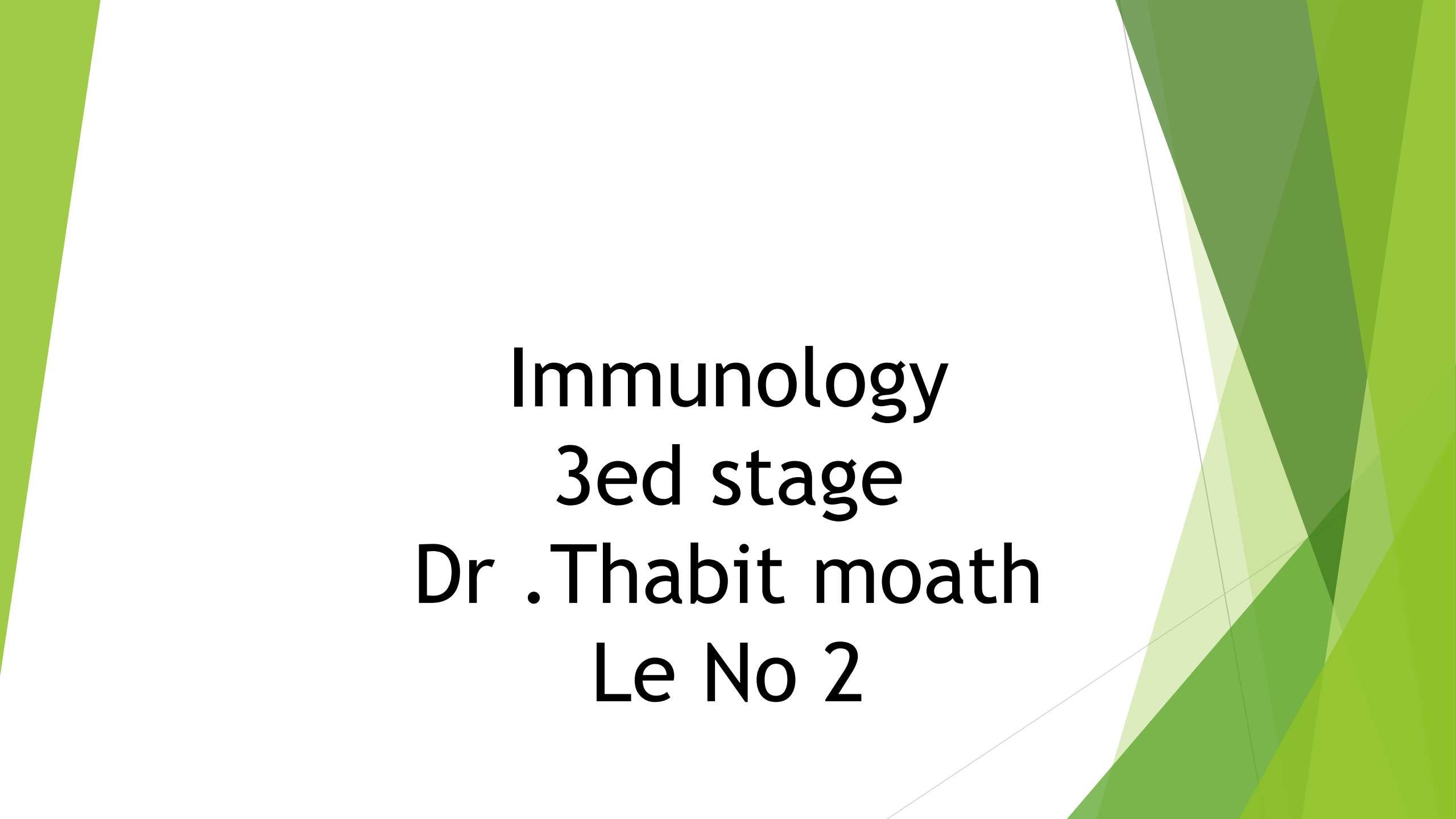
2. B cell

3. Natural killer (NK) cells.

- **B cells** are belong the **humoral** immunity component of the **adaptive** immune system by **Immunoglobulin** production

- T cells are belong the humoral immunity component of the adaptive immune system by Immunoglobulin production

- B cells, unlike the other two classes of lymphocytes, T cells and natural killer cells, express B cell receptors (BCRs) on their cell membrane.

- T cells, unlike the other two classes of lymphocytes, B cells and natural killer cells, express B cell receptors (BCRs) on their cell membrane

- Enumerate T cells types
- Three main subtypes of T cells can be distinguished according to their unique functions: **Helper**, **Cytotoxic**, and **regulatory T cells**.
- Sub types of T cells are
- The subtypes can be identified by the presence of the CD4+ and CD8+.
- The subtypes of B cells identified by the presence of the CD4+ and CD8+.
- **A cytotoxic T cell (also known as TC, cytotoxic T lymphocyte, CTL, T-killer cell, cytolytic T cell, CD8+ T-cell or killer T cell)** is a T lymphocyte that kills cancer cells, cells that are infected (particularly with viruses), or cells that are damaged in other ways.

Difference between Innate and Adaptive Immunity

S.N.	Characteristics	Innate Immunity	Adaptive immunity
1.	Presence	Not require exposure	require exposure
2.	Specificity	Non-Specific	Specific
3.	Response	Fights any foreign agent	Fight only specific agent
4.	Response	Rapid	Slow (1-2 weeks)
5.	Potency	Limited and Lower potency	High potency
6.	Time span	The immunity remains throughout the life.	The immunity remains throughout the life. Or short
7.	Inheritance	yes	none
8.	Presence	Present at birth	Develops during a person's lifetime and can be short-lived.
9.	Allergic Reaction	None	Immediate and Delay hypersensitivity
10.	Used Against	For microbes	Microbes and non-microbial substances called antigens
11.	Memory	No memory	Long term memory
12.	Speed	Faster response	Slower response
13.	Complement system activation	Alternative and lectin pathways	Classical pathway
14.	Anatomic and physiological barriers	Skin, Mucous membranes, Temp, pH, chemicals, etc.	Lymph nodes, spleen, mucosal associated lymphoid tissue.
15.	Example	White blood cells fighting bacteria, causing redness and swelling, when you have a cut.	Chickenpox vaccination so that we don't get chickenpox because adaptive immunity system has remembered the foreign body.

