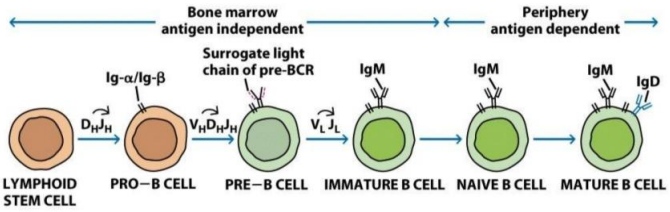
Humoral immunity

B Cells Maturation

•B cells matures in bone marrow independent of antigen, then continue to maturate in peripheral lymphoid organs with the presence of antigen
•Three main steps of maturation:
1. Progenitor- Ig alpha and beta- for signal transduction (long tails)
2. Pre-B cell- IgM heavy chain, and light chain
3. “mature”- IgD

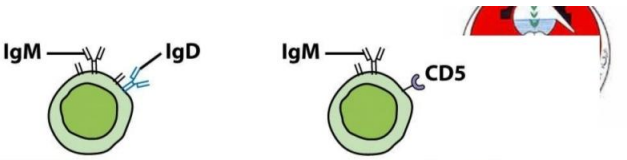


B cells Clonal Selection

-Self-reactive B cells are eliminated in bone marrow (BM).
- BM produces 5 x 107 B cells/day, but only 5 x 106 B cells/day or 10% actually enter the circulation.
- Some of this loss is due to negative selection and elimination or clonal deletion of immature B cells expressing auto antibodies to self-antigens.
- “Cross-linking” of mIgM by self Ag may lead to cell death or anergy
The clones of lymphocytes that can be interacted with corresponding Ag will be selected and lead to activation, proliferation, producing Ab and specific memory cells.

B-1 B cells

“Innate-like” subset of B cells.
•Appear during fetal life and express IgMbut little IgD and display CD5.
•Are also found in peritoneum and pleural space.
• Originates from stem cell in bone marrow, but also from proliferation of B1 Cells outside the BM.
• Responds poorly to protein antigen, but strongly to carbohydrate antigens.
• Antibodies produced are of low affinity.
• No memory produced



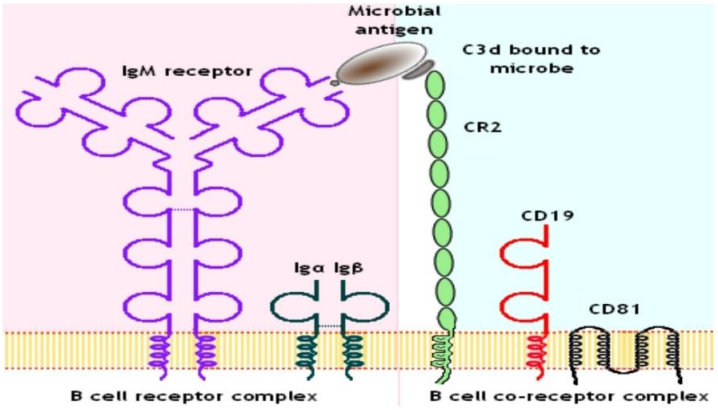
Attribute	Conventional B cells (B-2 B cells)	B-1 B cells
Major sites	Secondary lymphoid organs	Peritoneal and pleural cavities
Source of new B cells	From precursors in bone marrow	Self-renewing (division of existing B-1 cells)
V-region diversity	Highly diverse	Restricted diversity
Somatic hypermutation	Yes	No
Requirements for T-cell help	Yes	No
Isotypes produced	High levels of IgG	High levels of IgM
Response to carbohydrate antigens	Possibly	Definitely
Response to protein antigens	Definitely	Possibly
Memory	Yes	Very little or none
Surface IgD on mature B cells	Present on naive B cells	Little or none

T-independent B cell activation

• T cell independent Ags (LPS, lipids, nucleic acid and protein)
• B1 cells response to multivalent non-protein antigens with repeating determinants, such as polysaccharides, some lipids, and nucleic acids, does not require antigen-specific helper T lymphocytes.
• Multivalent Antigens (called so because each antigen molecule contains multiple identical epitopes) are therefore called T independentantigens.
• These responses are elicited by engagement of the B cell receptor (BCR) with the antigen (mainly B1 cells)

