Immunity

It is the ability of host body to prevent or overcome invasion by virulent microorganism. It is known as resistance or immunity.

Types of immunity:

A. Natural immunity

B. Acquired immunity/Adaptive Immunity

A. Natural immunity

It is also called non-specific immunity because it exists in all humans and is present from earliest time of life. It provides defense against infections by a number of chemical and mechanical barriers. These natural barriers include skin, mucous membrane, secretions, and component of blood, enzymes and often body fluids. These barriers form First line of defense against infections and diseases.

Susceptibility:

Lack of such natural resistance is called susceptibility.

Components of innate immunity :

1. Skin layer:

It provides a protective covering to all body tissues.

2. Mucous membrane:

It secretes sticky mucous, that trapped the air born particles and sweep them out by ciliary movement.

3. Acidity of stomach and vagina:

Acidic PH is toxic to most micro organisms.

4. Bile:

Bile is inhibitory to most micro organisms.

5. Lysozymes:

It is present in saliva and digest cell wall of gram positive bacteria.

6. Microbial flora:

The normal microbiota prevent pathogens from colonizing the host by competing with them for nutrients (competitive exclusion)

Second line of defense:

Once microorganism succeed in passing first line of defense, they enter the deeper tissues and are attached by specific cells of the body, which may ingest or destroy them. These cells are called **phagocytes** and they form second line defense.

Types:

There are two types

a. Free phagocytes.

b. Fixed phagocytes.

a)Free phagocytes:

It includes neutrophils that are present in blood stream.

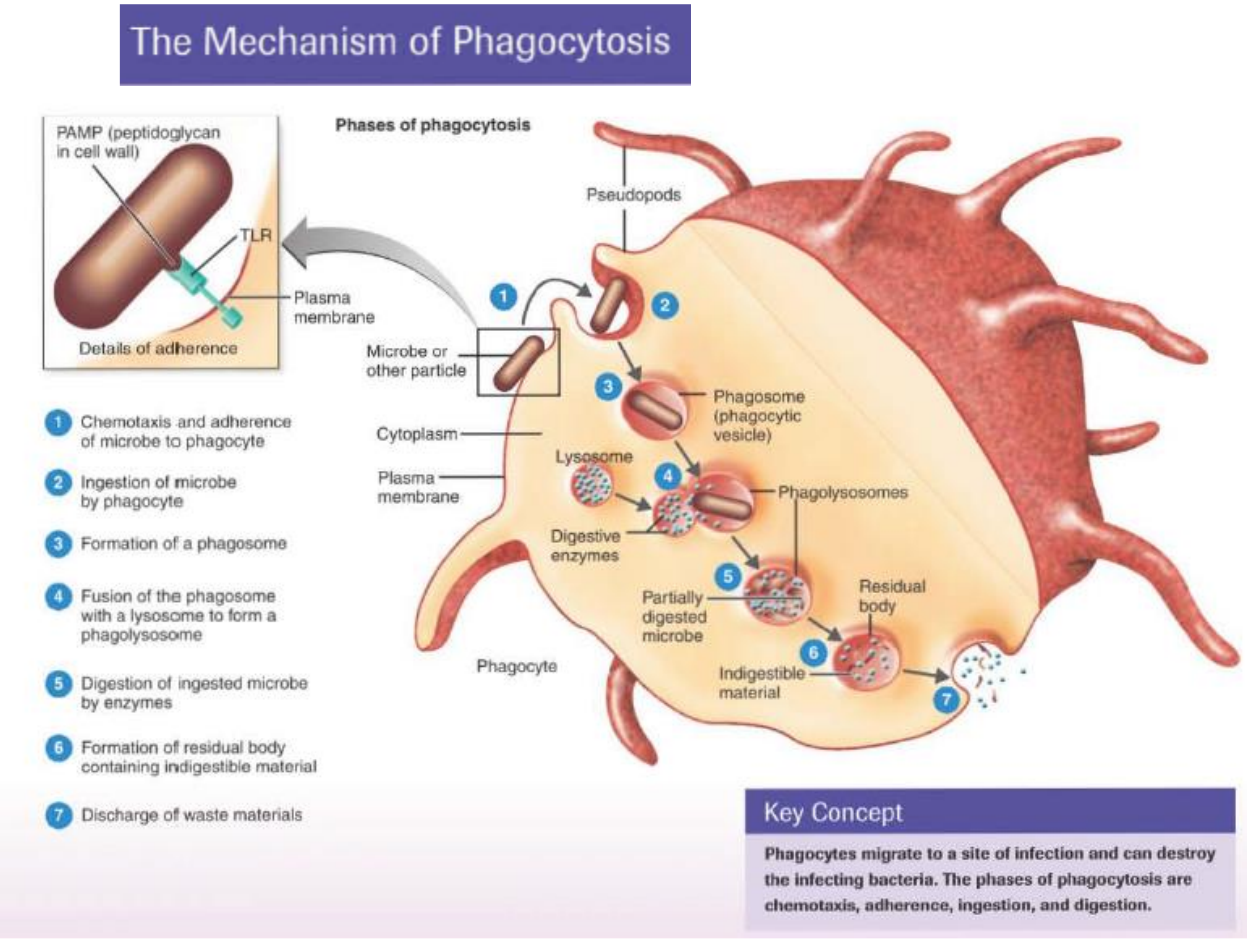
b)Fixed phagocytes:

These are the macrophages of reticular endothelial system. They are present in tissues.

Phagocytosis:

Greek words meaning eat and cell is the ingestion of a microorganism or other substances (such as debris) by a cell. It is the process by which phagocytes engulf the foreign particles in the form of **phagosomes.** It involves following steps.

1. Adequacy of blood flow.
2. Leukocytes adhesion to capillary walls and passing through it.
3. Chemo taxis is the chemical attraction between parasites and phagocytes mediated by a substance released by parasite.
4. Amoeboid engulfment of parasite in the form of phagosome**.**
5. Fusion of phagosomes with lysozymes and final digestion.
6. Removal of undigested particles outside the phagocytes.



II. Opsonization(Enhanced phagocytosis):

Sometimes, the pathogens repels the phagocytes (-ve chemotaxis) in such situation the component of complement system **opsonin** bind the parasite to phagocyte it. This enhanced phagocytosis called **opsonization**.

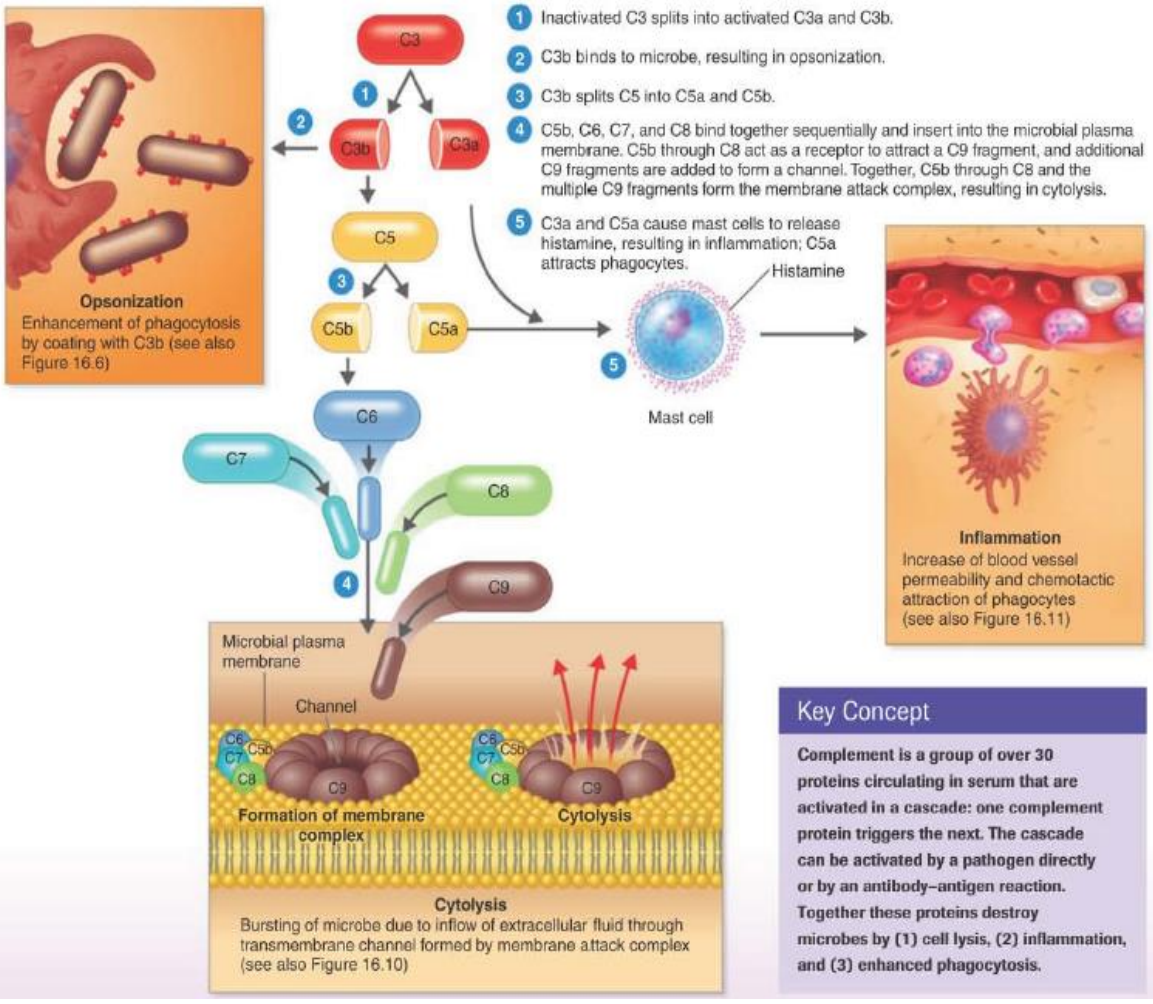
Opsonin:

They are serum protein that increases the susceptibility of parasite to phagocytosis.

III. Complement system:

It is a group of complex system of 30 serum proteins and other factors produced in liver and are present in normal serum of all vertebrates .They play a major role in animal defense.

Outcomes of Complement Activation



Key Concept

Complement is a group of over 30 proteins circulating in serum that are activated in a cascade: one complement protein triggers the next. The cascade can be activated by a pathogen directly or by an antibody–antigen reaction. Together these proteins destroy microbes by (1) cell lysis, (2) inflammation, and (3) enhanced phagocytosis.

Inflammation

Damage to the body's tissues triggers a local defensive response called inflammation, another component of the second line of defense. The damage can be caused by microbial infection (lipopolysaccharide of microorganism may induce inflammatory response), physical agents (such as heat, radiant energy, electricity, or sharp objects), or chemical agents (acids, bases, and gases).

Inflammation has the following functions:
(1) To destroy the injurious agent, if possible, and to remove it and its by-products from the Body. Influx of phagocytes from capillaries to tissues is facilitated by increased permeability of capillaries.

(2) If destruction is not possible, to limit the effects on the body by confining or walling off the injurious agent and its by-products

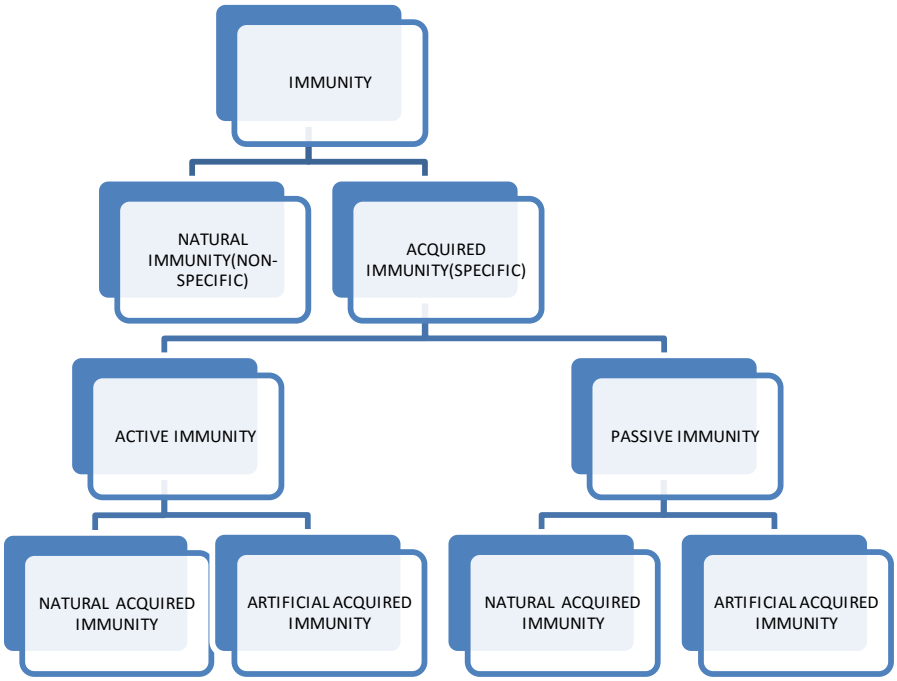
(3) To repair or replace tissue damaged by the injurious agent or its by-products

Fever

An abnormally high body temperature, a third component of the second line of defense .The most frequent cause of fever is infection from bacteria (and their toxins) or viruses.
Up to a certain point, fever is considered a defense against disease. Interleukin-I helps step up the production o f T cells. High body temperature intensifies the effect of antiviral interferon and increases production of transferrins that decrease the iron available to microbes. Also, because the high temperature speeds up the body's reactions, it may help body tissues repair themselves more quickly. However death results if temperature rises above 44-46C.

Interferones

Interferons (IFNs) are a class of similar antiviral proteins produced by certain animal cells, such as lymphocytes and macrophages, after viral stimulation. One of the principal functions of interferons is to interfere with viral multiplication.



Acquired immunity(specific defense):

It involves the formation of antibodies(**third line of defense**) as a result of stimulation by foreign particles (**Antigen**).acquired immunity is developed during individuals life time.

Adaptive immunity displays four characteristic attributes:

■ Antigenic specificity

The antigenic specificity of the immune system permits it to distinguish subtle differences among antigens. Antibodies can distinguish between two protein molecules that differ in only a single amino acid.

■ Diversity

The immune system is capable of generating tremendous diversity in its recognition molecules, allowing it to recognize billions of unique structures on foreign antigens

■ Immunologic memory

Once the immune system has recognized and responded to an antigen, it exhibits immunologic memory; that is, a second encounter with the same antigen induces a heightened state of immune reactivity.

■ Self/nonself recognition

Finally, the immune system normally responds only to foreign antigens, indicating that it is capable of self/nonself recognition

It is of two types.

A. Active immunity:

It is the immunity that develops from exposure to antigen that lead to production of antibodies. Exposure to antigen may be intentional or unintentional.

Types of active immunity

It is of two types

a)Natural acquired Active immunity:

It results due to unintentional exposure of antigen **e.g.** Immunity that develops after an infection such as mumps and measles etc.

b)Artificial acquired Active immunity:

It develops as a result of intentional exposure to antigen **e.g.** vaccine administration.

B. Passive immunity:

It is the immunity that develops from infusion of antibodies from outside source.

Types of passive immunity:

It is of two types.

a)Artificial acquired Passive immunity:

It is due to intentional injection of antibodies rich serum into circulation, **e.g**Hepatitis A ,antiserum, chicken pox antiserum .

b)Natural acquired Passive immunity(congenital immunity):

