THEORIES OF ANTIBODIES PRODUCTION

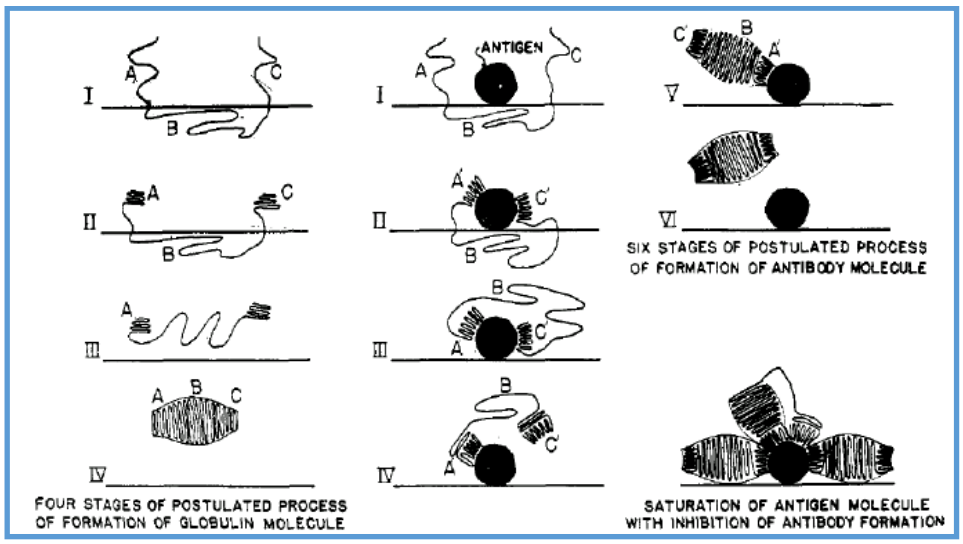
1. **Instructive Theories**
   a. Direct Template Theory
   b. Indirect Template Theory
2. **Selective Theories**
   a. Natural Selection Theory
   b. Side Chain Theory
   c. Clonal Selection Theory
   d. Immune Network Theory

INSTRUCTIVE THEORIES

Instructive theories suggest that an immunocompetent cell is capable of synthesizing antibodies of all specificity. The antigen directs the immunocompetent cell to produce complementary antibodies. According to these theories the antigen play a central role in determining the specificity of antibody molecule. Two instructive theories are postulated as follows:

Direct template theory:

This theory was first postulated by Breinl and Haurowitz (1930). They suggested that a particular antigen or antigenic determinants would serve as a template against which antibodies would fold. The antibody molecule would thereby assume a configuration complementary to antigen template. This theory was further advanced as **Direct Template Theory** by Linus Pauling in 1940s.



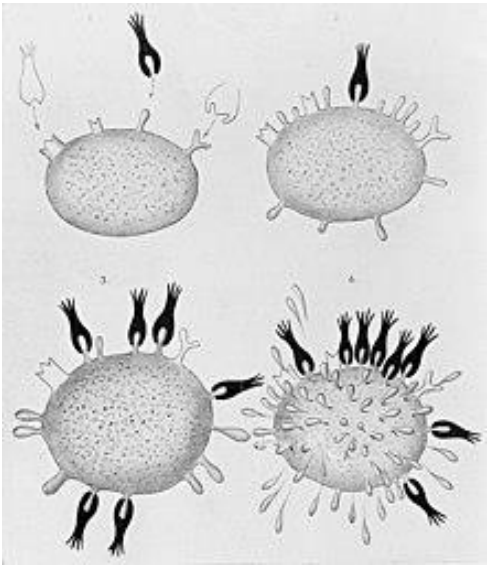
Indirect template theory:

This theory was first postulated by Burnet and Fenner (1949). They suggested that the entry of antigenic determinants into the antibody producing cells induced a heritable change in these cells. A genocopy of the antigenic determinant was incorporated in genome of these cells and transmitted to the progeny cells. However, this theory that tried to explain specificity and secondary responses is no longer accepted.