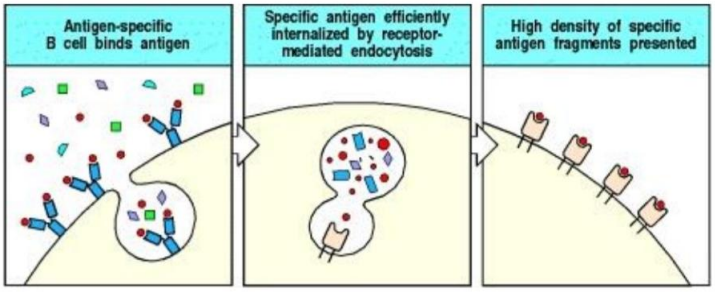
• B cell activation needs 3 signals
– Recognizing the antigen by membrane Igs and the signal transduce to inside cells by Ig alpha and beta
– Recognizing C3d complement on microbe by CR2 on B cell
– activation of Toll-like receptors (TLRs) on B cells by molecules (pathogen-associated molecular patterns [PAMPs]) derived from the microbe
• Both 3 signals enough to activate B cells and for proliferation and IGM antibody formation
• Some activated B cells differentiate into short-lived antibody secreting plasma cells,

• Low affinity Igs, mainly IGM, no memory B cells



B cells antigen presentation function

• Surface immunoglobulin (IGM or IGD) allows B cells to bind and internalize specific soluble intact antigen very efficiently. The internalized antigen is processed in intracellular vesicles where it binds to MHC class II molecules. These vesicles are then transported to the cell surface where the MHC class II-antigen complex can be recognized by Th2 cells. Because of high specificity, it is perfect when Ag concentration is low.



T-dependent B cell activation

Antigen binding in B cells

• T cell dependent Antigens (always proteins)

• First, the antigen that is presented to B cells is generally in its intact, native conformation and is not processed by antigen presenting cells. This, of course, is one of the important distinctions between the forms of antigens recognized by B and T lymphocytes

• Second, the receptor internalizes the bound antigen into endosomal vesicles, and if the antigen is a protein, it is processed into peptides that may be presented on the B cell surface for recognition by helper T cells.

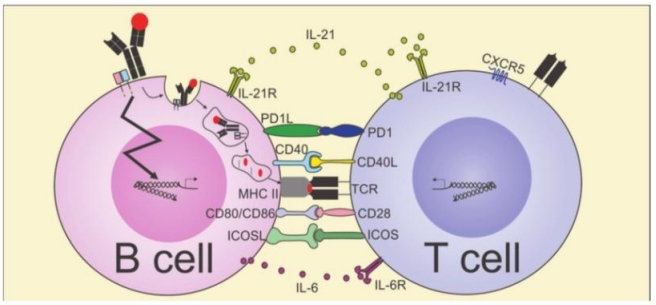
T dependent B cell activation, protein antigens,
– Helper T cell–dependent B cell responses to protein antigens require initial activation of naive T cells in the T cell zones and of B cells in lymphoid follicles in lymphoid organs to the same antigen. The activated lymphocytes migrate toward one another and interact at the edges of follicles, where the B cells present the antigen to helper T cells..

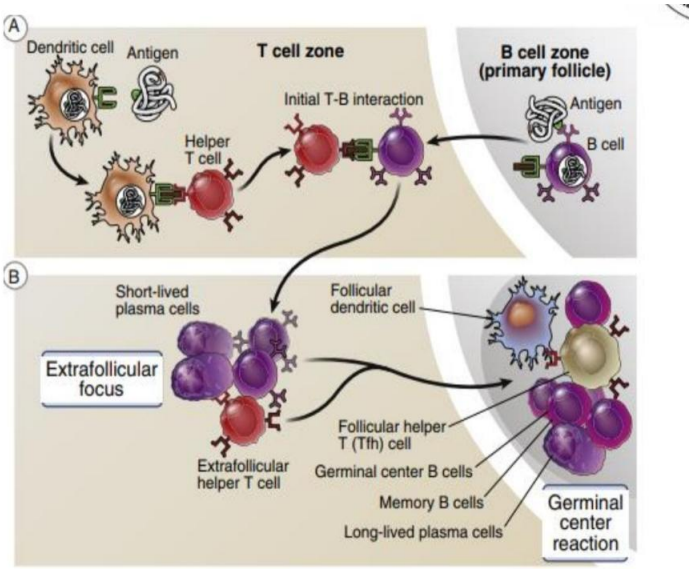
Activated T cells reduce expression of the chemokine receptor CCR7, which recognizes chemokines produced in T cell zones, and increase expression of the chemokine receptor CXCR5, which binds a chemokine produced in B cell follicles. Activated B cells undergo precisely the opposite changes, decreasing CXCR5 and increasing CCR7 expression.

