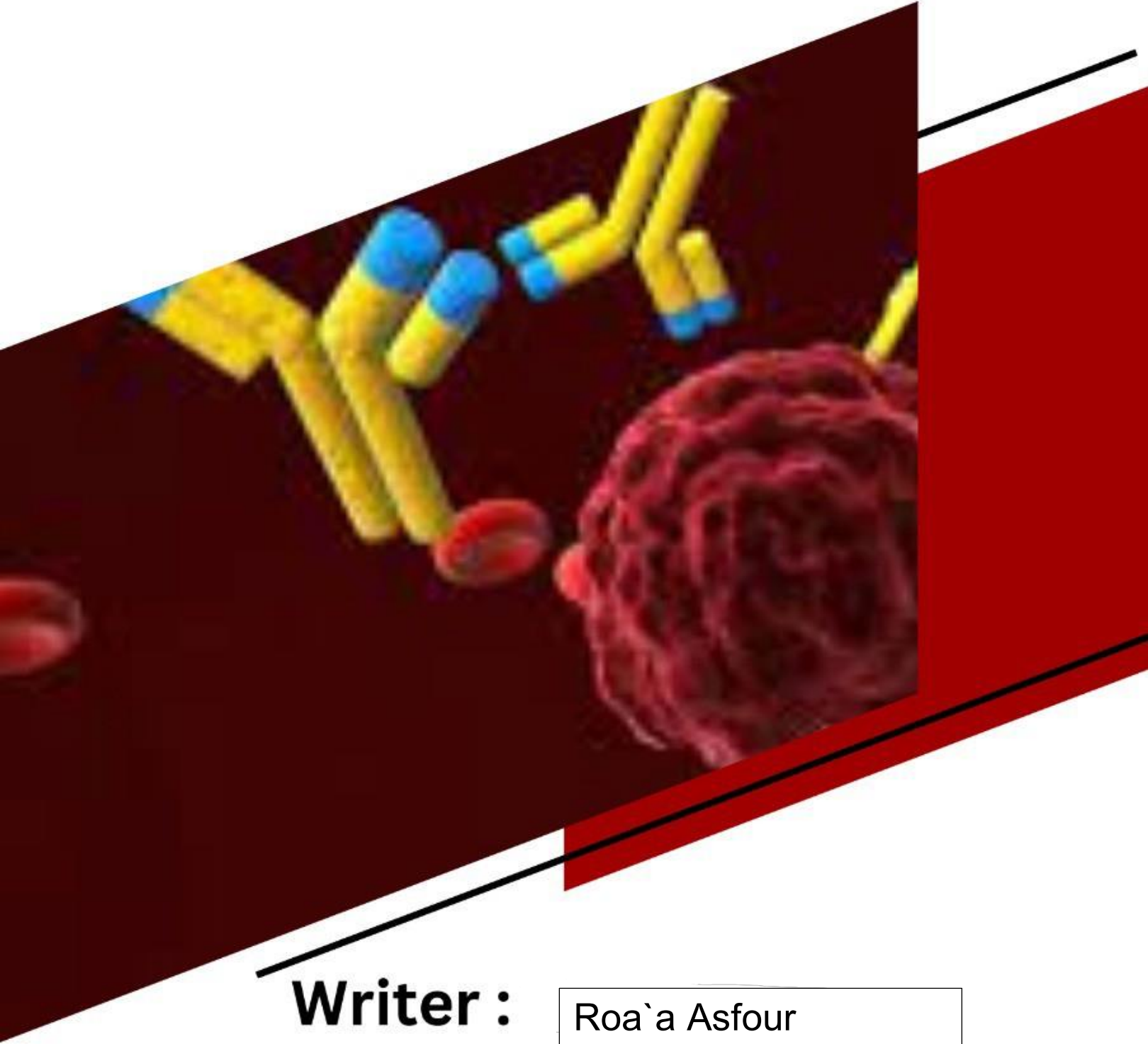


Doctor 021

IMMUNOLOGY

Sheet no.13



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Review

The topic of the last lecture was a bridge between innate and adaptive immunity. It is better to see innate and adaptive immunity as a continuum instead of separate entities. This is because these processes interplay with each other; cells of innate immunity interact with cells of adaptive immunity and vice versa.