Immune cell and Tissue

Innate

cells
Macrophage
Granulocytes
Basophile
Mast cell
Natural killer cell
Dendritic cell

Soluble components
Complement system
Cytokines

Pattern Recognition Receptors (PRR)

Adaptive

cells:
T Lymphocyte
B Lymphocyte

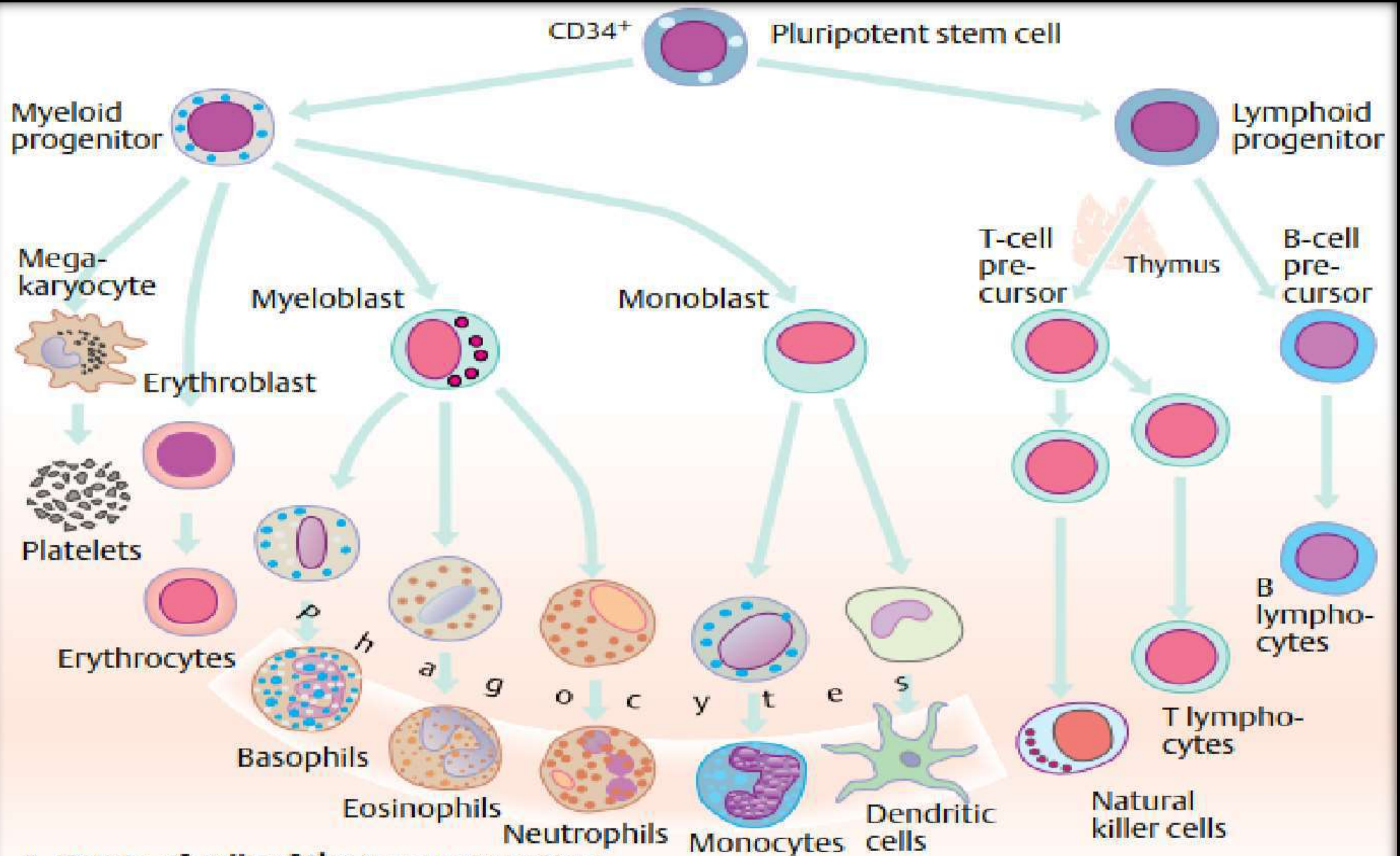**Soluble components**
Antibodies
Cytokines
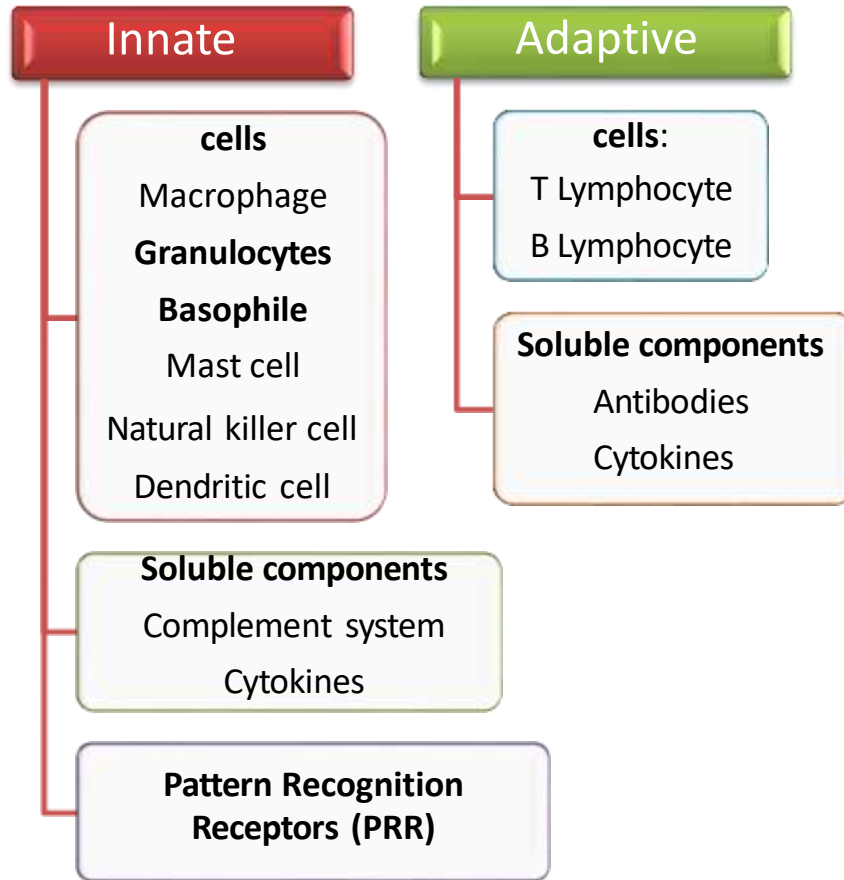
Introduction:

White blood cells (WBCs), or **Leukocytes**, in the peripheral blood play a key role in both innate and adaptive immunity. There are five principal types of leukocytes in peripheral blood: **Neutrophils, Eosinophils, Basophils, Monocytes**, and **Lymphocytes**. The first four types are all parts of innate immunity, while lymphocytes are considered part of adaptive immunity

All blood cells arise from a type of cell called a **hematopoietic stem cell**.