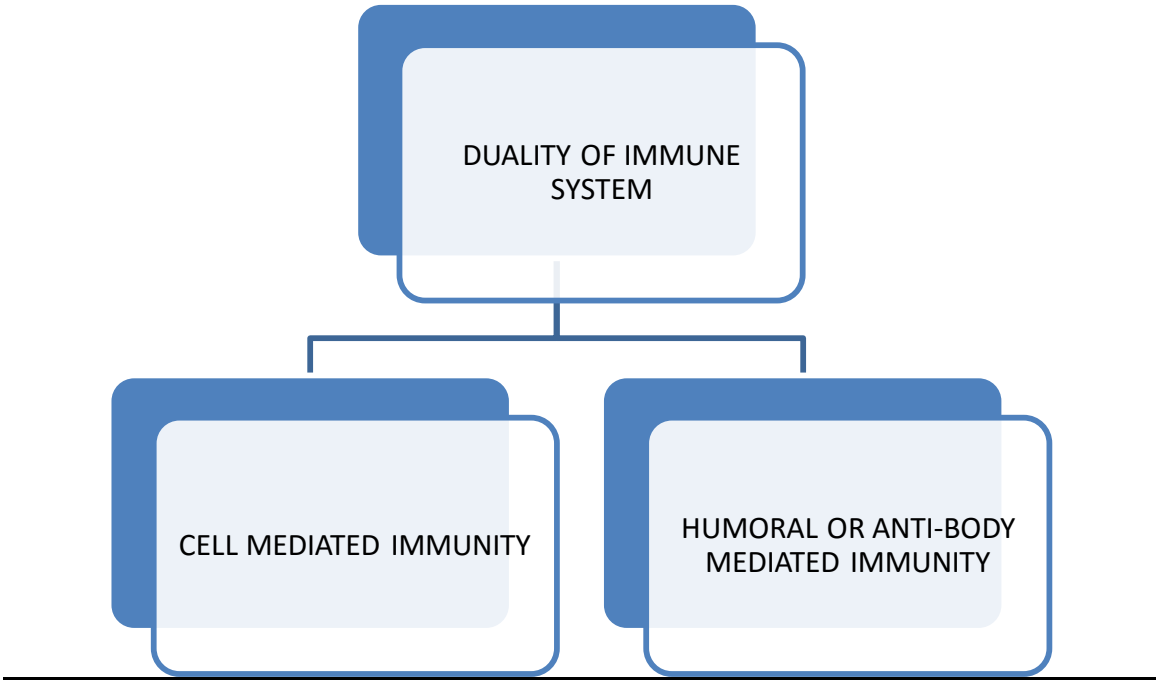
This immunity develops when antibodies pass into fetal circulation from mother blood through placenta and also form colostrums.

Difference between active and passive immunity

	Active immunity		**Passive immunity**
1.	Active immunity is the immunity that develops from exposure to antigen that lead to production .	**1.**	immunity develops from anti-bodies
2.	it develop slowly not become fully effective even for several weeks.	2.	It is the fast process because we directly introduce anti-bodies. Established quickly the time depends upon route of administration.
3.	Once developed it is long lasting. it after gives protection for many years even when it begins to fade a booster dose of antigen quickly restore the effective anti-bodies level.	3.	It is very short lived because body recognize the antibodies as foreign substance and gradually eliminate them. Protection is only for 2-3 weeks. exception is, when human anti-bodies are given, the rate of elimination is slower in this case.
4.	Active immunity is mainly used for prevention of diseases.(prophylaxis)	4.	It is both for prophylaxis therapeutic purpose.
5	Examples: polio vaccine, tuberculosis vaccine.	5.	Examples: Tetanus anti toxin to prevent tetanus after road accident, Diphtheria anti toxin.

IV. Duality of immune system:

In vertebrates two parallel immune responces have been award namely cellular or **cell mediated** immune response and **humoral** or anti-body mediated immune response to cope with widely different categories of invaders to defend some classes of organism both humoral or cellular responsees are needed. Antigens, which are generally very large and complex, are not recognized in their entirety by lymphocytes. Instead, both B and T lymphocytes recognize discrete sites on the antigen called antigenic determinants, or epitopes



A. <u>Cell mediated immune response:</u>

This response is not due to antibodies but it is due to sensitized **thymus derived lymphocytes** that is **T-lymphocytes**'-cells develop from stem cells in the bone marrow. Mature in thymus gland where they are ready to in counter antigen. The cell-mediated branch (T cells) recognizes protein epitopes displayed together with MHC molecules on self-cells, including altered self-cells such as virus-infected self-cells and cancerous cells. T-lymphocytes recognize invading organism as foreign and initiate chain reaction as follows.

1. Cytotoxic destruction of invading cells.
2. Activation of phagocytic macrophages. This system is primarily responder to transplanted tissues such as foreign skin graft. It response is to reject foreign tissue. It is also an important factor in our defense against cancer.
3. The cell mediated immune response is most effective against bacteria, viruses located within phagocytic and infected host cells and against fungi, protozoa, and helminth.

B. <u>Humoral or anti-body mediated immunity:</u>

It involves the production of anti-bodies that act against foreign organism and substances. Cells called B-cells or B- lymphocytes are responsible for production of anti-bodies. These anti-bodies are found in extra-cellular fluid such as plasma, lymph and mucous secretion. The humoral branch (B cells) recognizes an enormous variety of epitopes: those displayed on the surfaces of

bacteria or viral particles, as well as those displayed on soluble proteins, glycoproteins, polysaccharides, or lipopolysaccharides that have been released from invading pathogens. The humoral immune response defend primarily against bacterial toxin, bacteria and virus that are circulating freely in the body fluids. It is also a factor in some reactions against transplanted tissues.

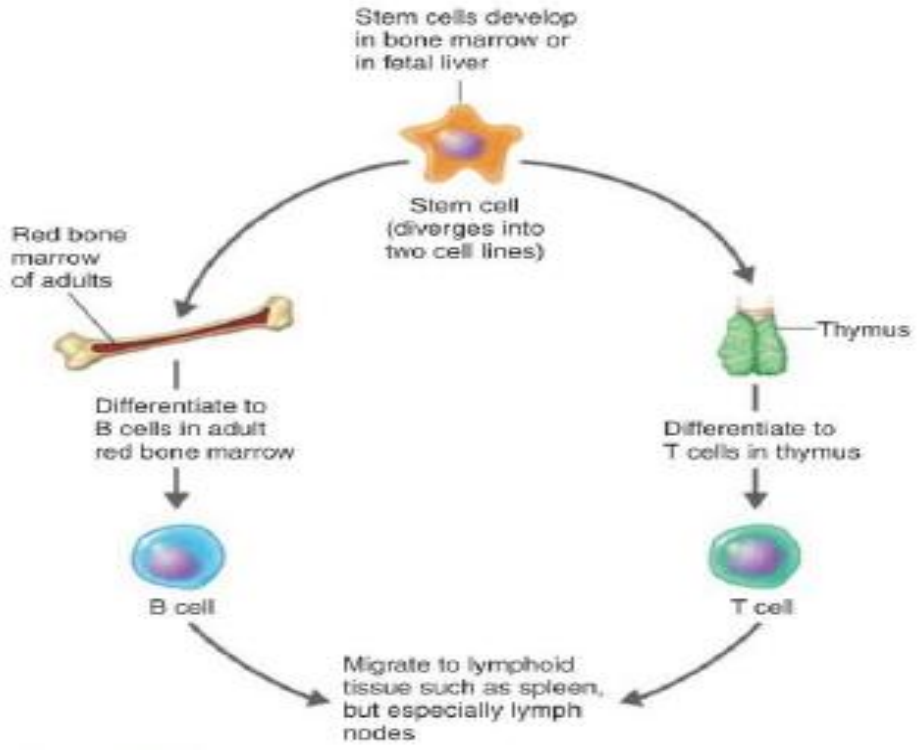
The lymphatic system

The lymphatic system consists of a fluid called *lymph,* vessels called *lymphatic vessels,* a number of structures and organs containing *lymphoid tissue,* and *red bone marrow,* where *stem* cells develop into blood cells, including lymphocytes

Lymphocytes:

Lymphocytes are basic cell responsible for both humoral and cellular immunity. Bone marrow stem cells are ultimate organ of erythrocytes and all leucocytes including lymphocytes production. Many lymphocytes pass through thymus where they become processed by hormonal micro environment try to release. These lymphocytes are now called **thymus derived** or **lymphocytes T-cells**. During its maturation within the thymus, the T cell comes to express a unique antigen-binding molecule, called the T-cell receptor, on its membrane. Unlike membrane-bound antibodies on B cells, which can recognize antigen alone, T-cell receptors can recognize only antigen that is bound to cell-membrane proteins called major histocompatibility complex (MHC) molecules. There are two well-defined subpopulations of T cells: T helper (TH) and T cytotoxic (TC) cells. T helper and T cytotoxic cells can be distinguished from one another by the presence of either CD4 or CD8 membrane glycoproteins on their surfaces.T cells displaying CD4 generally function as TH cells, whereas those displaying CD8 generally function as TC cell

By majority of bone marrow derived lymphocytes which do not enter or become processed by thymus are called **B-lymphocytes** or **B-cells**.



Lymphatic system consist of

A. Primary lymphoid system

B. Secondary lymphoid system

C. <u>Primary lymphoid system:</u>

Primary lymphoid organ provide environment for development, differentiation and maturation of cells. Thymus and bone marrow.