- The word “lymphocytes” refers to B and T cells.
- B and T cells can recognize different antigens. T cell receptors are limited, they can only recognize linear peptides of a certain length that must be bound to the cleft of MHC molecules. This is why T cells exhibit MHC restriction. B cells, however, can recognize a wider range of antigens. They can recognize peptides, proteins, carbohydrates, lipids and metal ions.
- MHC II presents extracellular antigens to CD4 T cells, while MHC I presents intracellular antigens to CD8 cells. There is a difference in the processing of these molecules and the antigens they present.
- The antigen goes to the lymphocyte - The number of lymphocytes is limited, therefore, it would not be efficient for lymphocytes to go and look for microbes that enter from the multiple portals of entry (skin, GI tract, respiratory tract) or are blood-borne. Instead, antigen presenting cells (APCs) can be found at these sites, and they capture the antigens (this is the first step of antigen presentation to lymphocytes). Then the antigen can be brought to the T cell
- There are five APCs – All express MHC II. The most efficient type is dendritic cells, which can activate naïve T lymphocytes. Macrophages and B cells activate effector T lymphocytes. There are also thymic epithelial cells (important for lymphocyte development) and vascular epithelial cells
- Once the dendritic cell captures the antigen, it is no longer a resting/immature dendritic cell. The cells mature and start expressing co-stimulators and the homing receptor CCR7 (chemokine receptor 7). In T cell rich zones, CCL19 and CCL21 are secreted and they are ligands for the homing receptor. This will recruit dendritic cells to the T cell rich zone. During this process, the dendritic cells will complete their maturation and process the antigen.
- MHC I – Intracellular antigens such as viruses and mutated proteins of tumor cells They get tagged by ubiquitin, which sends them to the proteasome for degradation. At the same time, MHC synthesis occurs in the ER. The degraded peptide attaches to the TAP transporter, which will translocate the peptide into the ER. The TAP has a high affinity for tapasin, which will allow the TAP to bring the peptide with it into the ER. Then, the peptide is loaded onto the MHC molecule, passed through the Golgi apparatus, and finally sent through an exocytotic vesicle to the cell surface

MHC II – Extracellular Antigens – The antigen enters the cell in a membrane bound vesicle known as an endosome. This endosome becomes a phagosome, then it fuses with a lysosome to become a phagolysosome. This phagolysosome has a very low pH and enzymes such as cathepsins which cause degradation of proteins into peptides. MHC II synthesis occurs in the ER and immediately a protein, known as the invariant chain, binds to the cleft. This ensures that peptides that are meant to bind to MHC I won't accidentally bind to MHC II. The MHC II molecule is then sent to the phagolysosome and that's where loading will occur.

Antigen Recognition in the Adaptive Immune System and Lymphocytes Development

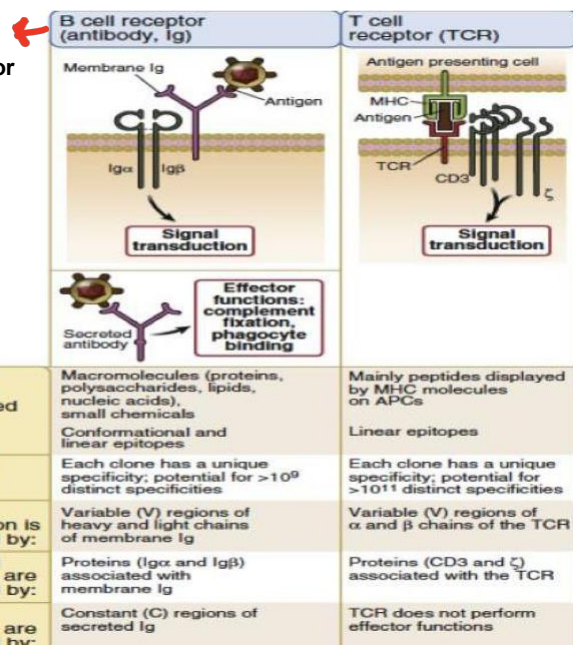
بسم الله الرحمن الرحيم
من هون رح نبدأ بالشيت

This lecture discusses adaptive immunity, so the focus is on B and T lymphocytes. Lymphocytes must have receptors so that they can recognize antigens. Although B and T cell receptors differ in many ways, they both have constant and variable regions and antigens bind to the variable region.