❊ **Hematopoietic lineages**:
To form WBCs, the pluripotent hematopoietic stem cell gives rise to two distinct types of precursor cells: **1. myeloid precursors (MP)** and **2. lymphoid**

CD34+ Pluripotent stem cell

Myeloid progenitor

Lymphoid progenitor

Mega-karyocyte

Myeloblast

Monoblast

T-cell pre-cursor

Thymus

B-cell pre-cursor

Erythroblast

Platelets

Erythrocytes

p h a g o c y t e s

Basophils

Eosinophils

Neutrophils

Monocytes

Dendritic cells

Natural killer cells

B lympho-cytes

T lympho-cytes

A. Origin of cells of the immune system
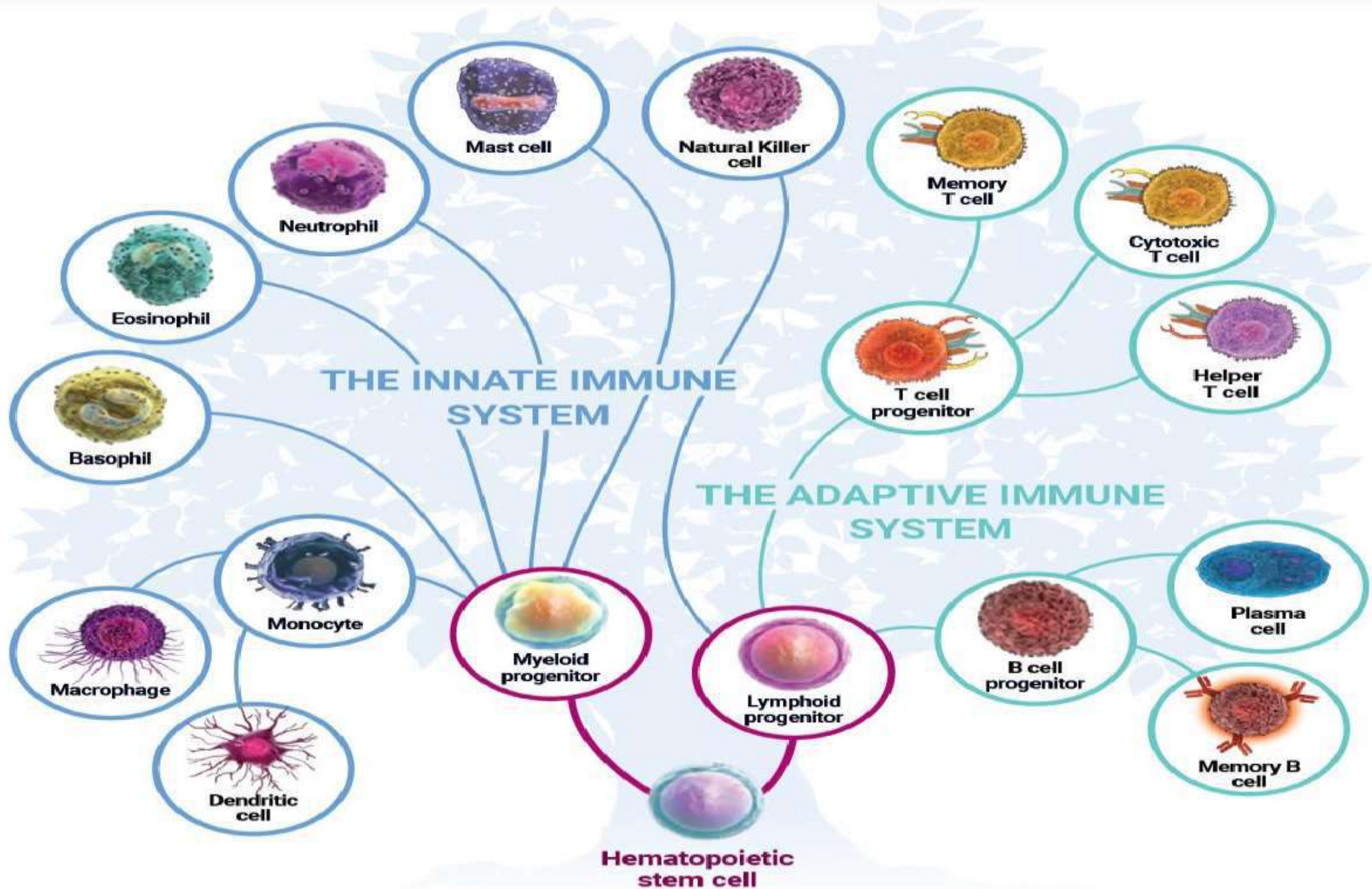
precursors (LP).

1. **Myeloid Precursors** give rise to the WBCs that participate in phagocytosis, which are known as the myeloid line. Phagocytic cells are key to innate immunity, but they are also important in processing antigens for the adaptive response.

2. Lymphocytes arise from Lymphoid Precursors and form the basis of the adaptive immune response.

Leukocytes may be broadly classified by the absence (A granular) or presence (granular) of cytoplasmic inclusions or granules.

A. **A Granular Leukocytes**
    1. B Lymphocyte (B cell)
    2. T Lymphocyte (T cell)
    3. Plasma cell
    4. Dendritic cell
    5. Monocyte

B. **Granular Leukocyte**
    1. Neutrophil
    2. Eosinophil
    3. Basophil
    4. Natural killer cell

A. Cells of the Innate Immune System

❈ **Leukocytes in Peripheral Blood**

1) Neutrophils

Neutrophilic granulocytes are the most numerous leukocyte in adults. They are compromised approximately **50%-70%** of the peripheral blood leukocytes. They are also called PolyMorphoNuclear (PMN) cells because of their variable number of nuclear segments (two to five) which distinguished them from other WBC of lymphoid or myeloid origin, such as **lymphocytes and monocytes.**

- They are short-lived (half-life of approximately 7h)

- and highly mobile, as they can enter parts of tissue where other cells/molecules cannot.

- Neutrophils are a type of **phagocyte** and they are very effective at **killing** bacteria so, the **main function of neutrophils is phagocytosis, resulting in the destruction of foreign particles**.