SELECTIVE THEORIES

Selective theories suggest that it is not antigen, but the antibody molecule that play a central role in determining its specificity. The immune system already possess pre-formed antibodies of different specificities prior to encounter with an antigen. Three selective theories were postulated as follows:

Side chain theory: This theory was proposed by Ehrlich (1898). According to this theory, immunocompetent cells have surface receptors that are capable of reacting with antigens, which have complementary side chains. When antigens are introduced into host, they combine with those cell receptors that have a complementary fit. This inactivates the receptors. There is an overproduction of the same type of receptors that circulate as antibodies.



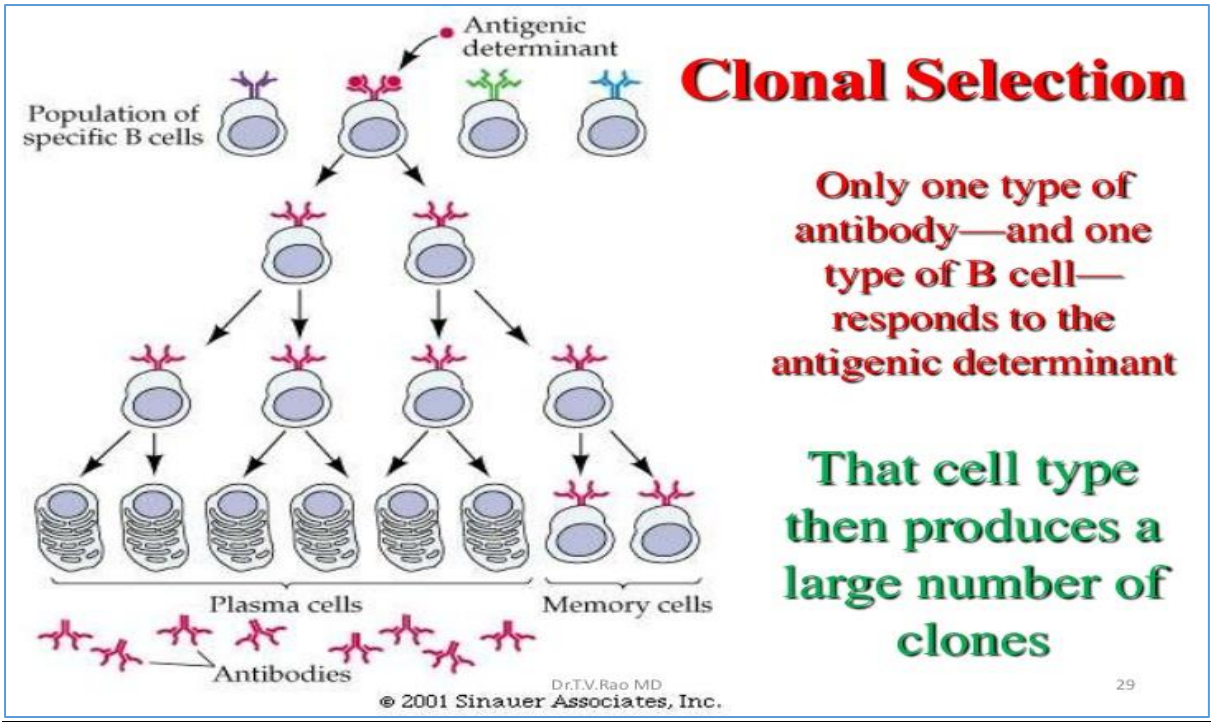
Natural selection theory: This theory was proposed by Jerne(1955). According to this theory, during the embryonic life, millions of globulin molecules were formed against all possible range of antigens. The antigen when introduced to the host combines selectively with the globulin molecule that has the nearest complementary fit. This globulin then stimulates antibody forming cells to produce more quantities of the same type of antibody.

BURNET'S CLONAL SELECTION THEORY

In 1959, F.M. Burnet proposed the clonal selection theory of antibody production. The clonal selection hypothesis states that an individual B cell expresses receptors specific to the distinct antigen, determined before the antibody ever encounters the antigen. The clonal selection hypothesis has become a widely accepted model for how the immune system responds to infection and how certain types of B and T lymphocytes are selected for destruction of specific antigens invading the body.