B and T cell receptors differ in many ways. This includes:

- 1. Form of Antigens Recognized –** As mentioned earlier, B cell receptors can recognize a larger variety of molecules, while T cells have MHC restriction.
- 2. Location –** B cell receptors may be membrane bound or in a secreted soluble form. T cell receptors are always membrane bound.
- 3. Effector Functions –** T cell receptors do not have any effector functions, while B cell receptors have effector functions done by the Fc portion of the antibody. Effector functions include complement fixation (activation) and phagocyte binding.

4 polypeptide chains (2 heavy and 2 light chains). This receptor can be bound or Soluble



2 polypeptide chains, alpha and beta . (Always membrane-bound)

The main difference between BCR and TCR

what they see/interact with. T cell receptors interact with processed proteins on MHC molecules (MHC restriction phenomena), while B cell receptors can interact with chemicals/ Carbs/lipids/nucleic acids/proteins.

TCR recognizes linear peptides (peptides that have NO 3D conformation and NO charge, and they have to be of a certain length). Unlike BCRs, they recognize antigens in their 3D STRUCTURE.

The diversity of both receptors is very huge. And this is due to the utilization of genetic recombination

Antibodies

Antibodies = B Cell Receptors (BCRs) = Immunoglobulins (Ig)

These are all different names for the same molecule!

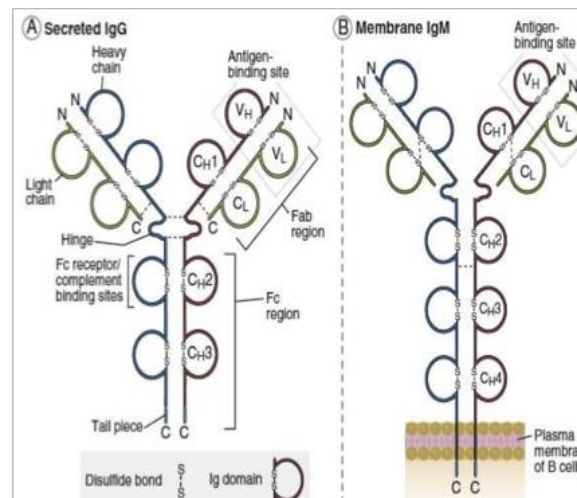
Structure

An antibody molecule is composed of four polypeptide chains—two identical heavy (H) chains and two identical light (L) chains—with each chain containing a variable region and a constant region.

- The light chain consists of 1 variable domain and 1 constant domain
- The heavy chain consists of 1 variable domain and 3 (when secreted) or 4 (when membrane-bound) constant domains.

Each light chain is attached to a heavy chain and the two heavy chains are attached to each other by disulfide bonds.

Each variable domain also has hypervariable regions (also known as complementarity-determining regions {CDRs}). This is what gives the fine specificity of the receptor. The fine specificity of the antigen comes from the epitope (also known as the determinant). This basically means that these are the components that allow for the highly specific binding between an antibody and its antigen.



Parts of the Antibody:

1. Fab (fragment of antigen binding) region - The part of the antibody that binds to the antigen. Each antibody has two, identical Fab regions.

a. The Fab region is composed of the variable and constant domain of the light chain and the variable and first constant domain of the heavy chain.

b. As a monomer, an antibody can bind to 2 antigens for the two Fab regions (or two epitopes of the same antigen). However, antibodies can also exist as dimers, trimers, or pentamers. Therefore, they can be bound to up to 10 epitopes, for the 10 Fab regions present in an antibody that exists as a pentamer.

2. Fc (fragment that crystallizes) region - Responsible for most of the biological activity and effector functions of the antibodies.

3. Hinge region – It is a flexible region between the Fab and Fc parts. It allows the two antigen-binding Fab regions of each antibody to move independently of each other. This allows them to bind to different epitopes separately.

in the heavy chain, the constant domain can be longer or shorter depending on the state of the BCR, if its membrane bound or soluble.