- During the beginning **(acute) phase of inflammation**, particularly as a result of bacterial infection, environmental exposure,and some **cancers,**

- neutrophils are one of the first responders of inflammatory cells to migrate toward the site of inflammation. They migrate through the blood vessels and then through interstitial tissue, following chemical signals by **interleukins** in a process called **chemotaxis**.

The increasing of neutrophils in peripheral blood is often an indication for acute infection.

Segmented neutrophil

Eosinophils:

Eosinophil

is one of the immune system components responsible for combating multicellular **parasites and certain infections** in vertebrates.

Eosinophil are approximately 12-15 μm in diameter and normally make up between 2% of the circulating WBCs in a **non-allergic person**.

Their number increases in an **allergic reaction** or in response to certain **parasitic infections**.

The nucleus of eosinophil is usually **bi-lobed** granulocytes with cytoplasmic granules are spherical and evenly distributed throughout the cell, contain a large number of previously synthesized proteins that contain basic proteins.

Eosinophil take up the **acid eosin dye and the cytoplasm** is filled with large orange to reddish- orange granules.

Eosinophil is capable **of phagocytosis** but are much less efficient than neutrophils because they are present in smaller numbers and they lack digestive enzymes.

They are able to neutralize basophil and mast cell products.

In addition, they can use cationic proteins to damage cell membranes and kill larger parasites that can not be phagocytized
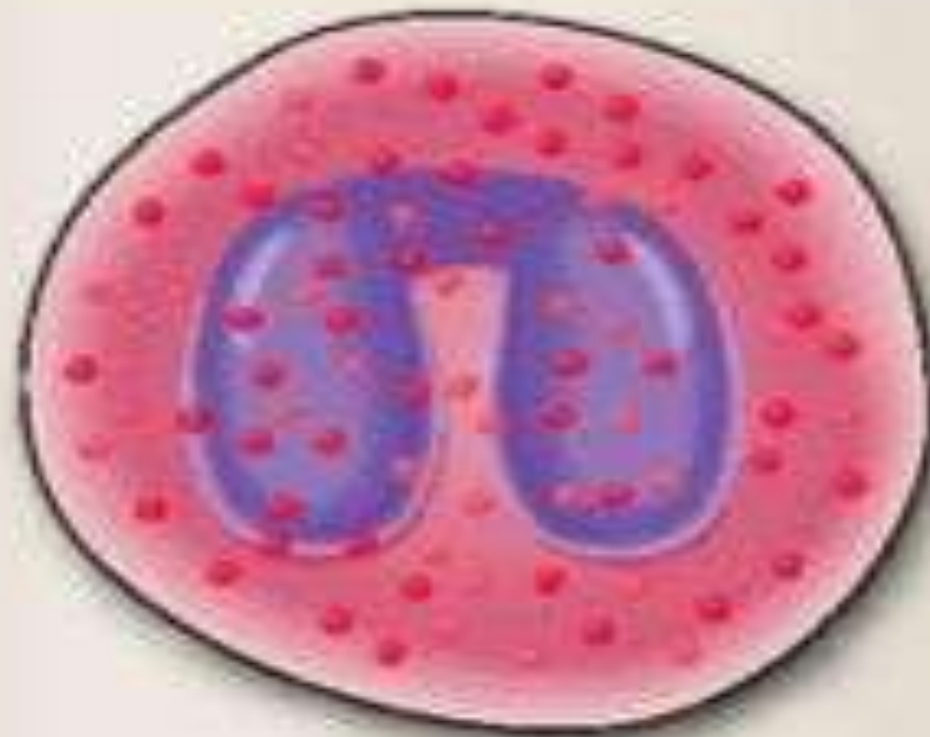
Eosinophil

3) Basophils

Basophils are the least numerous of WBCs found in peripheral blood, representing less than 1% of all circulating WBCs.

The smallest of the granulocytes, basophils are slightly larger than RBCs (between 10-15 μm in diameter)

and contain rough, densely staining deep-bluish-purple granules that often obscure the nucleus. Constituents of these granules include **histamine**, **cytokines**, **growth factor**, and a small amount of **heparin**, all of which have an important function in inducing and maintaining **allergic reactions**. <u>Histamine</u> contracts smooth muscle and heparin is an anticoagulant. In addition, basophils **regulate** some **T helper (Th) cell responses and stimulate B cells to produce the antibody IgE.**

Basophils have a **short life** span of only a few hours in the bloodstream.