D. <u>Secondary lymphoid system:</u>

Those organ where mature B and T lymphocytes settle and initiate immune response **e.g.** lymph nodes ,spleen, liver etc

ANTIGEN:

ANTIGEN is derived from two words. ANTI means antibody and GEN means generation. Antigen are substances which help in generation of antibody.

DEFINITION:

Substances which when inoculated into animal body provoke new response by stimulating the cells of immune system to produce modified globulin called anti bodies.

PROPERTIES OF ANTIGEN:

Antigen exhibit following properties.

IMMUNOGENECITY:

Antigen should have ability to stimulate immune system.

REACTIVITY:

Antigen should have ability to react with immune components

CHEMICAL NATURE OF ANTIGEN

Most antigens are either protein or large polysaccharide. Lipids and nucleic acids are antigen only when combined with proteins and polysaccharides

TYPES

There are following types of antigen.

1-Microbial antigen

2-Non microbial antigen

MICROBIAL ANTIGEN:

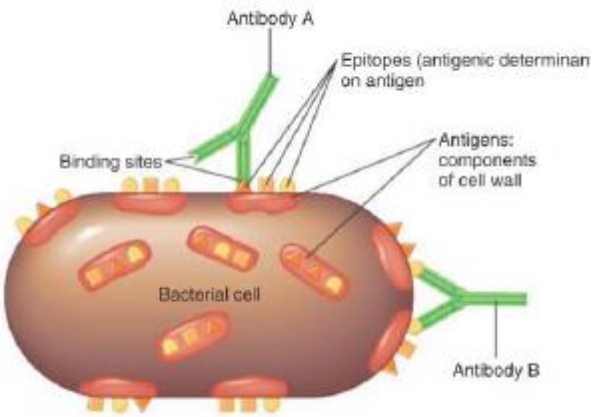
Antigenic compounds are often comprises of invading microorganism such as capsules, cellwall, flagella,pilli,and toxins of bacteria,Coats of viruses or surface molecule of other types of microbes.

NON MICROBIAL ANTIGEN:

They include pollens, egg white ,blood cell surface molecules, Serum proteins from other individual or species and surface molecules of transplanted tissue and organs.

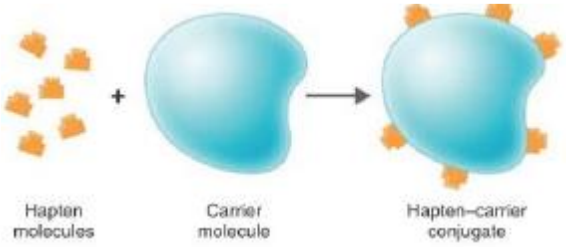
ANTIGENIC DETERMINENT

Generally antibodies recognize and interact with specific region on antigen called antigenic determinant or epitope.



Most antigen have a molecular weight of 10000 Dalton or higher. A foreign substance that has a low molecular weight is often non antigenic unless attached to a corner molecule .These small molecules are hapten.

e.g. Penicillin is a good example of hapten.



This drug is non antigenic by itself but some people develop an allergic reaction to it. In the people penicillin combine with serum proteins. The resulting combined molecule initiate immune response.

ANTIBODIES

Antibodies are proteins that are made in response to antigen and can recognize and bind to that antigen. Antibodies can therefore help to neutralize or destroy that antigen. Antibodies are highly specific in recognizing the antigen and antigens such as bacterium or virus may have several

antigenic determinents or epitopes that cause production of different anti bodies. Each antibody has at least two identical sites that bind to antigenic determinants. These sites are known as antigen binding sites.

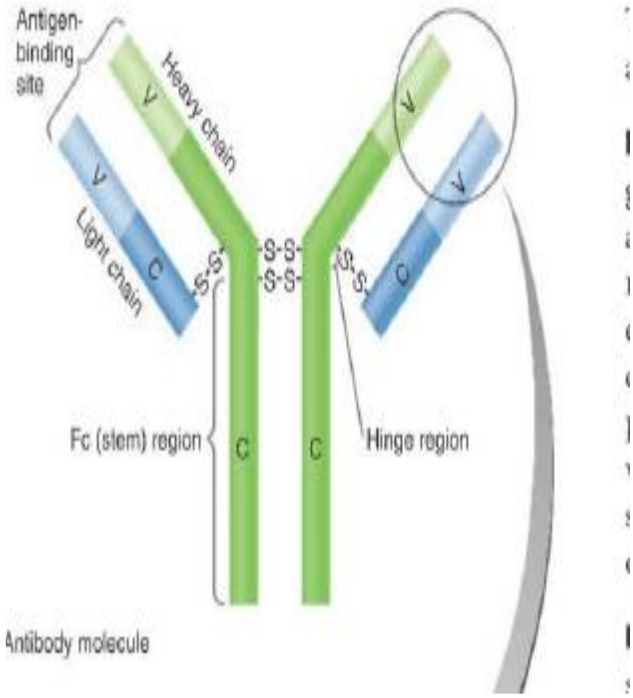
VALENCY:

No. of antigen binding sites is called valency of that antibody.

e.g. Most human antibodies have two binding sites therefore are Bivalent.

ANTIBODY'S STRUCTURE:

Because of bivalent antibody have simple molecular structure, it is called as monomer. A typically monomer have 4 proteins chains. Two identical light chains[L CHAIN],two identical heavy chain[H CHAIN].Light and heavy chains refer to their relative molecular weight .These chain are joined by disulphide links to form a ''Y''shaped structure. Constant and variable region are exist in each chain.



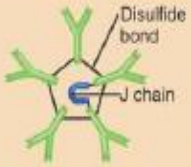
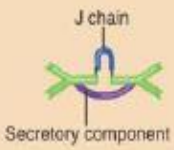
VARIABLE REGION :

Variable region structure reflects the nature of antigen for which they are specific.

CONSTANT REGION:

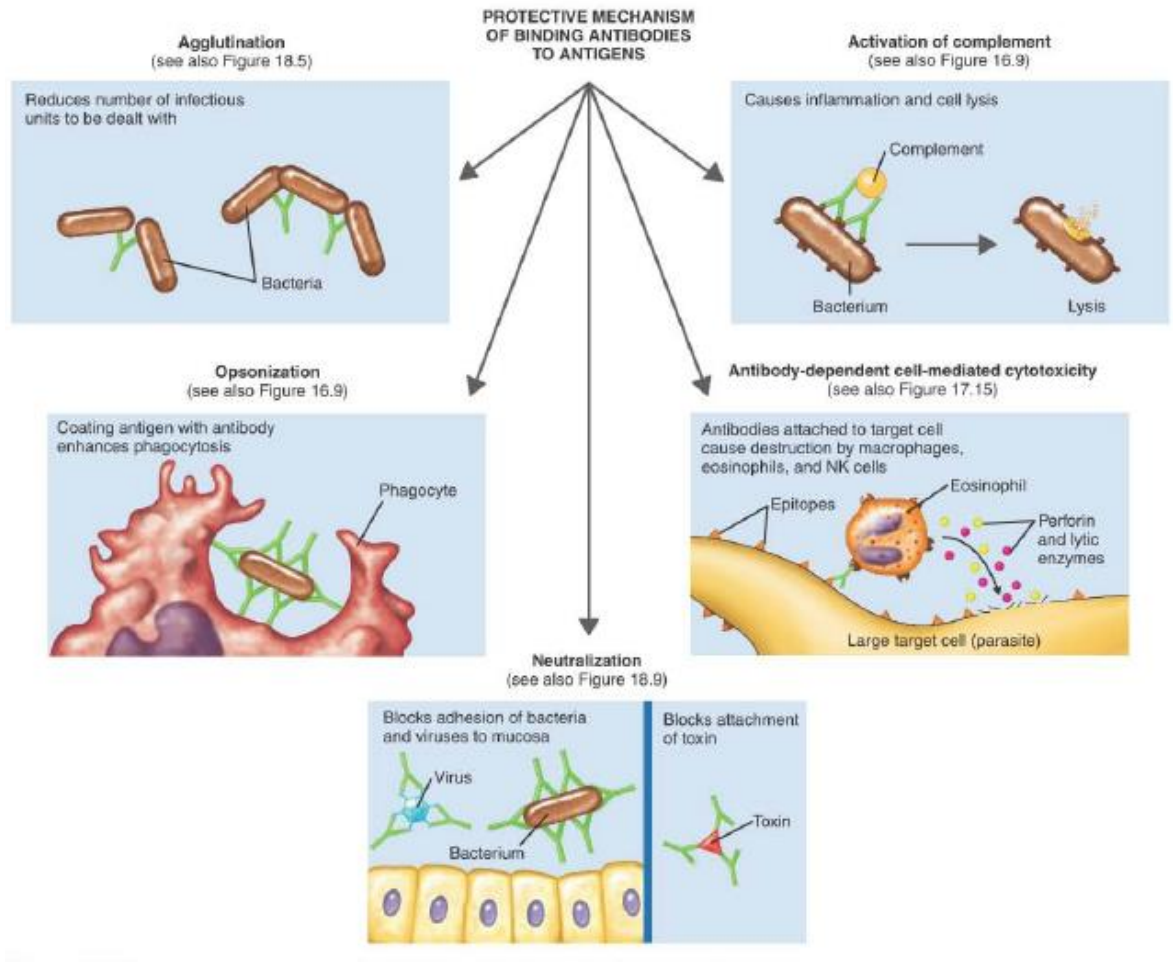
There are 5 major types of constant for 5 major classes of immunoglobulins. Stem of 'Y'shaped monomer antibodies called Fc region Fc fragment. So named because in the early days when antibody structure was being identified,it was a fragment that crystallized in cold storage. Fc region are often important in immunological reactions.