**The different forms of the Fab region are called IDIOTYPES .
The different forms of the heavy chain are called ISOTYPES .**

• Examples on enzymes that can cleave antibodies are pepsin and papain, they cleave in different ways

Isotypes

There are five types of heavy chains, called μ , δ , γ , ϵ , and α , which differ in their constant regions. Antibodies that contain different heavy chains belong to different classes, or isotypes, and are named according to their heavy chains (IgM, IgD, IgG, IgE, and IgA).

The antigen receptors of naive B lymphocytes, which are mature B cells that have not encountered any antigens, are membrane-bound IgM and IgD. After stimulation by Antigen and helper T lymphocytes, the antigen-specific B lymphocyte clone may expand and

These same B cells may produce antibodies of other heavy-chain classes. This change in Ig isotype production is called heavy-chain class (or isotype) switching. For example, a class switch may be from an IgM which recognizes antigen X to an IgG which also recognizes antigen X. The heavy chains (more specifically, the Fc portion) can change during the B cell life span, but the light chain (which can be of κ -kappa or λ -lambda types) doesn't.

The next table illustrates the different isotypes. While most of what the professor stated is in the table, there are a few additional points mentioned:

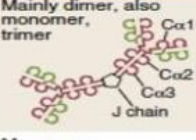


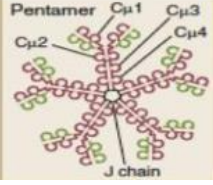
- Out of the four IgG subclasses, IgG1 and IgG3 are the most common in humans. IgG antibodies have multiple functions, including complement fixation (activation), opsonization, and neonatal immunity (immunity transferred from the mother to the fetus since IgG antibodies are the only antibodies that can pass through the placenta)
- IgM in its soluble form is a pentamer. Therefore, it can bind up to 10 different antigens or 10 different epitopes of the same antigen.

(IgD is concerned with RNA splicing that is happening to the RNA encoding the constant domain of the heavy chain).

Immature B lymphocytes express IgM only

Mature NAÏVE B lymphocytes express IgM and IgD

The same B cells may produce antibodies of other heavy-chain classes. This change in Ig isotype production is called heavy-chain class (or isotype) switching. (This doesn't change specificity but changes the function (effector mechanism))

Isotype of antibody	Subtypes (H chain)	Serum concentration (mg/ml)	Serum half-life (days)	Secreted form	Functions
IgA	IgA1,2 ($\alpha 1$ or $\alpha 2$)	3.5	6	Mainly dimer, also monomer, trimer 	Mucosal immunity
IgD	None (δ)	Trace	3	Monomer	Naive B cell antigen receptor
IgE	None (ϵ)	0.05	2	Monomer 	Defense against helminthic parasites, immediate hypersensitivity
IgG	IgG1-4 ($\gamma 1$, $\gamma 2$, $\gamma 3$ or $\gamma 4$)	13.5	23	Monomer 	Opsonization, complement activation, antibody-mediated cytotoxicity, neonatal immunity, feedback inhibition of B cells
IgM	None (μ)	1.5	5	Pentamer 	Naive B cell antigen receptor (monomeric form), complement activation

Binding

The parts of antigens that are recognized by antibodies are called epitopes (or determinants).

The strength with which one antibody binds to one epitope of an antigen is called the affinity of the interaction. Affinity can mature (affinity maturation).

The total strength of binding is much greater than the affinity of a single antigen-antibody bond and is called the avidity of the interaction. This considers the binding of all the units of the antibody (so it takes into account whether the antibody is monomeric, dimeric, etc.)

Antibodies produced against one antigen may bind to other, structurally similar antigens. Such binding to similar epitopes is called a cross-reaction.

Cross-reaction measures the extent to which different antigens appear similar to the immune system

Cross-reactivity between antigens occurs when an antibody raised against one specific antigen has a competing high affinity toward a different antigen)

These similar antigens can be self-antigens sometimes.