Basophil
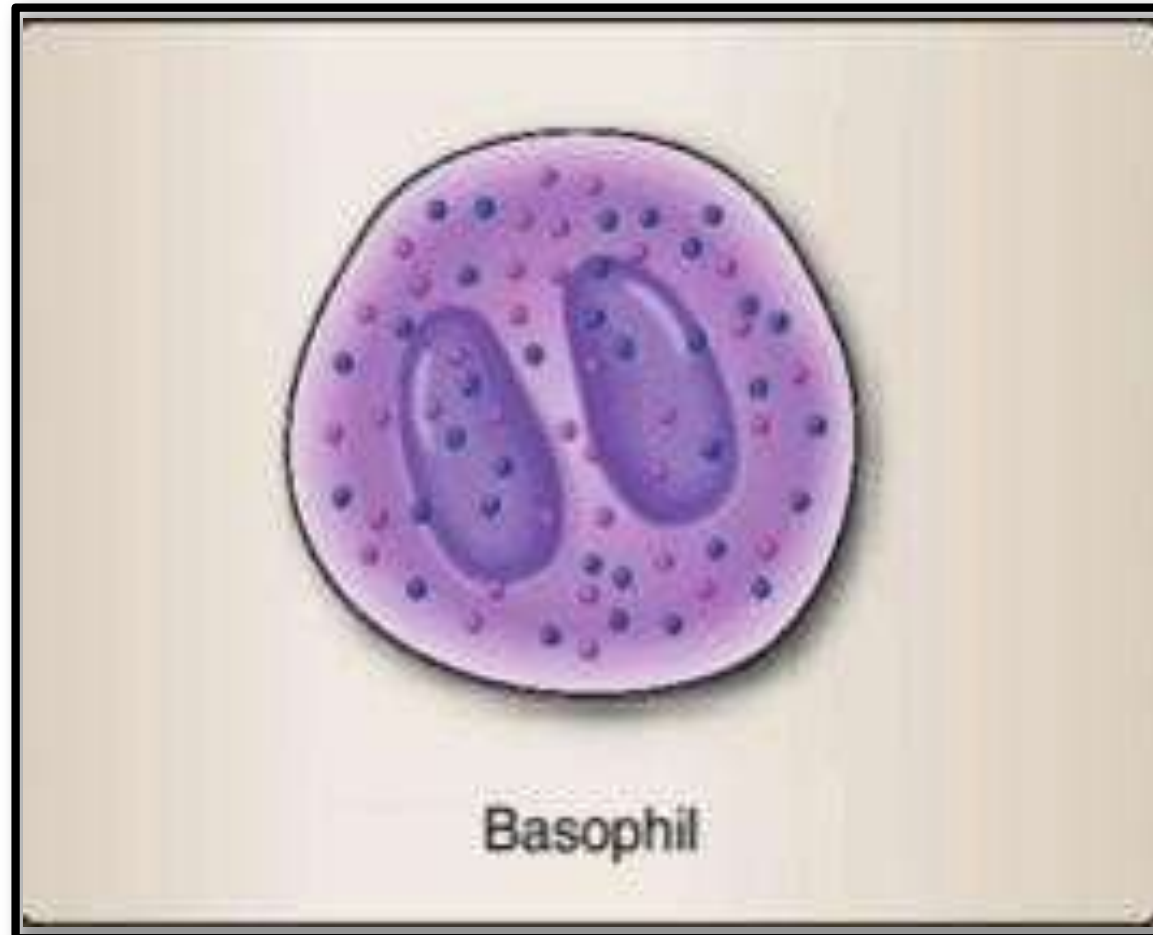
4) Monocytes

Monocytes are the largest type of leukocyte. They are looks amoeboid in appearance,

and have non-granulated cytoplasm. Containing unilobar nuclei, these cells are one of the types of mononuclear leukocytes ground-glass appearance because of the presence of fine dust-like granules.

These cells are the scavengers of the body.

They phagocytize, or pick up cellular debris, foreign cells, and particles and degrade them enzymatically.

Monocytes make up between 4%-10% of total circulating WBCs; however

, they do not remain in the circulation for long.They stay in peripheral blood for up to 30 hours, they then migrate to the tissues and become known as **macrophages**.

Monocyte serve multiple roles in immune function. Such roles include:

1. Replenishing resident macrophages under normal conditions
2. Migration within approximately 8–12 hours in response to inflammation signals from sites of infection in the tissues.
3. Monocytes also can influence the process of adaptive immunity.
4. Differentiation into macrophages or dendritic cells to effect an immune response
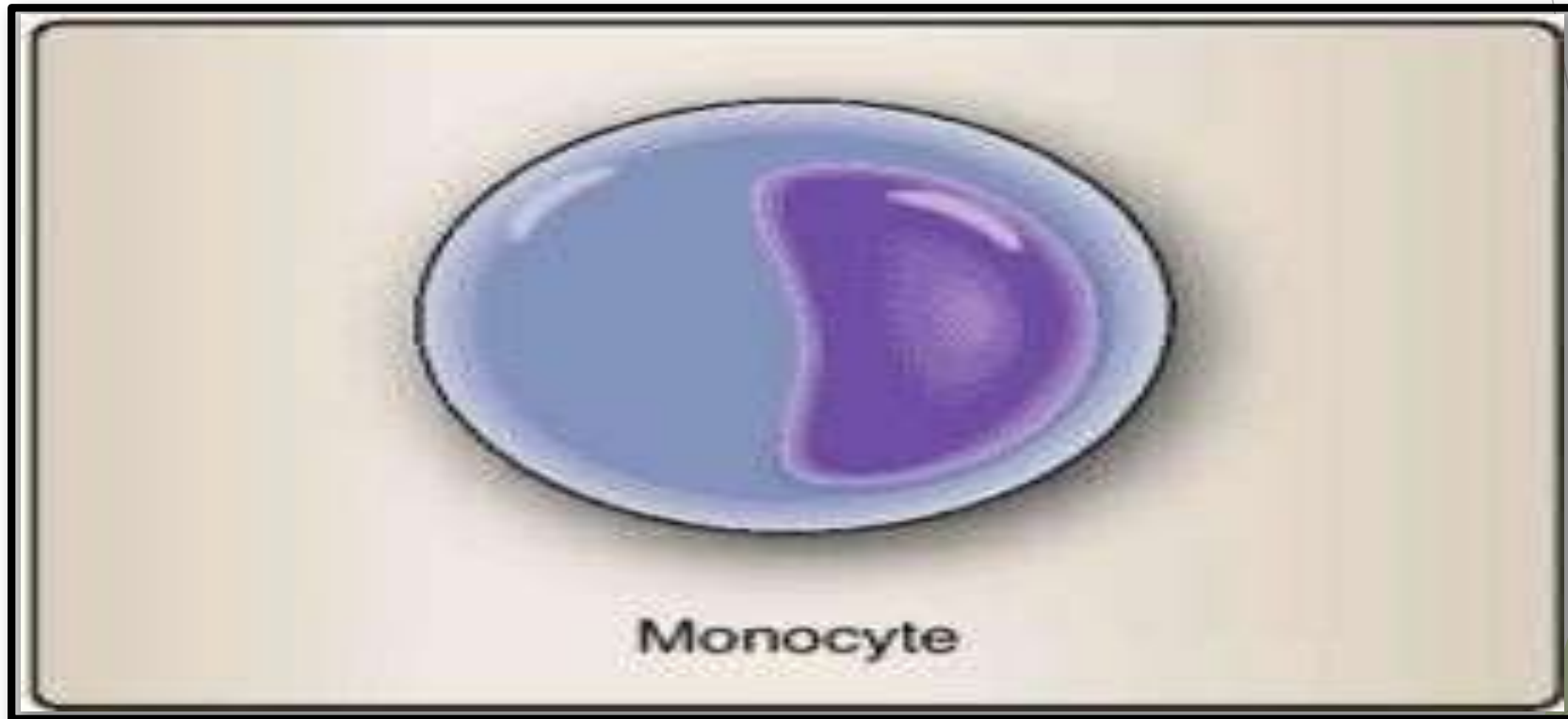
Monocyte

Tissue Cells

1) Macrophage

All macrophages arise from monocytes, which can be thought of as macrophage precursors because additional differentiation and cell division **takes place in the tissues**. The transition from monocyte to macrophage in the tissues is characterized by progressive cellular enlargement to between 25-80 μm. **Unlike monocytes, macrophages don't have peroxidase.**

Macrophages have specific names according to their particular tissue location:

✳ In the lung are alveolar macrophages
✳ In the liver, Kupffer cells
✳ In the brain, Microglial cells
✳ In the bone, Osteoclasts
✳ In connective tissue, Histiocytes.

❖Macrophages **may not be as efficient as neutrophils in phagocytosis** because **their motility is slow compared with that of the neutrophils**. Some macrophages progress through the tissues by means of amoeboid action, whereas others are immobile.