– Activation of B cell by direct contact with T cell and by secreted cytokines from T helper.
– Activation of B cells by antigen results in increased expression of
• Class II major histocompatibility complex (MHC) molecules and B7 co-stimulators.
• Express the receptor CD40 which engage CD40 ligand (CD40L), on T cells (needed for isotype switch).
• Increase in cytokine receptors on activated B cells

B cells costimulatory receptors

• Costimulatory receptors provide so-called second signals for lymphocytes (antigen recognition provides the first signal) and ensure that immune responses are optimally triggered by infectious pathogens.
• B7-1 (CD80) and B7-2 (CD86), ligands on B cells and antigen presenting cells (APCs), they bind CD28 on T cells (activation), (signal 2)
• CD40 is a glycoprotein present on B cells and binds CD40L on T cells. Lead to B cell activation and isotype switch

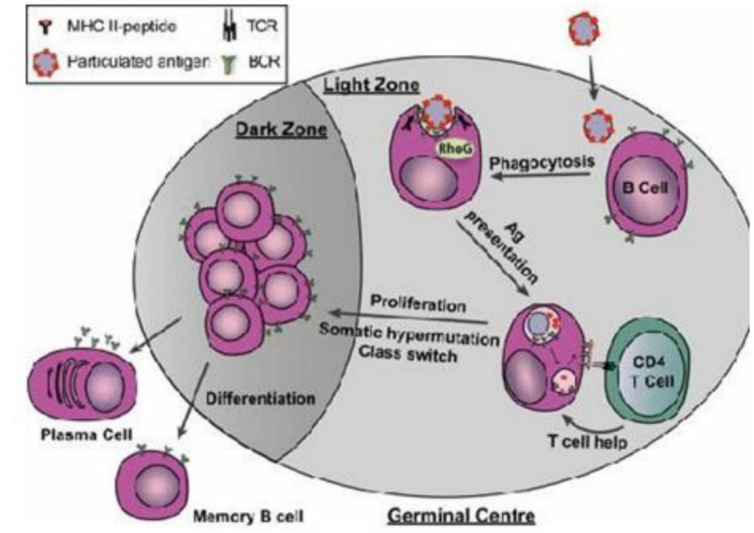




	TD Antigens	TI Antigens
Chemical nature	Proteins	Polymeric antigens, especially polysaccharides; also glycolipids, nucleic acids
Features of Response		
Primary B cell subset	Follicular B (B2) cells	MZ(/B1) B cells
Germinal center formation	Yes	No
Secondary isotypes (isotype switching)	Yes; IgG, IgE, and IgA	Little; some IgG and IgA
High affinity Ab's (affinity maturation)	Yes	No
Secondary response and memory B cells	Yes	Limited, only for some antigens
Long-lasting serum antibody titers (long-lived PCs)	Yes	No/limited

Results of B cell activation

1. Activated B cells migrate to germinal centers,
2. The proliferation of each B cell in response to one antigen result in one clone of cells with receptors of identical specificities.
3. B cells differentiate into antibody secreting plasma cells by switching membrane form Ig to secreting Ig,
4. Immunoglobulin isotype switching
5. Somatic hyper mutation of produced antibody

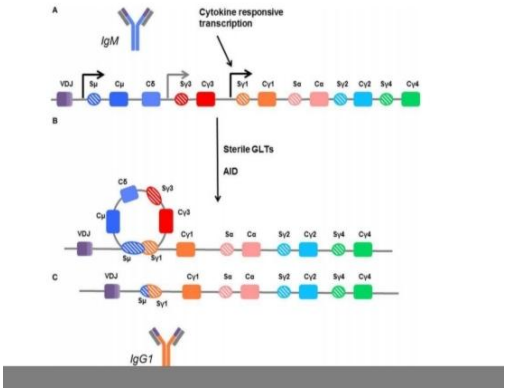


Isotype switch

• During B cell development in BM, naïve B cells are formed by combining Cμ or Cδ to the V-D-J of the heavy chains.
• Whereas in activated B cells in germinal center, isotype switch happen to other antibody isotypes, by combining other constant as Cγ for IGG, Cα for IGA and Cε for IGE.