Table 17.1 A Summary of Immunoglobulin Classes

Characteristics	IgG	IgM	IgA	IgD	IgE
					
Structure	Monomer	Pentamer	Dimer (with secretory component)	Monomer	Monomer
Percentage of Total Serum Antibody	80%	5–10%	10–15%*	0.2%	0.002%
Location	Blood, lymph, intestine	Blood, lymph, B cell surface (as monomer)	Secretions (tears, saliva, mucus, intestine, milk), blood, lymph	B cell surface, blood, lymph	Bound to mast and basophil cells throughout body, blood
Molecular Weight	150,000	970,000	405,000	175,000	190,000
Half-Life in Serum	23 days	5 days	6 days	3 days	2 days
Complement Fixation	Yes	Yes	No†	No	No
Placental Transfer	Yes	No	No	No	No
Known Functions	Enhances phagocytosis; neutralizes toxins and viruses; protects fetus and newborn	Especially effective against microorganisms and agglutinating antigens; first antibodies produced in response to initial infection	Localized protection on mucosal surfaces	Serum function not known; presence on B cells functions in initiation of immune response	Allergic reactions; possibly lysis of parasitic worms

*Percentage in serum only; if mucous membranes and body secretions are included, percentage is much higher.
†May be yes via alternative pathway.

ANTIGEN ANTIBODY REACTIONS



Immunology serves as diagnostic tool, used for the detection of diseases by interaction ofantibodies with antigens. These reactions include:

1: Agglutination reaction

2: Precipitation reaction

3: Neutralization reaction

1: Agglutination reaction:

Serological test in which antigen molecules are attached to large particles when combined with patient's serum containing complementary antibodies result in the visible clumps.

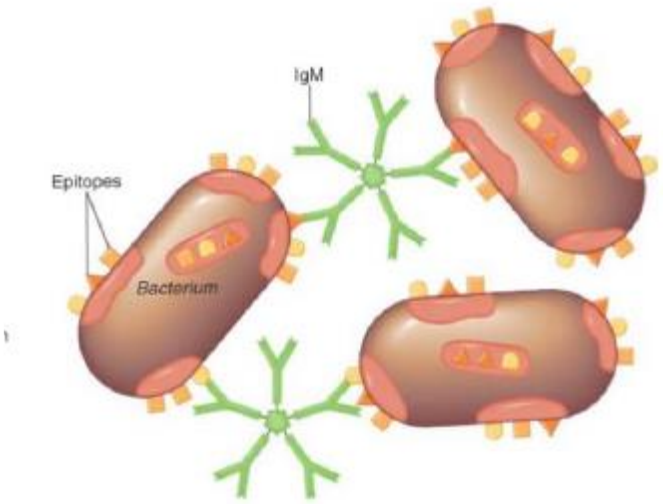


Figure 18.5 **An agglutination reaction.** When antibodies react with

Types:

A: Direct agglutination test

B: Indirect agglutination test

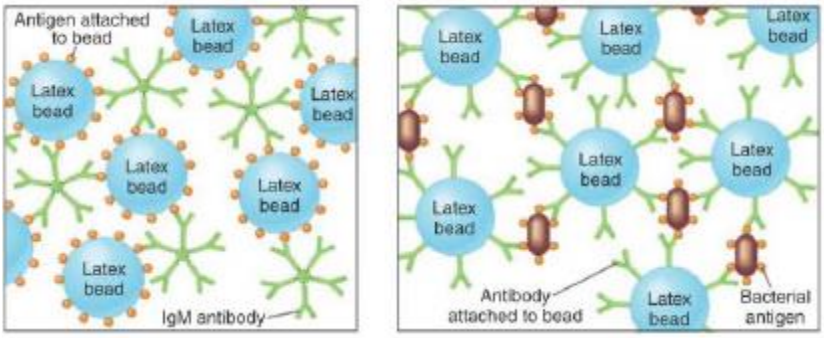
C: Heme agglutination test

Direct agglutination reaction:

Antigens are part of large molecules such as bacterium.

Indirect agglutination reaction:

Antigens or antibodies are absorbed on the surface of latex beads.



Heme agglutination reaction:

Agglutination reaction in which clumping of RBC's is carried out is called heme agglutination reaction.

Examples of agglutination reactions:

I: Widal test:

Purpose. It is used for diagnosis of typhoid.

TEST	OBSERVATION	RESULT
Typhoid bacilli	Agglutination	Positive
Person serum	No agglutination	Negative

II: Weil-Fliex Test:

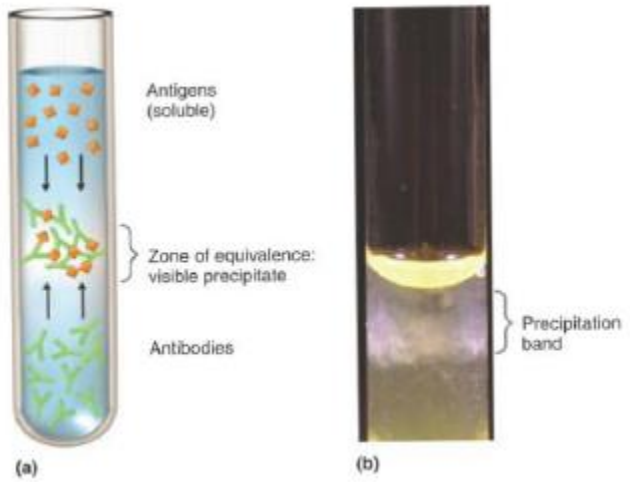
Purpose. It is used for diagnosis of Rickettsial infection.

TEST	OBSERVATION	RESULT
Proteus org. + patient serum	Agglutination	Serumhas complementary antibodies so person has rickettsial infection.
Proteus org. patient serum	No agglutination	Serum has no complementary antibodies so person doesn't have rickettsial infection.

III: TPA Test: (Treponema Pallidium Agglutination Test).

TEST	OBSERVATION	RESULT
Living or dead treponema + patient serum.	Agglutination	Positive
Living or dead treponema + patient serum.	No agglutination	Negative

2: Precipitation Reaction:



Antibody and soluble antigen interacting in aqueous solution form a lattice that eventually develops into a visible precipitate. Antibodies that aggregate soluble antigens are called precipitins. Although formation of the soluble Ag-Ab complex occurs within minutes, formation of the visible precipitate occurs more slowly and often takes a day or two to reach completion.

Precipitation reaction should be used in following ways:

A: Tube precipitation test

B: Slide precipitation test

C: Gel effusion test

Diagnostic application:

It is used for diagnosis of anthrax by Ascoli Test. Although various modifications of the precipitation reaction were at one time the major types of assay used in immunology, they have been largely replaced by methods that are faster and, because they are far more sensitive, require only very small quantities of antigen or antibody. Also, these modern assay methods are not limited to antigen-antibody reactions that produce a precipitate.

3: Neutralization Test:

It is an **antigen-antibody reaction** in which the harmful effects of bacterial exotoxin or virus are blocked by specific antibodies. The antitoxin combines with the exotoxin to neutralize it. Antitoxins produced in animals can be injected in humans to provide passive immunity against the toxin. A more frequently used neutralization test is the viral heme agglutination inhibition test. This test is used in the diagnosis of influenza, measles, mumps and a number of other infections caused by viruses that can agglutinate RBC's.