However, their life span appears to be in the range of months rather than days.

❖Function of Macrophage in an Innate immune system:

1.Microbial killing

2.Anti-tumor activity

3.Intracellular parasite eradication

4.Phagocytosis

5.Secretion of cell mediators.

❖<u>**Macrophages play a major role in the adaptive immune response by presenting antigens to T and B cells.**</u>

Mast Cells

Tissue mast cells resemble basophils, but they come from a different lineage.

Mast cells are distributed throughout the body in a wide variety of tissues such as skin, connective tissue, and the mucosal epithelial tissue of the respiratory, genitourinary, and digestive tracts.

Mast cells are larger than basophils with a small round nucleus and more granules. **Unlike basophils, they have a long life span of between 9-18 months**.

The enzyme content of the granules in mast cells helps to distinguish them from basophils because they contain acid phosphatase, alkaline phosphatase, and protease, as well as histamine.

Mast cells play a role in allergic reactions,

but they can also function as antigen-presenting cells (APCs).

**They can both enhance and suppress the adaptive immune response**.

3)Dendritic Cells

Dendritic cells are so named because they are covered with long membranous extensions that make them resemble nerve cell dendrites.

Progenitors in the bone marrow give rise to dendritic cell precursors that travel to lymphoid as well as non-lymphoid tissue.

They are classified according to their tissue location in a similar manner to macrophages. After **capturing an antigen in the tissue by phagocytosis or endocytosis, dendritic cells present the antigen to T lymphocytes** to initiate the adaptive immune response in a similar way as macrophages. Dendritic cells, however, are considered the **most effective APC** in the body, as **well as the most potent phagocytic cell.**

Activation of adaptive immune response in a draining lymph node
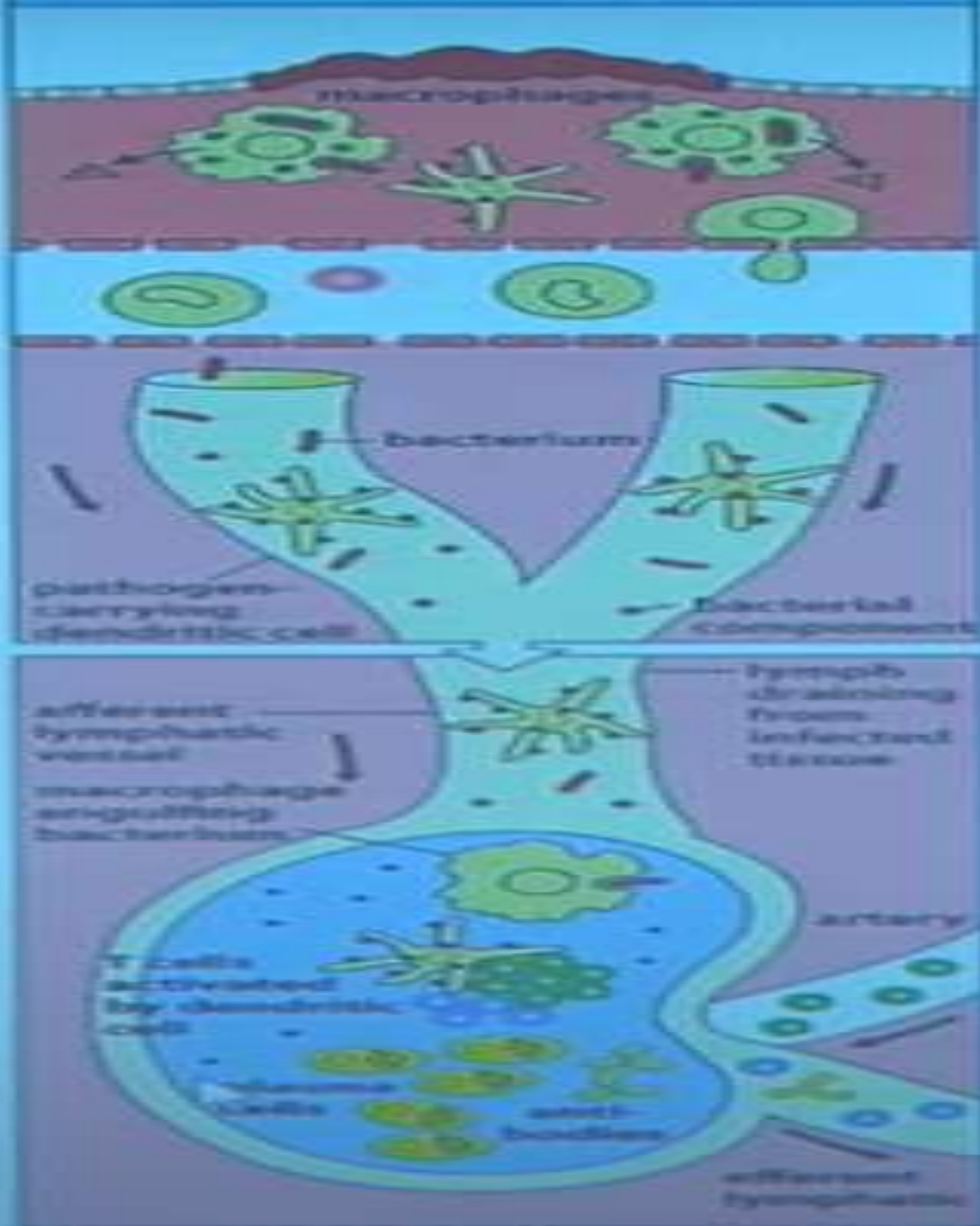
Pattern Recognition Receptors (PRRs)

PRRs are molecules expressed by cells of the innate immunity, which are capable of sensing "patterns," triggering the reactions of innate immunity such as inflammation and taking part in adaptive immune responses.

Toll-like receptors (TLRs) and NOD-like receptors (NLRs) are the most important type of PRR.

Toll-like receptors (TLRs) are expressed on the:

1. **Membranes** of macrophages, neutrophils, dendritic cells, lymphocytes, epitheliocytes, platelets, splenocytes, etc.
. **Present in the endosomes of the cells.**

The term "toll" comes from drosophila, in which TLRs were first identified. TLRs can recognize both PAMPs and DAMPs and provide a l**ink between innate and adaptive immunity** as they can interact with antigens too. A pathogen bound to TLRs may be engulfed, **digested,** and presented to lymphocytes in antigenic form.