• The molecular mechanism of isotype switching is a process called DNA recombination, in which B cells change the isotypes of the antibodies they produce by changing the constant regions of the heavy chains but the specificity (variable region) of the antibodies remains unaltered.
• The key enzyme required for isotype switching (and affinity

maturation) is activation induced deaminase (AID)
• CD40 signals work together with cytokines to induce isotype switching.
• Activation-induced cytidine deaminase (AID) plays a key role in both class switch and Somatic hyper mutation
• Deficiencies of AID underlie some forms of the hyper-IgM syndrome
• Mutations in the CD40L gene result in a disease called the X-linked hyper-IgM syndrome, which is characterized by defects in antibody production, isotype switching, affinity maturation, and memory B cell generation

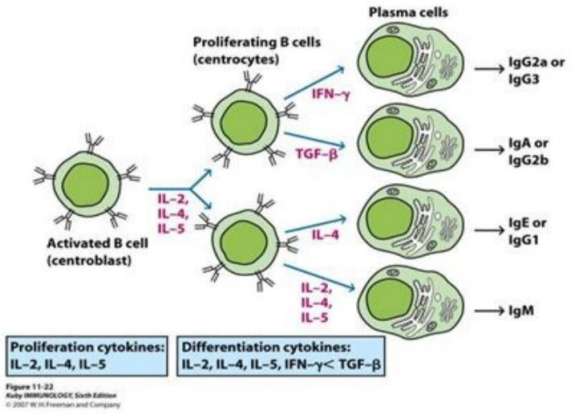


Isotype determinants

• Isotype switching in response to different types of microbes is regulated by cytokines produced by the helper T cells that are activated by these microbes.. Antibody response require
– protein antigens
– T-dependent B cell activation.

• Microbe type
– The response to many viruses and intracellular bacteria involves the production of IgG antibodies
– The humoral response to many helminthic parasites is mainly driven by IgE antibodies, which participate in eosinophil- and mast cell–mediated elimination of the Helminths– Allergen more IGE

• In addition, B cells in different anatomic sites switch to different isotypes. Specifically, B cells in mucosal tissues switch to IgA,
• a prior history of antigen exposure, first exposure more IGM, 2nd more IGG

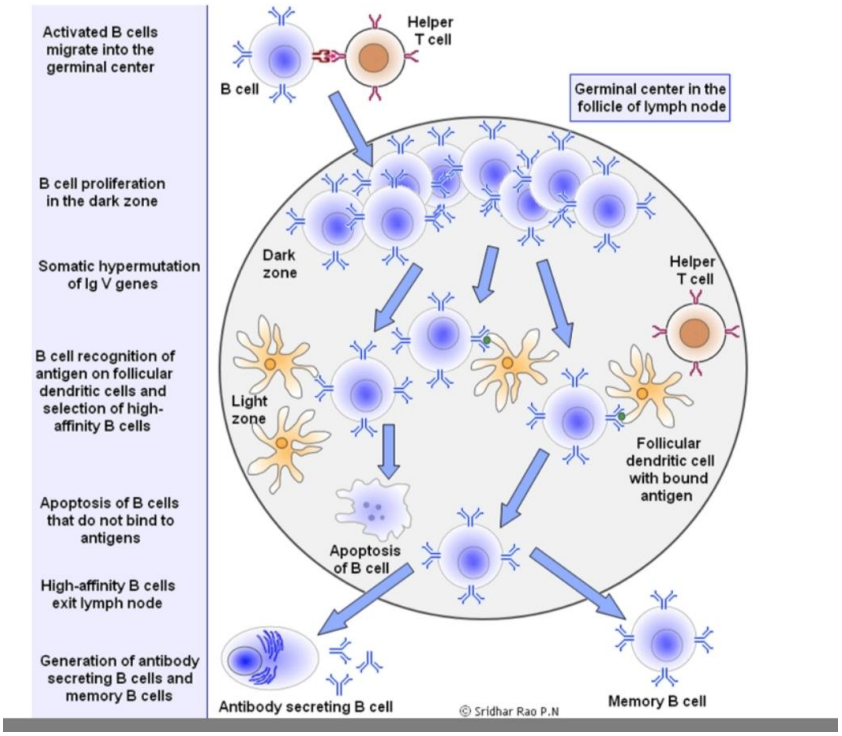


Somatic hypermutation

• Affinity maturation or somatic hyper mutation is the process that leads to increased affinity of antibodies, and it is the result of somatic mutation of Ig genes followed by selective survival of the B cells producing the antibodies with the highest affinities.