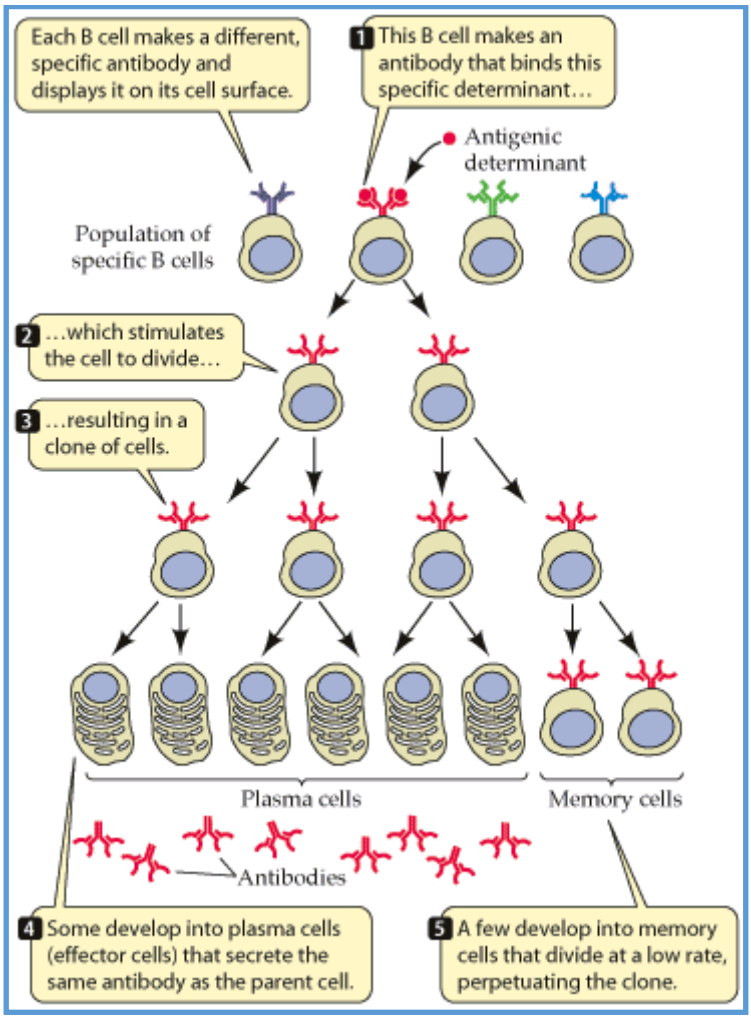
According to clonal selection hypothesis:

- The lymphoid stem cells differentiate randomly to produce different mature lymphocyte each carrying an Ag binding receptor.
- Each lymphocyte bears a unique type of receptor with a unique specificity (derived by V(D)J recombination), determined before the antibody ever encounters the antigen
- The occupation of a receptor by an antigen (epitope) is required for cell activation **(clonal selection)**, which is followed by proliferation **(clonal expansion)** of this cell to form clones.
- The activated cells of a clone differentiate into Ab producing cells, effector cells & memory cells.
- The differentiated effector cells derived from an activated lymphocyte clone will bear receptors of identical specificity as the parental cell.
- The specificity of Ab produced by a lymphocyte is identical to that of its Ag receptor.
- Those lymphocytes bearing receptors for self molecules will be deleted at an early stage.



Steps of Clonal Selection:

1. In the primary immune response, clonal selection produces effector cells and memory cells that may confer life-long immunity.
2. In the secondary immune response, memory cells are activated by a second exposure to the same antigen, which initiates a rapid and anamnestic response.
3. Random somatic hypermutations during clonal expansion cause the production of B cells with **increased antibody-binding affinity** for their antigens.
4. The **clonal selection** hypothesis may explain why secondary immune responses are so effective at preventing reinfection by the same pathogen.



It generally takes 4-5 days for a naive B- lymphocyte that has been activated to complete **clonal expansion** and differentiate into effector B-lymphocytes.Although Burnet proposed this theory for B cells and antibody production, but it is equally applicable to T cells also.