It's called also "molecular mimicry ":(the Ag of the microbe is similar to self antigens)

Molecular mimicry is a hallmark of the pathogenesis of rheumatic fever where the streptococcal group A carbohydrate epitope, structurally mimics cardiac myosin in the human disease, rheumatic carditis (rheumatic).

○ Monoclonal Antibodies

The realization that one clone of B cells makes an antibody of only one specificity (recognizes one antigen) has been exploited to produce monoclonal antibodies. They have therapeutic and research uses.

Making Monoclonal Antibodies

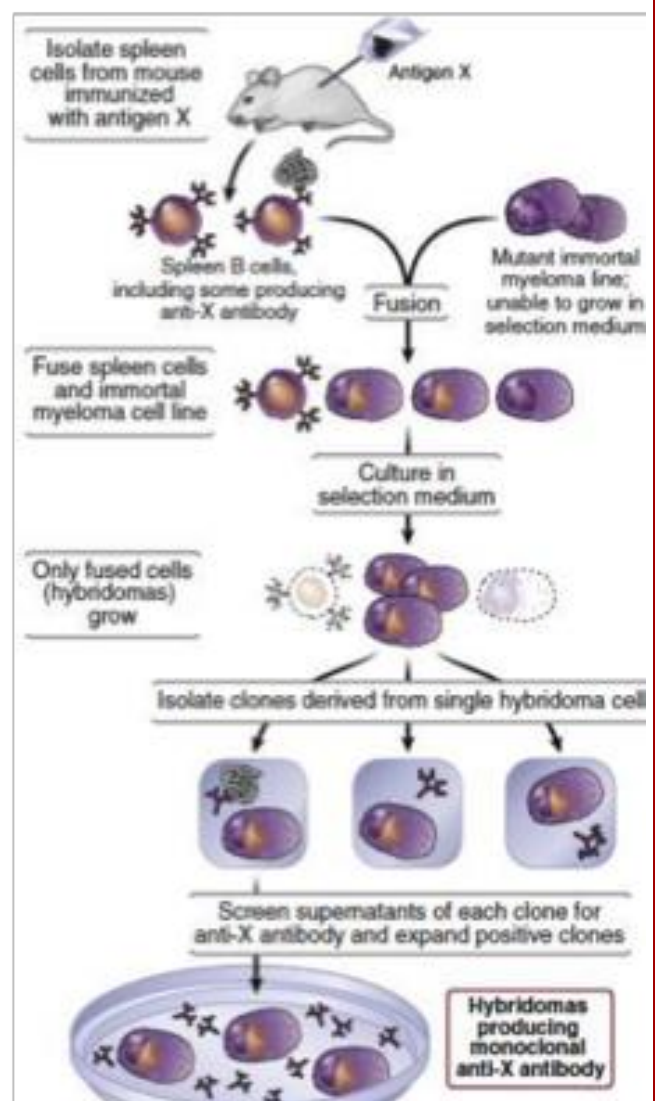
1. Antigen X is injected into a mouse. Then, the B cell population of the spleen is taken, as the spleen has clones of the B cells that produce antibodies that are specific to antigen X.

2. B cells have a short life span in vitro. Therefore, they are fused with plasma myeloma cells, which are immortal, tumor cells. This fusion results in a hybridoma cell.

a. The plasma myeloma cell must be unable to produce its own antibodies. This is because we only want the antibodies produced by the B cell clones of the mouse.

3. Selection and expansion are done on the clones that produce antibodies against antigen X.

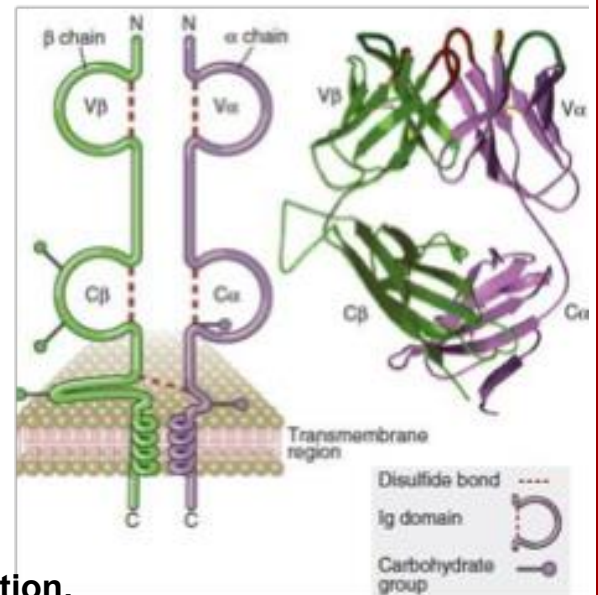
The issue with using these antibodies for medical purposes is that they originate from a mouse, which the immune system will recognize as foreign. Recently, monoclonal antibodies have been generated by using recombinant DNA technology to clone the DNA encoding human antibodies of the desired specificity.