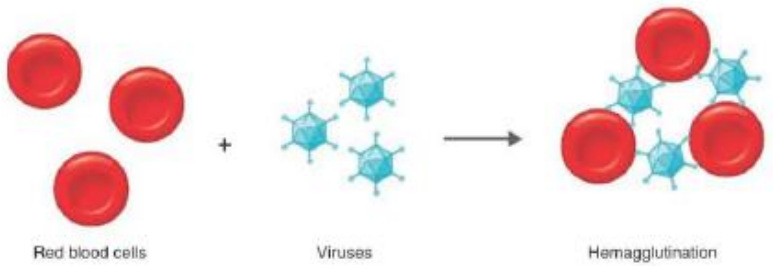
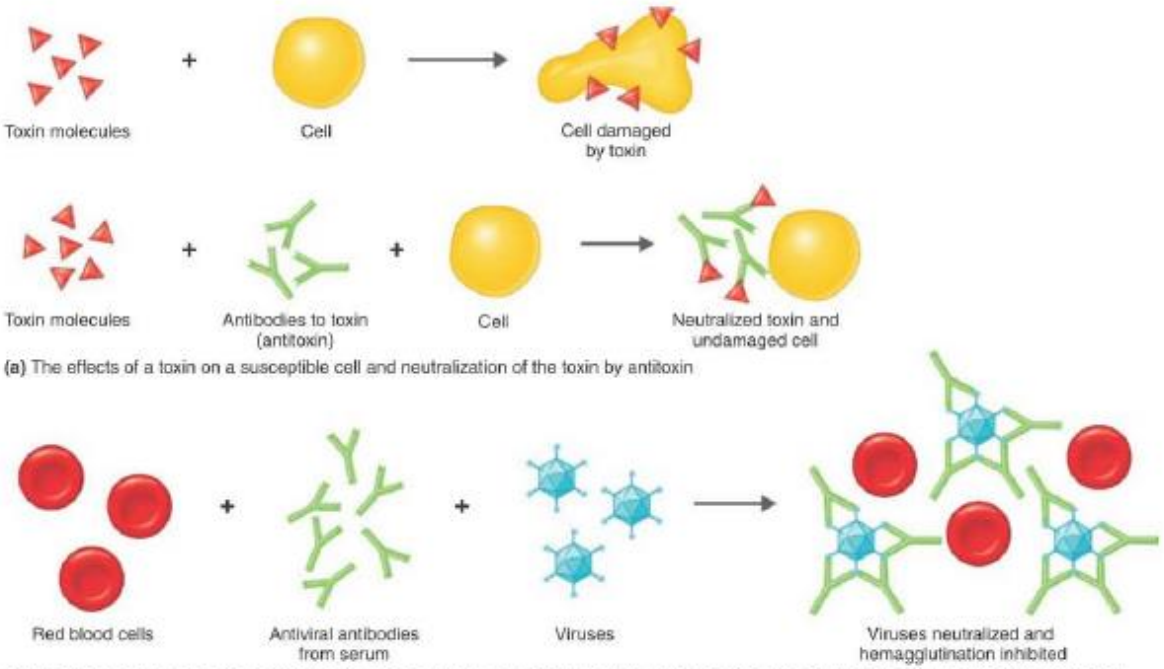


Figure 18.8 Viral hemagglutination. Viral hemagglutination is not an antigen–antibody reaction.

Q **What causes agglutination in viral hemagglutination?**

If a person serum contains antibodies against these viruses, antibodies will react with the viruses and neutralize them e.g. if heme agglutination occurs in a mixture of measles virus and red blood cell but does not occur when the patient serum is added in the mixture. Result indicate that serum contain antibody that have bound to and neutralize the measles virus.



(a) The effects of a toxin on a susceptible cell and neutralization of the toxin by antitoxin

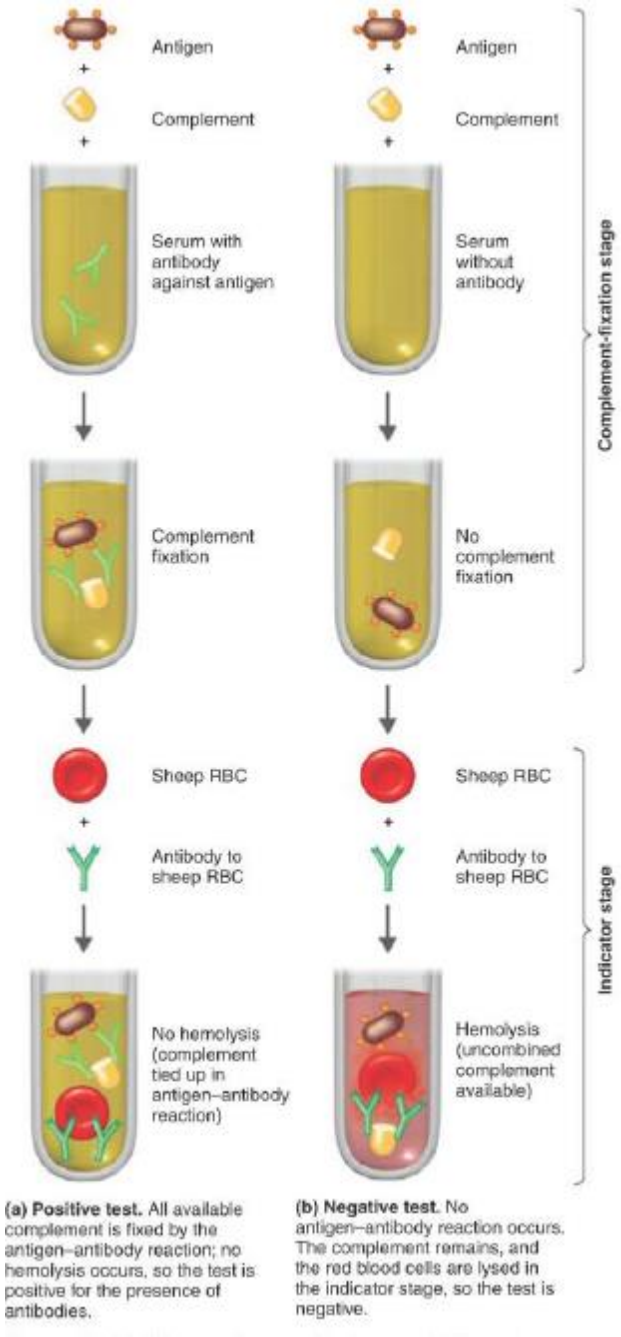
(b) Viral hemagglutination test to detect antibodies to a virus. These viruses will normally cause hemagglutination when mixed with red blood cells. If antibodies to the virus are present, as shown here, they neutralize and inhibit hemagglutination.

Figure 18.9 Reactions in neutralization tests.

Complement-Fixation Reactions

During most antigen- antibody reactions, complement binds to the antigen-antibody complex and is used up, or fixed. This process of complement fixation can be used to detect very small amounts of antibody. Antibodies that do not produce a visible reaction, such as precipitation or agglutination, can be demonstrated by the fixing of complement during the antigen- antibody reaction. Complement fixation was once used in the diagnosis of syphilis (Wassermann test) and is still used to diagnose certain viral, fungal, and rickettsial diseases. The complement-fixation test requires great care and good controls, one reason the trend is to replace it with newer, simpler tests.