Clone: A **clone** is defined as a group of identical cells derived from a single cell.

Clonal selection: When an antigen encounters the immune system, its epitopes eventually will react only with B-lymphocytes with B-cell receptors on their surface that more or less fit and this activates those B-lymphocytes. This process is known as **clonal selection.**

JERNE'S IMMUNE NETWORK THEORY (INT)

Niels K. Jerne, M.D., was awarded the 1984 Nobel Prize in Physiology or Medicine for developing three theories that form the basis of modern cellular immunology. He shared the Nobel jointly with César Milstein and Georges J.F. Köhler (immunologists honored for developing the hybridoma technique for producing monoclonal antibodies), for his theories concerning "the specificity in development and control of the immune system."

Jerne's three main theories challenged widely held views concerning the development of antibodies and laid new foundations for contemporary immunology. Jerne published his first theory, the "natural-selection theory" of antibody formation, in 1955. At the time, immunologists believed that specific antibodies were nonexistent until their corresponding antigens entered the system and served as templates upon which the antibodies were created. Another leading theory at the time held that antigens introduced into cells were modified by enzymes and that repeated antigen exposure caused replication of antibodies that were partial replicas of these enzyme-modified antigens. Jerne challenged both of these notions, hypothesizing that all antibodies are formed during fetal development and are present in the body from birth. He suggested that when an antigen enters the body, it binds to a pre-existing complementary antibody and stimulates the rapid production of identical antibodies.

With his second theory, first set forth in 1971, Jerne sought to explain how the immune system learns to distinguish self from non-self. Immunologists at the time thought that the body's self-tolerance could not be inherited as a standard pattern but must be learned. Jerne theorized that this "learning" takes place in the thymus gland in the upper chest, where different populations of lymphocytes are exposed to histocompatibility antigens. Lymphocytes that recognize self-antigens are suppressed, whereas non-self lymphocytes, which have accumulated spontaneous mutations, develop and multiply into lymphocytes that can detect foreign antigens.

In 1974, Jerne published his third and most significant theory, his "network theory," which revolutionized the way immunologists thought about adaptive immunity and immune regulation. Jerne posited that an antibody can be produced and bind to the antigen-specific variable region of another antibody, being called as anti-antibodies, a process, which, in turn, triggers a successive cascade of anti-anti-antibody production. This cascade broadens the diversity of the antibody population, and the network attains a state of balance under normal conditions, which can be perturbed and restored during additional antigen exposures..

The idiotypic network hypothesis, formulated by Niels Jerne in 1974, postulates that the immune response may be regulated by responses to idiotypes, unique determinants originally described on B cell, and now also on T-cell receptors. This thesis is based on the dual characteristics of the B- and T-cell receptors (BCR and TCR, respectively), such that they both react with an antigen through their antigen-binding sites, these are also immunogenic, via their expression of idiotypic determinants, unique antigenic structures present in the variable region of the antibody and recognized by anti-idiotypic antibody and anti-idiotypic T cells. These interactions create a network of clones of B and T cells that express distinct idiotypic specificities that interact with each other to regulate the immune response. (Originally, Jerne viewed the idiotypic network as a BCR/Ab -cell network in which

interactions between lymphocytes required complementary V-region structures present on antibody molecules, but later on it was expanded to include T cells and their TCR.)

The variable region of an antibody molecule constitutes not only its "combining site", but also presents an antigenic profile (named its idiotype) against which anti-idiotypic antibodies can be induced in other animals. Moreover, it turned out that this antigenic, idiotypic profile of the variable region of a given antibody molecule is not a single site, but consists of several distinct sites against which a variety of different anti-idiotypic antibody molecules can be made. These individual sites are now named idiotopes, implying that the idiotype of one antibody molecule can be described as a set of different immunogenic idiotopes**. (Thus every antigen combining site of an Ig molecule expresses its own particular set of antigenic determinants called idiotopes and sum of all idiotopes is collectively called as idiotype).** Further, the immune system of a single animal, after producing specific antibodies to an antigen, continues to produce antibodies to the idiotopes of the antibodies which it has itself made. The latter anti-idiotopic antibodies likewise display new idiotypic profiles, and the immune system turns out to represent a network or web of idiotypic interactions (**idiotype anti-idiotype network)**.