T Cell Receptors

The antigen receptor of MHC-restricted CD4+ helper T cells and CD8+ cytotoxic T lymphocytes (CTLs) is a heterodimer consisting of two transmembrane polypeptide chains, designated TCR α and β , covalently linked to each other by a disulfide bridge between extracellular cysteine residues.

Each chain is made up of a variable and constant domain. The variable domains of the α and β chains form the antigen binding site of T cell receptors. Of course, there are also hypervariable regions within the variable region. The chain also has a transmembrane sequence.



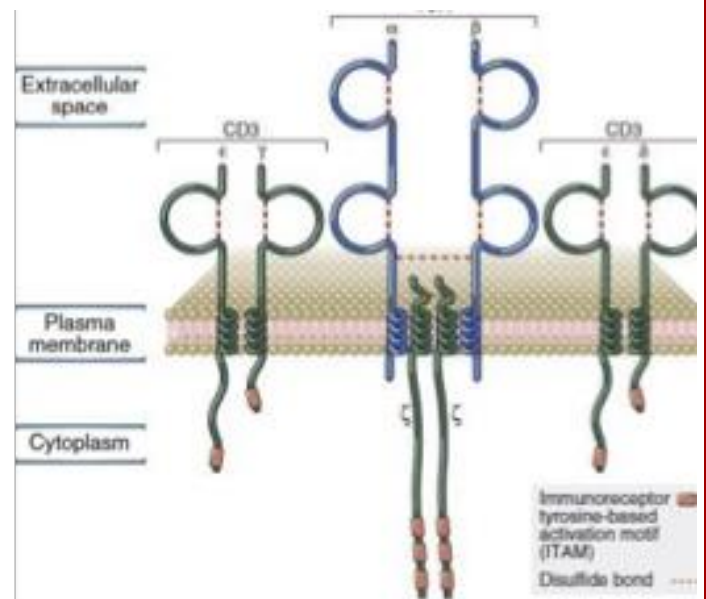
TCRs do not exhibit class switching or affinity maturation.

While most T cells receptors are $\alpha\beta$, there is a small population of $\gamma\delta$ (gamma delta).

The T cell receptor complex is a complex of the TCR and associated signaling molecules.

These signaling molecules help in signal transduction after the antigen binds. For T cells these signaling molecules are CD3 and zeta (ζ).

- Allelic exclusion occurs in both T+B cell receptors.
- In B cell receptor light chain(kappa and lambda)



Antibodies also have signaling molecules, Ig α and Ig β . So, the B cell receptor along with these signaling molecules form the B cell receptor complex.

Each gene segment encodes for alpha/ beta chain if we are talking about T cell receptor, heavy/ light chain if we are talking about B cell receptor, you have 2 copies on each chromosome one from your mother and the other from your father, so if the encodes the cope from your father and its functional and good then the other copy from your mother will be excluded(shut off) this is called allelic exclusion. So only one specific antigen binding site would result

The Immune Repertoire

As mentioned earlier, the CDRs give the fine specificity of the receptor. But why do they need this high specificity?

As adaptive immunity is very specific, the antigen receptors of lymphocytes must be able to bind to and distinguish between many, often closely related, chemical structures. So each lymphocyte clone must have a unique receptor that can differentiate between distinct antigens. The process of lymphocyte maturation first generates a very large number of cells (perhaps as many as 10^9) before any encounter with antigens. This large collection of distinct lymphocytes makes up the immune repertoire. However, not all these cells have useful receptors. The selection process now comes into play and promotes the survival of cells with receptors that can recognize antigens. Cells that cannot recognize antigens or have the potential to cause harm will be eliminated.