The test is performed in two stages: complement fixation and indicator



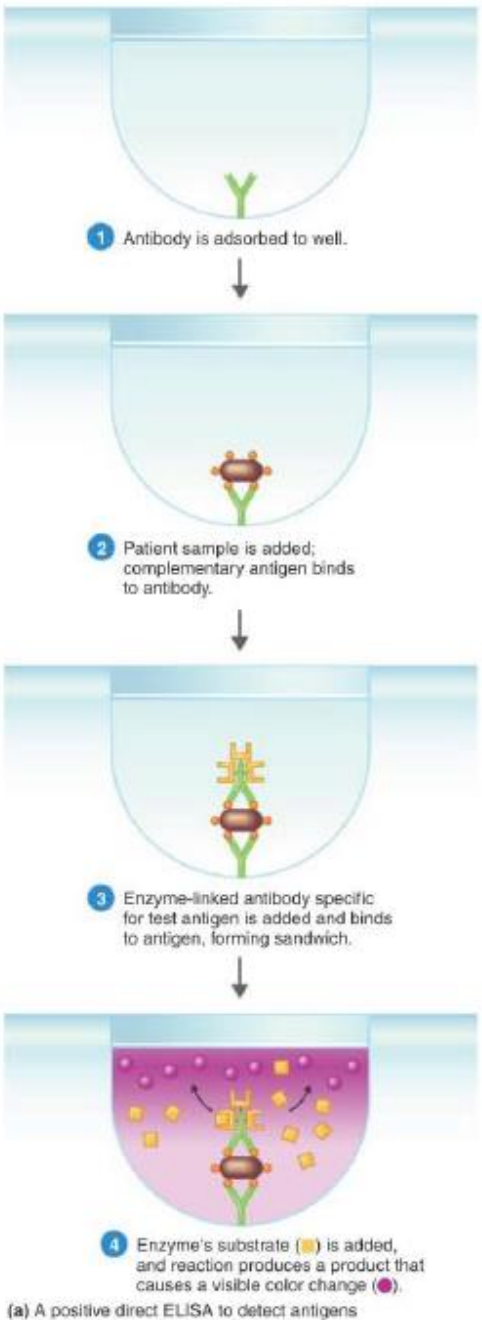
(a) Positive test. All available complement is fixed by the antigen–antibody reaction; no hemolysis occurs, so the test is positive for the presence of antibodies.

(b) Negative test. No antigen–antibody reaction occurs. The complement remains, and the red blood cells are lysed in the indicator stage, so the test is negative.

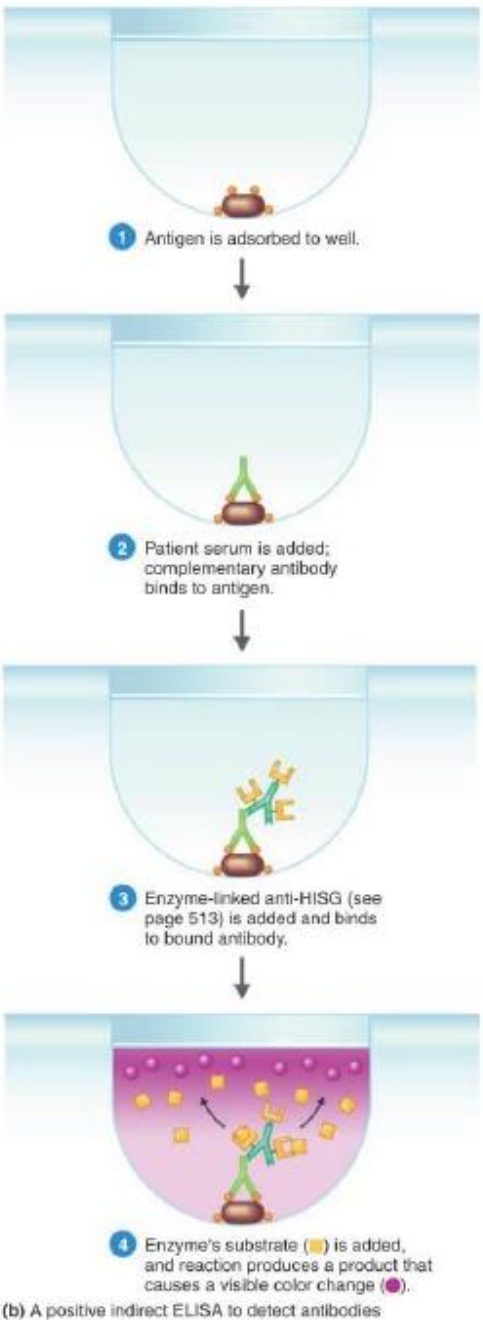
ELISA

The enzyme-linked immunosorbent assay (ELISA) is the most widely used of a group of tests known as *enzyme immunoassay (EIA).* There are two basic methods. The *direct ELISA* detects antigens, and the *illdirect ELISA* detects antibodies. A microtiter plate with numerous shallow wells is used in both procedures Variations of the test exist; for example, the reagents can be

bound to tiny latex particles rather than to the surfaces of the microtiter plates. ELISA procedures are popular primarily because they require little interpretive skill to read; the results tend to be clearly positive or clearly negative



(a) A positive direct ELISA to detect antigens



(b) A positive indirect ELISA to detect antibodies

VACCINATION

Vaccine is derived from Latin word **VACCA** means **Cow**. **Vaccination** is artificial introduction of active immunity by administration of non-pathogenic form of disease causing agent.

Edward Jenner was founder of vaccination. It is very easy and cost affecting way of preventing illness. It is also called **prophylactic immunization.**

VACCINE

Vaccine is a formulation containing pathogenic agents modified to make them non-pathogenic and administration of which induce immune response in the recipient sufficient to prevent susceptible disease.

OR

Vaccine is the suspension of attenuated or killed micro-organisms or their antigenic portion presenting to a potential host to induce and immunologically mediated resistance to disease.

PRINCIPLE OF VACCINATION

To induce in an individual a prime state that on contact with relevant infection a more rapid and effective immune response could be generated leaving to prevention of disease.

Vaccination depends on the ability of both B & T lymphocyte to respond to specific antigen and develop into memory cell and this represent a form of actively enhanced adopted immunity. The primary aim is to eliminate the disease.

PROPERTIES OF AN IDEAL VACCINE

1: Non-pathogenic:

A vaccine should not cause disease.

2: Immunogenicity:

A vaccine should be strongly immunogenic to induce high concentration of antibodies.

3: Immunogenic specificity:

It should produce antibodies that react specifically with disease producing agent.

4: Effectiveness:

It should be effective after a single dose.

5: Long lived immunity.

6: Low cost.

7: Compatibility to co-administer vaccine.

8: Stability:

It should be stable (genetically and thermal stability).

9: Response:

It should induce a wide range of appropriate responses (humoral, cellular and mucosal immunity).

Types of Vaccine:

There are three types of vaccine.

1: First Generation Vaccine.

2: Second Generation vaccine.

3: Third Generation Vaccine.

First Generation Vaccine:

Those vaccines which are prepared from whole organisms whether live attenuated or killed (bacterial or viral) are **first generation vaccines**.

A: Live attenuated vaccines:

Attenuation is a process of reducing virulence or toxic properties of micro-organism. Aim of attenuation is to decrease toxicity and maintain antigenicity.

Methods to achieve attenuation:

1: Growing the organism at low temperature e.g. attenuation of influenza virus by growing at low temperature.

2: By passing through unnatural host e.g. polio virus was attenuated by passage through monkey kidney cell.

3: By treatment with mutagenic.

DISADVANTAGES:

1: Probability of reversal of attenuation.

2: They should not be given to immune-compromised persons.

3: Difficult and expensive to develop due to need to prove that micro-organism is non-pathogenic.

ADVANTAGES:

It gives strong and long lasting immunity due to amplification of immunogenic stimulant by growth of micro-organism in the body.

B: Killed Vaccines:

Here pathogens are killed in such a way that the pathogenicity is reduced and terminated and antigenicity is maintained.

METHODS:

1: By heating at 56^0C for 1 hour.

2: By treatment with chemical such as phenol 0.5% for bacterial vaccine.

3: β-propiolactone for viral vaccine.

ADVANTAGES:

1: These are relatively safe.

2: Relatively easy and cheap to produce.

3: Their safety testing is simpler.

DISADVANTAGES:

1: Immunity is weak and short lived as these organisms have no capacity to multiply so boaster dose is essential for killed vaccine.

2: Allergic reactions may occur in some patient.

Second Generation Vaccines

Those vaccines which consist of defined naked or recombinant component derived from organisams by biochemical purification or Genetic engineering.

1)Purified macromolecules as vaccines

In this case specific molecules are used as vaccines Forms of such vaccine include

1)Inactivated Exotoxins (Toxoid Vccine)

1)Capsular polysaccharide or polypeptide

Toxoid Vaccine

Some bacterial pathogens produce exotoxins such as *Cornebacterium diptheriae* and *Clostridium tetani* causing diphtheria and tetnus. These exotoxins can be inactivated with formaldehyde to form toxoid. In the production of antisera conditions are so adjusted to achieve detoxification without excessive modification of epitope structure.

Vaccination with toxoid induces antitoxoid antibodies which are capable of binding to toxin and neutralizing the toxic effects.