These antibodies are not echoes of the invading antigen, but were already available to the animal in its repertoire of B cells before the antigen arrived. Thus, in its dynamic state, the immune system is mainly self-centered, generating anti-idiotypic antibodies to its own antibodies, which constitute the overwhelming majority of antigens present in the body. The system also somehow maintains a precarious equilibrium with the other normal self-constituents of our body, while reacting vigorously to invasions into our body of foreign particles, proteins, viruses, or bacteria, which incidentally disturb the dynamic harmony of the system. The idiotype anti-idiotype network also regulates the antibody production.

Subsequently it was shown that the idiotype anti-idiotype interactions may lead to clonal selection through expansion or elimination of specific clones. Consequently, the elements of network will be in dynamic equilibrium and the position of equilibrium is changed when antigen enters the system and induces clonal expansion of specific sets of cells. These networks of clones of B and T cells that express distinct idiotypic specificities interact with each other to regulate the immune response **(Burnet-Jerne Theory of Antibody Production)**. Idiotypes found on both antibodies that are specific for foreign and self-antigens are involved in the regulation of responses to antigen.

Idiotypic cascade - A foreign antigenic epitope (x) induces an immune response characterized by the production of Ab1 antibody, which is anti-x and contains several idiotopes, which can either be paratopic or nonparatopic (framework). Ab1 can elicit an anti-idiotypic response (Ab2), which can be of three types: (1) Ab2β, which is directed to the paratope of Ab1, and presents the internal image of antigen epitope x; (2) Ab2γ, which is directed to the paratope of Ab1 but does not carry the internal image; (3) Ab2α, which is directed to non-paratopic or framework idiotopes of Ab.