• In proliferating germinal center B cells in the dark zone, Ig V genes undergo point mutations at an extremely high rate to produce high affinity Ab. For this reason, mutation in Ig V genes is also called somatic hypermutation

• The antibodies produced have variable affinities (binding strength) to the antigen so that B cells producing high affinity Ab proliferate and become (antibody secretors) plasma cells and (non antibody secretors) memory B cells. While cells producing low affinity Ab die. This is called selection.



T cell Dependent B cell activation

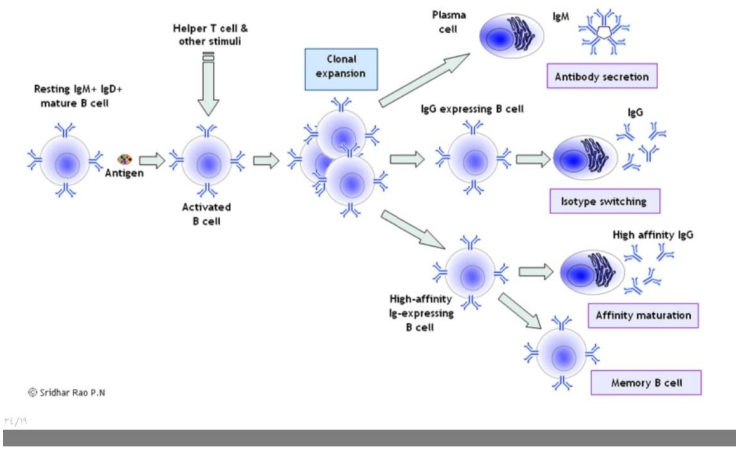


TABLE 11-2 Properties of thymus-dependent and thymus-independent antigens

Property	TD antigens	TI antigens	
		Type 1	Type 2
Chemical nature	Soluble protein	Bacterial cell-wall components (e.g., LPS)	Polymeric protein antigens; capsular polysaccharides
Humoral response			
Isotype switching	Yes	No	Limited
Affinity maturation	Yes	No	No
Immunologic memory	Yes	No	No
Polyclonal activation	No	Yes (high doses)	No

Plasma cells

• Short-lived plasma cells are generated during T-independent responses and early during T cell–dependent responses. These cells are generally found in secondary lymphoid organs and in peripheral non-lymphoid tissues.

• Long-lived plasma cells are generated in T-dependent germinal center responses to protein antigens.

– Signals from the B cell antigen receptor and IL-21 cooperate in the generation of plasma cells

– identified as antibody-secreting cells that do not express CD20, a marker of mature B cells.

– Some of (plasma cells) generated in germinal centers enter the circulation and home to the bone marrow where they differentiate into long-lived plasma cells. Some stay in medulla of secondary LN.

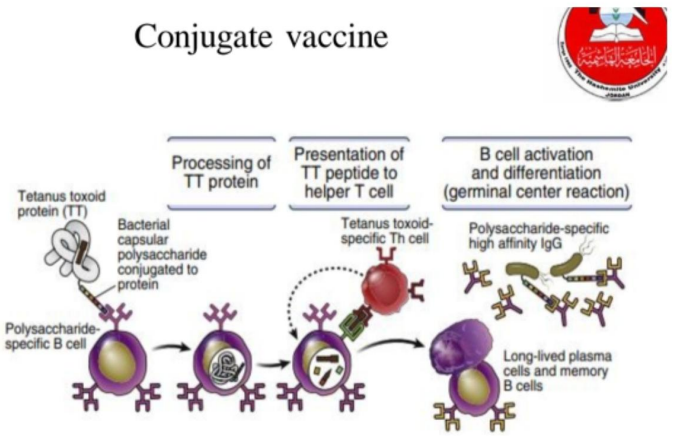
Humoral immunity shut down and formation of memory B cells

• After antibodies are capable of killing invading microorganisms, most of activated B cells die by programmed cell death

• Furthermore, circulating IgG antibodies that binds to antigen in periphery induce negative feedback mechanism to inhibit further antibody production

• Memory B cells are formed and stay for long time to facilitate faster antibodies production when the body is exposed to same antigen next time

the polysaccharide is covalently linked to a foreign protein to form the equivalent of a hapten-carrier conjugate, which does activate helper T cells. Such vaccines, which are called conjugate vaccines