The term "toll" comes from drosophila, in which TLRs were first identified. TLRs can recognize both PAMPs and DAMPs and provide a l**ink between innate and adaptive immunity** as they can interact with antigens too. A pathogen bound to TLRs may be **engulfed, digested,** and **presented** to lymphocytes in antigenic form.

pathogen –associated molecular patterns

Damage-associated molecule patterns

| TLR	Ligands (PAMP, DAMP)
TLR1	Triacyl lipopeptides of *Mycobacterium tuberculosis*, DAMP
TLR2	Diacyl and triacyl lipopeptides of Gram-positive bacteria, lipoteichoic acid, peptidoglycan, yeast zymozan, DAMP
TLR3	Viral dsRNA
TLR4	Lipopolysaccharide (endotoxin, LPS) of Gram-negative bacteria, DAMP
TLR5	Flagellin of bacterial flagella
TLR6	Diacyl lipopeptides of Gram-positive bacteria
TLR7	Viral ssRNA
TLR8	Viral ssRNA
TLR9	Unmethylated CpG nucleotides of bacterial and viral DNA
TLR10	Unknown

Immunology
3ed stage
Dr .Thabit moath
Le No 1

Introduction to immunity

A wide different of **organisms** and **their associated molecules** form a constant risk to the human body. The action human immune system is the defensive mechanisms that **identify** and **distinguish "no self" organisms and molecules from "self**," that which belongs within the body. And **remove** this risk later. Forging body(organisms) may enter the body from the **outside** (infectious organisms or toxic agents) or may **arise** from harmful changes occurring within the body (the malignant **transformation** of a previously normal cell into a cancer cell). Fortunately, the immune system consists of three layers of defense.

1.The first line of defense is provided by a set of **mechanical**, **chemical**, and **biologic** barriers that protect the body. If these barriers are breached,

2.the second and third lines of protective systems are activated: first the **innate immune** **system** and

3.then the **adaptive immune system**.
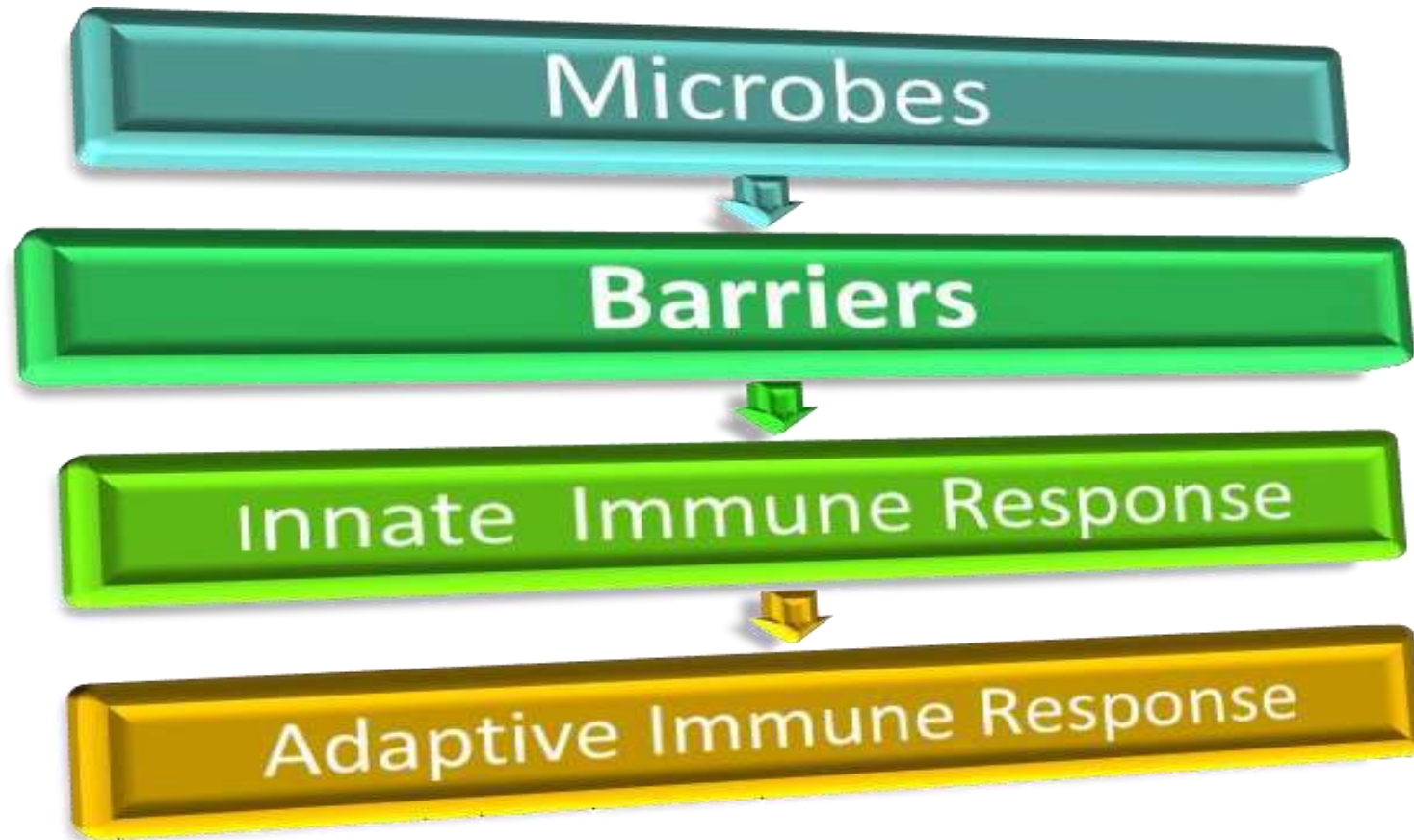
Classification of Immunity

Barriers to Infection (non-specific innate immunity)

The body has several **mechanical (physical)**, **chemical**, and **biologic** barriers that provide the first line of defense against the entry of microbes into the aseptic, environment of our tissues.

1. Physical Barriers

The initial mechanical barriers that protect the body against invasive microbes include:

A. Skin
   where consists of several **thick layers** prevent the entry of microbes into the body

B. Mucous membranes
  The epithelium of mucous membranes lines all of the body's cavities that come into contact with the environment, such as the respiratory, gastrointestinal, and urogenital

- In the respiratory tract, the mucus traps inhaled bacteria, fungi, and other particles.
- In the gastrointestinal tract, the mucus and mucous membranes **help to protect the epithelial cells and underlying tissues from damage by digestive enzymes and to propel ingested matter** through the tract. Mucosal surfaces of the moist epithelium facilitate the exchange of molecules with the environment while also resisting microbial invasion.