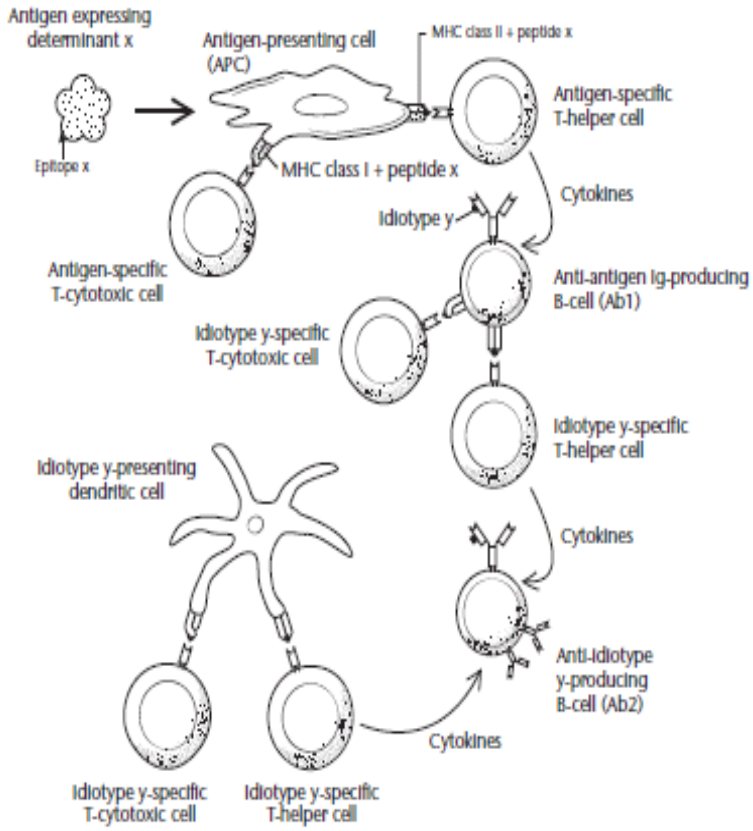


Figure 1 An antigenic epitope (x) is processed and presented by an antigen-presenting cell (APC) in association with Class II MHC to an antigen-specific T-helper cell or, in association with Class I MHC, to an antigen-specific cytotoxic T cell. Signals from the T-helper cell lead to the activation of B cells that produce anti-epitope x antibody (Ab1), which will express idiotype y. Anti-x producing B cells, as APCs, present idiotypic peptide y in association with Class II to idiotype y-reactive or specific T-helper cell and, in association with Class I, to idiotype y-specific cytotoxic T cells. Signals from the activated idiotype y-reactive T cells lead to the activation of anti-idiotope y-producing B cells (Ab2). Dendritic cells may also present idiotypic peptides to T cells.

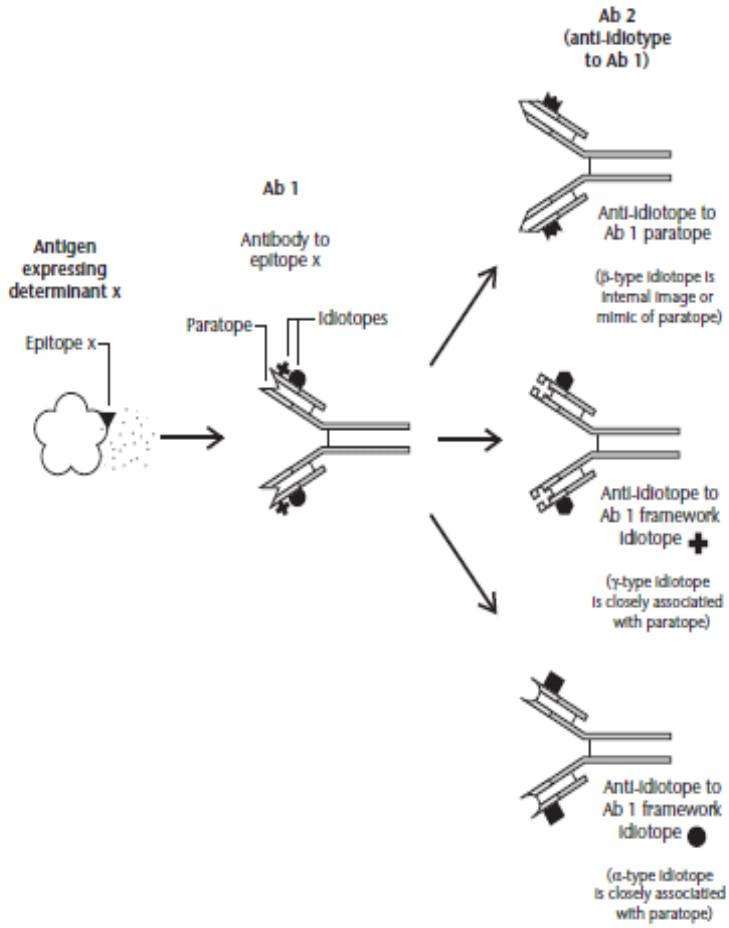


Figure 2 Idiotypic cascade. A foreign antigenic epitope (x) induces an immune response characterized by the production of Ab1 antibody, which is anti-x and contains several idiotopes, which can either be paratopic or nonparatopic (framework). Ab1 can elicit an anti-idiotypic response (Ab2), which can be of three types: (1) Ab2β, which is directed to the paratope of Ab1, and presents the internal image of antigen epitope x; (2) Ab2γ, which is directed to the paratope of Ab1 but does not carry the internal image; (3) Ab2α, which is directed to nonparatopic or framework idiotopes of Ab1.