Remember that this specificity is for the sake of antigen binding. Therefore, although each clone of T lymphocytes recognizes a different antigen, the antigen receptors transmit biochemical signals that are fundamentally the same in all lymphocytes and are unrelated to specificity.

Lymphocyte Development

DEVELOPMENT OF IMMUNE REPERTOIRES

As the clonal selection hypothesis predicted, there are many clones of lymphocytes with distinct specificities, perhaps as many as 10^9 , and these clones arise before an encounter with antigen.

- The process of lymphocyte maturation first generates a very large number of cells each with a different antigen receptor and then preserves the cells with useful receptors.
- receptors are expressed on developing lymphocytes, and selection processes come into play that promotes the survival of cells with receptors that can recognize antigens, such as microbial antigens, and eliminate cells that cannot recognize antigens in the individual or that have the potential to cause harm.

Lymphocyte Development

If we think about the receptors we need for all the antigens, microbes, fungi, and cancers we would need all the earth's DNA to make a separate antigen binding site for every segment on T/B cell receptors .

site of maturation for

Bcells=>bone

marrow

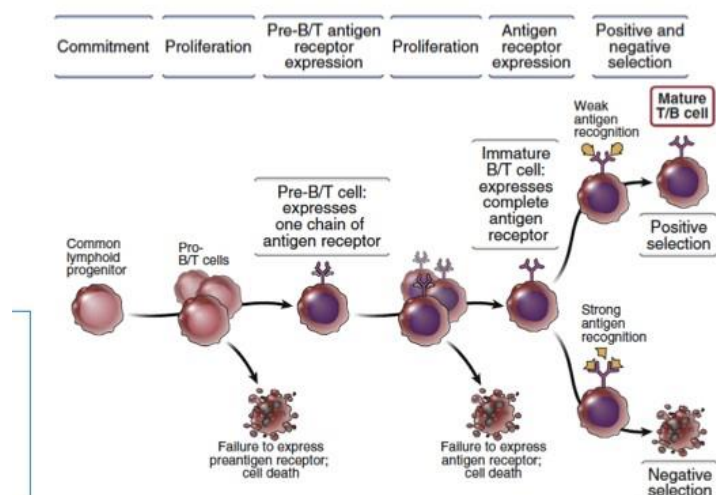
• Tcells=>leave bone

marrow and mature

in thymus.

B cells also mature at the spleen but we

Wil not focus on this.



These lymphocytes possess a molecular mechanism and the centers of these mechanisms are called antigen receptor gene rearrangements, this has a critical role in the development of lymphocytes and generation of the diversity of T/B cell receptors so it is diverse and at the same time specific towards unknown antigens once they are encountered.

• A Basic Idea:

The first step in lymphocyte development is the commitment of common lymphoid progenitor cells to either B cell lineages or T cell lineages. Then, the cells go through cycles of proliferation and antigen

receptor gene rearrangement

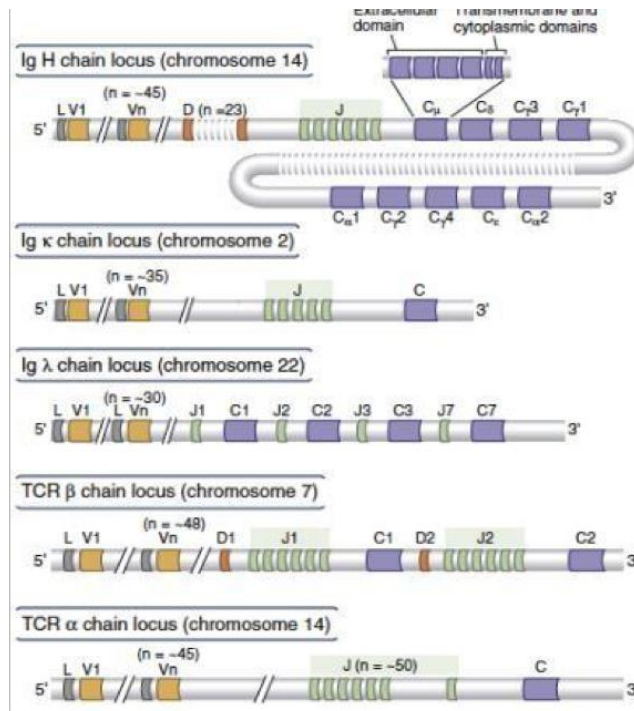
(this is a very important step!).