C. Respiratory tract

The **hair-like cilia** of the epithelia lining the respiratory tract passages help the tract clean by moving the secretions containing trapped microbes and particles outward for **expulsion by coughing and sneezing**. The rhythmically beating cilia of the respiratory epithelium is commonly **disrupted** by chronic **smoking and chronic alcohol consumption**, leading to an **increased risk** of respiratory infections.

D. Urinary tract

Similar to the outward movement of secretions of the respiratory tract, **urination helps to inhibit movement of microbes from the environment up into the bladder and kidneys**.

The periodic voiding of sterile urine provides an externally **directed fluid pressure** that inhibits the inward movement of microbes along the urinary tract.

2. Chemical and Environmental Barriers

A. pH

Most pathogens are very sensitive to an acidic environment, **where an acid pH (less than 6) inhibits the growth of potential pathogens.**

1. **Skin**: The skin contains oil and sweat glands, some of whose products are slightly acidic.
   In general, the skin has a **pH of about 5.5.**

2. **Stomach**: Compared to the colon, the stomach has very few bacteria because of the highly
   acidic environment (normal pH of 1- 3). The acidic environment of the stomach prevents the colonization of the intestine ingested microbes.

3. **Vagina**: The acidic environment of the vagina and cervical in healthy women is normally
   pH 4.4 to 4.6. This acidic environment is the result of lactic acid production by the commensal bacteria *Lactobacilli* spp.

3. Microcidal action of secreted molecules

Several tissues that are in contact with the environment synthesize and secrete a variety of **microcidal molecules** that act **to inhibit or kill** microbes that are attempting to colonize.

1.Skin: The skin is protected in part by several **antimicrobial peptides** secreted by a variety of cell types found within the skin. Among these are **α-defensins**, **β-defensins**, and **cathelicidin.** All are able to inhibit microbial growth by direct action upon the microbes, perhaps by damaging the microbial membranes and causing lysis.

Other molecules with enzymatic activity are present in the skin as well. Sweat contains **lysozyme,** an enzyme that breaks down peptidoglycan (a constituent of most bacterial cell walls).

2.**Respiratory tract**: To protect the mucosal surfaces of the lungs, some cells of the respiratory epithelium secrete microcidal molecules such as **β-defensins**. These and other molecules in the respiratory tract can attach to microbes and make them more susceptible to ingestion and destruction **by phagocytic cells.**

3.Gastrointestinal tract: The gastrointestinal tract defends against pathogens in many ways.

In addition to the low pH of the stomach, some epithelial cells secrete microcidal molecules such as **α-defensins** and **cryptidin** that help to destroy many potential pathogens.

4. Biologic Barriers: Commensal Microbes

Commensal microbes are those that exist in a symbiotic relationship with the body. The skin

and the gastrointestinal tract are colonized by over 500 commensal bacterial and other

microbial species that are estimated to make up over 95% of the cells present in a normal

human body.

Commensal microbes colonizing the skin and gastrointestinal tracts "defend" their territory and inhibit the establishment of other potentially pathogenic microbes.

Skin	Gastrointestinal tract	Respiratory tract	Urogenital tract	Eyes
Epithelial cells joined by tight junctions				
Flow of fluid, perspiration, sloughing off of skin	Flow of fluid, mucus, food, and saliva	Flow of fluid and mucus, e.g., by cilia Air flow	Flow of fluid, urine, mucus, sperm	Flow of fluid, tears
Sebum (fatty acids, lactic acid, lysozyme)	Acidity, enzymes (proteases)	Lysozyme in nasal secretions	Acidity in vaginal secretions Spermine and zinc in semen	Lysozyme in tears
Antimicrobial peptides (defensins)				
Normal flora of the skin	Normal flora of the gastrointestinal tract	Normal flora of the respiratory tract	Normal flora of the urogenital tract	Normal flora of the eyes

Innate (natural) immunity:

Innate immune response is the individual's ability to resist infection by means of normally present body functions. These are considered **nonspecific** and are the same for all pathogens or foreign substances to which one is exposed. No prior exposure is required and the response **lacks memory** and specificity. Many of these mechanisms are subject to influence by such factors as **nutrition, age**, **fatigue**, **stress**, and **genetic determinants.**

Adaptive immunity:

Its unique cells and molecules, is the third level of defense against these potential threats to the body, following the barriers and the innate immune system. It is a type of resistance that is characterized by **specificity** for each individual pathogen, or microbial agent, and the ability to **remember a prior exposure**. **Memory** and specificity result in an increased response to that pathogen upon repeated exposure, something that does not occur in innate immunity.

- Adaptive immunity can be divided further into two types which are:
1. **Natural immunity**: consists of **passive** (maternal) and **Active**(infection) immunity

mechanical (physical)

Skin(thick layers)

Mucous membranes(epithelium)

Respiratory tract(mucus traps)

Urinary tract(urination)

Chemical

(oil and sweat glands)& PH& **Microcidal action of secreted molecules**
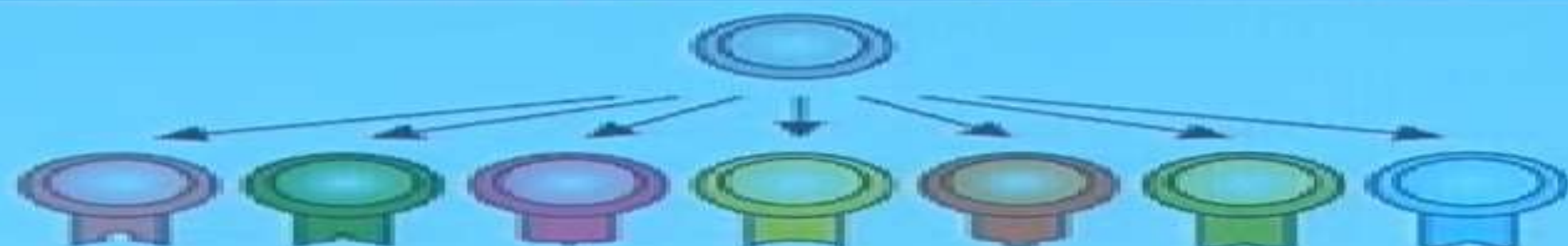
skin

Mucous membranes

Respiratory tract

Urinary tract

biologic barriers

commensal bacterial

During development, progenitor cells give rise to large numbers of lymphocytes, each with a different specificity

During infection lymphocytes with receptors that recognize the pathogen are activated

pathogen

Proliferation and differentiation of pathogen-activated lymphocytes give effector cells that terminate the infection

Effector cells eliminate pathogen

Figure 1.10 The Immune System, 3ed. (© Garland Science 2009)