Advantage

Reduced pathogenesity as specific purified macromolecules derived from pathogens are used

ii) Recombinant antigen

Recombinant DNA technology has paved the way for development of new generation vaccine.

A Number of genes encoding surface antigen from bacteria,virus and protozon pathogens have been successfully cloned in bacteria, yeast, insect or mammalian expression system and expressed antigens are used for vaccine development . Ist recombinant antigen vaccine produced is Hepatitis B vaccine .Now a number of vaccines have been developed by this technology

Third Generation Vaccine

This include Nucleic acid vaccine. Introduction of Naked nucleic acid in vivo as a plasmid for the purpose of generating an immune response is called a naked DNA vaccine. I t is the recent technology in the field of vaccine development. It involves

- Isolation of DNA from infectious agent bacteria, virus, fungi and protozoa
- Induce eukaryotic promoters in the form of gene to DNA
- Introduction in Plasmid (Vector)
- Injection into host cell
- Intramuscular injection into skeletal muscle cardiac or diaphgram

Advantages

1) Easy to produce large quantity of pure DNA within short period of time

2) Safest no chances of reversal

3) Highly stable

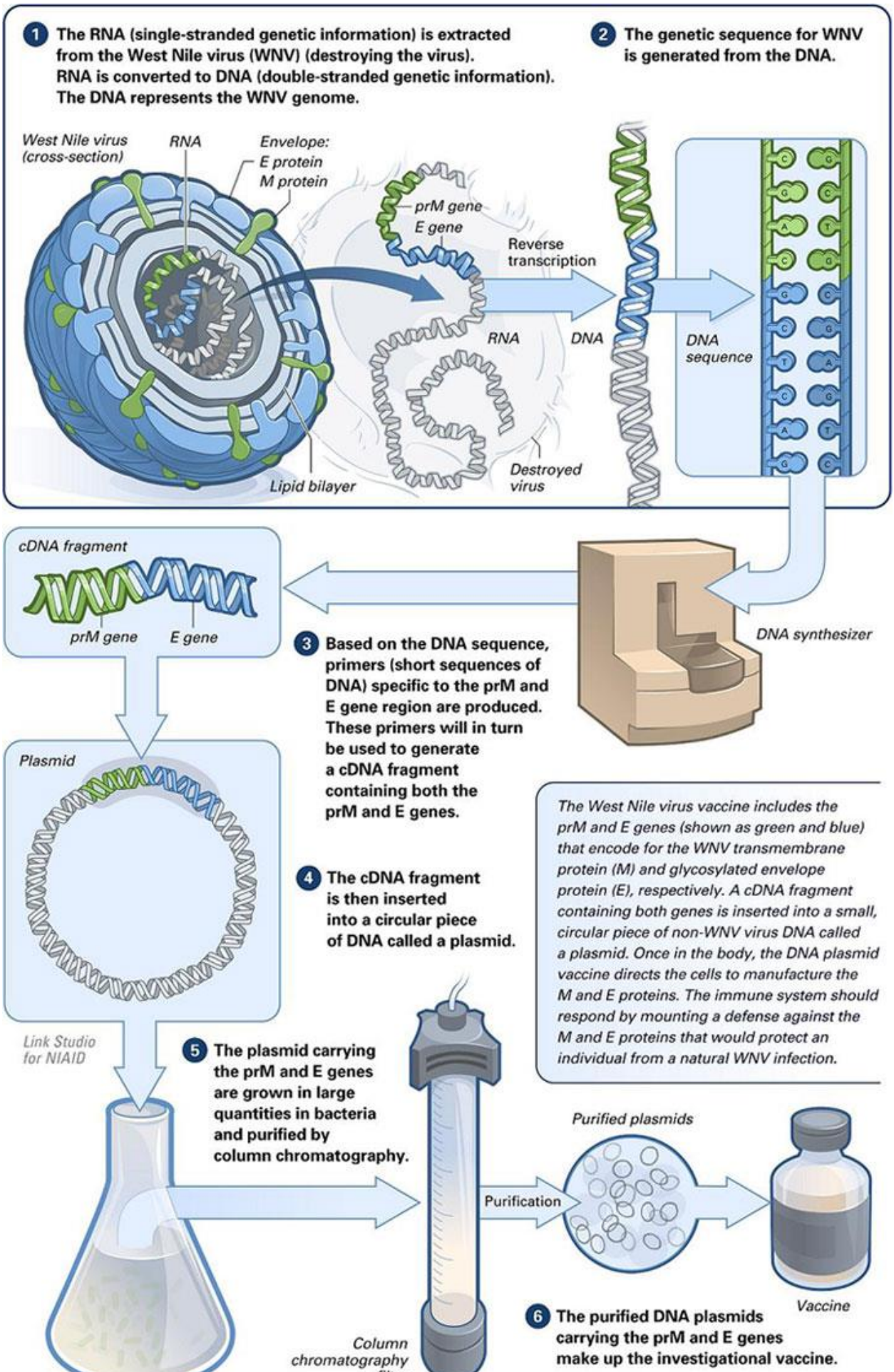

1 The RNA (single-stranded genetic information) is extracted from the West Nile virus (WNV) (destroying the virus). RNA is converted to DNA (double-stranded genetic information). The DNA represents the WNV genome.
2 The genetic sequence for WNV is generated from the DNA.
West Nile virus (cross-section)
RNA
Envelope: E protein M protein
Lipid bilayer
prM gene
E gene
Reverse transcription
RNA
DNA
DNA sequence
Destroyed virus
cDNA fragment
prM gene
E gene
DNA synthesizer
3 Based on the DNA sequence, primers (short sequences of DNA) specific to the prM and E gene region are produced. These primers will in turn be used to generate a cDNA fragment containing both the prM and E genes.
Plasmid
4 The cDNA fragment is then inserted into a circular piece of DNA called a plasmid.
The West Nile virus vaccine includes the prM and E genes (shown as green and blue) that encode for the WNV transmembrane protein (M) and glycosylated envelope protein (E), respectively. A cDNA fragment containing both genes is inserted into a small, circular piece of non-WNV virus DNA called a plasmid. Once in the body, the DNA plasmid vaccine directs the cells to manufacture the M and E proteins. The immune system should respond by mounting a defense against the M and E proteins that would protect an individual from a natural WNV infection.
Link Studio for NIAID
5 The plasmid carrying the prM and E genes are grown in large quantities in bacteria and purified by column chromatography.
Purification
Purified plasmids
Vaccine
Column chromatography
6 The purified DNA plasmids carrying the prM and E genes make up the investigational vaccine.

TABLE 18.2 Principal Vaccines Used in the United States to Prevent Viral Diseases in Humans

Disease	Vaccine	Recommendation	Booster
Influenza	Injected vaccine, inactivated virus (nasally administered vaccine with attenuated virus is now available for some)	For chronically ill, including children over 6 months. Adults over age 65. Healthy children aged 6–23 months (because higher risk of related hospitalizations). Health care workers and others in contact with high risk groups. Healthy persons aged 5–49 years can receive intranasal vaccine.	Annual
Measles	Attenuated virus	For infants aged 15 months	Adults if exposed during outbreak
Mumps	Attenuated virus	For infants aged 15 months	Adults if exposed during outbreak
Rubella	Attenuated virus	For infants aged 15 months; for women of childbearing age who are not pregnant	Adults if exposed during outbreak
Chickenpox	Attenuated virus	For infants aged 12 months	(Duration of immunity not known)
Poliomyelitis	Killed virus	For children, see Table 18.3; for adults, as risk to exposure warrants.	(Duration of immunity not known)
Rabies	Killed virus	For field biologists in contact with wildlife in endemic areas; for veterinarians; for people exposed to rabies virus by bites.	Every 2 years
Hepatitis B	Antigenic fragments of virus	For infants and children, see Table 18.3; for adults, especially health care workers, homosexual men, injecting drug users, heterosexual people with multiple partners, and household contacts of hepatitis B carriers.	Duration of protection at least
Hepatitis A	Inactivated virus	Mostly for travel to endemic areas and protecting contacts during outbreaks	Duration of protection estimated at about 10 years
Smallpox	Live vaccinia virus	Certain military and health care personnel	Duration of protection estimated
Herpes zoster	Attenuated virus	Adults over age 60	None recommended
Human papilloma virus	Antigenic fragments of virus	All females under age 26. Boys optional.	Duration at least 5 years