IDIOTYPES

1. The idiotype of each V region of a single immunoglobulin molecule may be comprised of as many as 15–20 idiotopes, which can be distinguished by monoclonal anti-idiotypic antibodies or defined by a specific and unique amino acid sequence.
2. The individual idiotypes may be located either in the heavy or in the light chains and contained in the primary amino acid sequences of the variable regions. These idiotypes are more likely to be sequence-dependent, and linear antigenic determinants.
3. The hypervariable regions (also known as complementarity determining regions or CDRs) are thought to be the primary immunogenic sites within the variable region, but any part of the variable region of immunoglobulin may contribute to the structure of an idiotype.
4. In a number of systems, the CDR3 region has been shown to be the highest contributor; however, contributions by CDR1 and CDR2 are not uncommon.
5. TCR of T cells is composed of two chains, α and b, either of which can bear idiotypic antigenic determinants. However, idiotypic determinants of TCRs are generally different from those of immunoglobulins or BCR of B cells, due to the low degree of homology between V genes of immunoglobulins and those of the TCR.
6. Idiotopes may be public, CRI or IdX, or private, major or minor.
7. Regulatory idiotypes - Idiotypes s found on both antibodies that are specific for foreign and self-antigens are involved in the regulation of responses to antigen.
8. anti-ids interact with idiotypic regions in several ways: **i)** binding to the variable region outside of the antigen binding site(Ab2α); **ii)** binding to the antigen binding site($Ab2_{\beta}$), and **iii)** binding near the antigen binding site ($Ab2_{\gamma}$). $Ab2_{\beta}$, and possibly $Ab2_{\gamma}$, would mimic the structure of the antigen (epitope) as it is structurally complementary to the antigen binding site (paratope) of the original antibody (Ab1), i.e. tThe structure of paratope of anti-idiotypic Ab produced against idiotypic determinant located in antigen binding site (paratope) of Ab1 (Id) will be exact copy of the original epitope (internal image idiotope).
9. The internal image idiotopes can serve as surrogates for foreign antigens, and thus anti-idiotypic antibody can be used as vaccines to bacterial, viral and parasitic antigens.
10. An anti-Id vaccine has reached the market. Racotumomab (Vaxira) is now the first approved anti-Id vaccine—with approval in Cuba and Argentina. Vaxira was shown to increase the survival of Non-Small Cell Lung Cancer patients in recurrent or advanced stages (IIIB/IV).

Figure 1. Antibody Structure

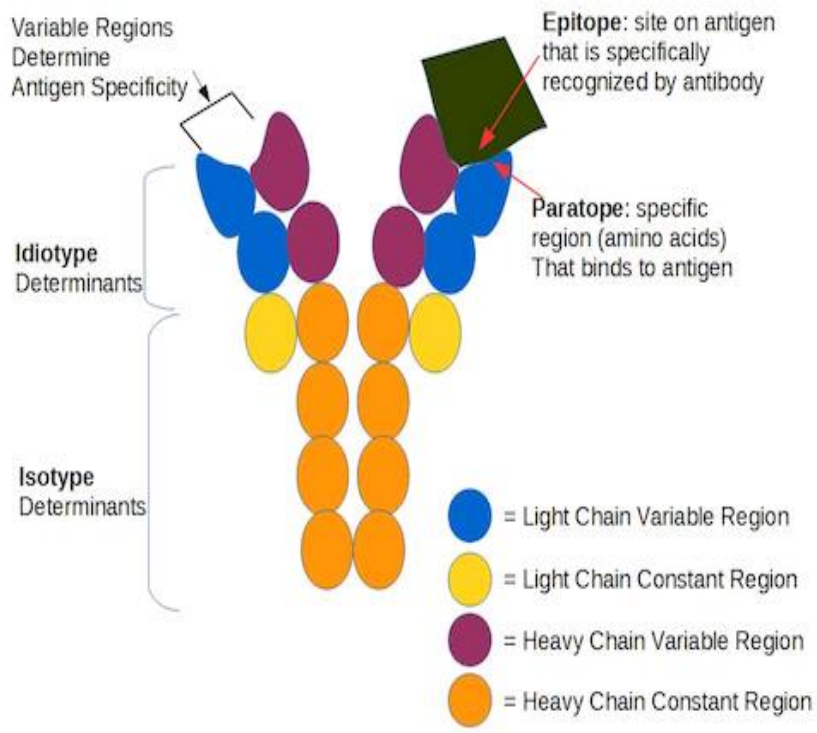


Figure 2. Anti-idiotype classes