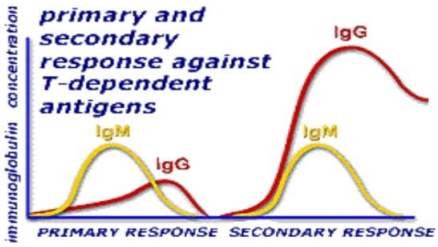


Primary and secondary immune response

• Primary and secondary antibody responses to protein antigens differ qualitatively and quantitatively.
• Primary responses result from the activation of previously unstimulated naive B and T cells, whereas secondary responses are due to the stimulation of expanded clones of memory B and T cells.
• Therefore, the secondary response develops more rapidly than does the primary response, and larger amounts of antibodies are produced in the secondary response. Heavy chain isotype switching and affinity maturation also increase with repeated exposure to protein antigens.
• Secondary immune response is mediated by memory cells

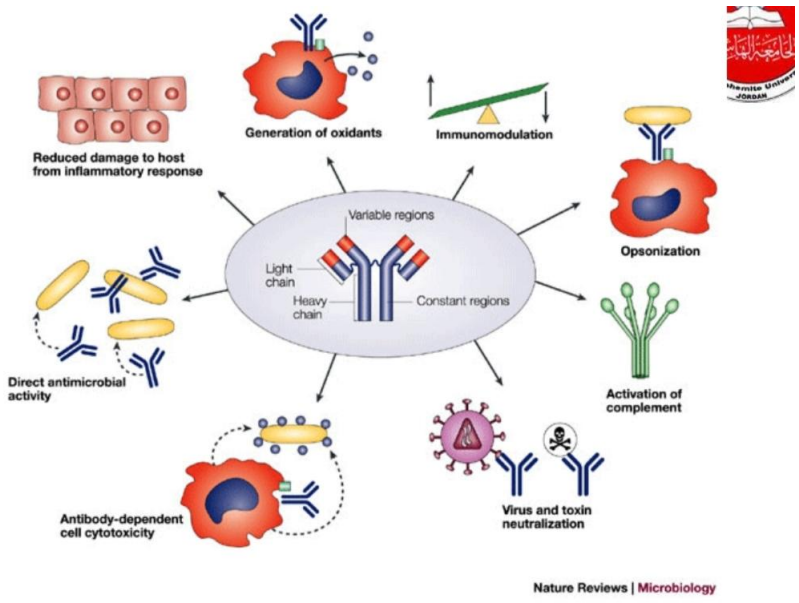
Difference Between Primary Response and Secondary Response.

	Primary Response	Secondary Response
Exposure to antigen	first exposure to a specific antigen	*after second exposure to the same* antigen
Time of onset	1-week delay	Within hours
Strength	weak potency	more potent
Duration	Short life , for only a few weeks	forms antibodies for many months
Type of antibody	IgM	IgG



Effector Mechanisms

•Neutralization
•Opsonization
•Complement activation
•Antibody dependent cell mediated toxicity (ADCC)
•Transcytosis- movement across epithelial cells



Done by:Dr.mohammed farhoud

In order to elicit an immune response, a carbohydrate must be:

A. Degraded.

B. Administered intranasally.

C. Made of polysaccharides.

D. Bound to a protein carrier.

E. Administered parenterally.

A 24-year-old patient recovers from a motor vehicle accident but requires a splenectomy. The doctor informs him that he can lead a normal life but will require which of the following?

A. Passive immunization with gamma globulin.

B. Regular transfusions.

C. Careful monitoring of lymph nodes for compensatory enlargement.

D. Immunization against encapsulated organisms.

E. Take TNF alpha inhibitors for life.

In order to generate an antigen-specific B-cell receptor (BCR) or TCR, the pre-B cell and pre-T cell are required to?

A. Encounter an antigen.

B. Rearrange their DNA for their receptors.

C. Splice their mRNA for their receptors.

D. Traffic to peripheral lymphoid organs.

E. More than one of the above.

Activation of B-cell receptor by binding of an epitope results in the formation of:

A. Plasma cells and cytotoxic T-cells.

B. Memory cells and cytotoxic T-cells.

C. Plasma cells for antibody production and memory cells for primary response.

D. Plasma cells for antibody production and memory cells for secondary response.

E. Plasma cells and helper T-cells.

Ans: D/D/B/C