Finally, either positive or negative selection occurs •

• Production of Diverse Antigen Receptors •

formation of functional genes that encode B and T lymphocyte antigen receptors is initiated by somatic recombination of gene segments that code for the variable regions of the receptors, and diversity is generated during this process.

- Early lymphoid progenitors contain Ig and TCR genes in their inherited(encoded), or germline, configuration. In this configuration, Ig heavy-chain and light-chain loci and the TCR α -chain and β -chain loci each contain multiple variable region (V) gene segments, numbering about 30 - 45 and one or a few constant region (C) genes.
- Between the V and C genes are groups of several short coding sequences called diversity (D) and joining (J) gene segments. (All antigen receptor gene loci contain V, J, and C genes, but only the Ig heavy-chain and TCR β chain loci also contain D gene segments.)
- There are 2 D segments in beta chains and about 23 D segments in the heavy chain of antibodies.
- There are pluripotent stem cells in the bone marrow then we have cells that are committed to becoming B/T lymphocytes(called commitment), the common lymphoid progenitor cell will either continue in the bone marrow and become B lymphocytes or will go to the thymus and be T lymphocytes, then, there will be the progenitor for B or T cells in this stage antigen receptor gene rearrangement occurs (recombination), they are segments that encode for B/ T cell receptor ,they are germ-line encoded(inherited) so they are present but what happens is that the segments are relocated, one segment from your father and the next one from your mother and then from your mother and so on...
-it will take the V region(variable region) and J region(joining region)(these are short coding segments) and then it binds them together by a constant region, this is called combinatorial diversity, enzymes do this called RAG enzymes(VDJ recombinase, RAG1,2) and these are in the bone marrow and the thymus, this will give so many possibilities.



Add to this the work done by an enzyme called TDT (terminal deoxynucleotidyl transferase), what it does is that it adds/ deletes nucleotides from the DNA that resulted from the RAG enzyme (after relocating the segments) and this is called junctional diversity.

• Germline organization of antigen receptor gene loci •

So the idea is that antigen receptor gene rearrangement recombination occurs at this stage.

- Then there is the pre stage ,in this case, the receptor is completed, if we are talking about B cell receptor so it's a heavy chain then the light chain is completed too.
- Starts with the heavy chain if we are talking about B cell receptor and with the beta chain if we are talking about the T cell receptor,
- then proliferation occurs if the result is a functional intact expression of the gene this is a survival signal, if not intact this gives an apoptotic signal.
- If we are talking about the T cell receptor Beta chain then the alpha chain is completed too.
- After that, positive/negative selection occurs, if the receptor interacts very strongly with a self-peptide in the bone marrow or the thymus then does the negative selection(deletion)
- Primary lymphoid organs: bone marrow + thymus
- Low affinity/avidity does the positive selection, in both cases, we are talking about SELF Ag because they are primary lymphoid organs(sterile= no antigens)

• Recombination and expression of immunoglobulin (Ig) genes. •

All this lymphocyte development happens before lymphocytes face any antigens.

• So by reacting with these self-antigens if the reaction was very strong then it's possible that it's a self-reactive B/T lymphocytes(it will undergo apoptosis) .

• Low affinity/avidity will continue and undergo positive selection, so I have commitment and between critical stages there is proliferation steps through the signal of the previous step survival signal or lastly by positive/negative selection.

• The lymphocyte is committed to become either T or B cell, it either becomes pro-B cell and goes to the bone marrow or becomes proT cell and goes to the thymus.

• In recombination step in pro there is combining of gene segments, so we have the heavy chain on B cell receptor or the beta chain of T cell receptor.

• In pre we continue with the light chain of B cell receptor or the alpha chain of T cell receptor.

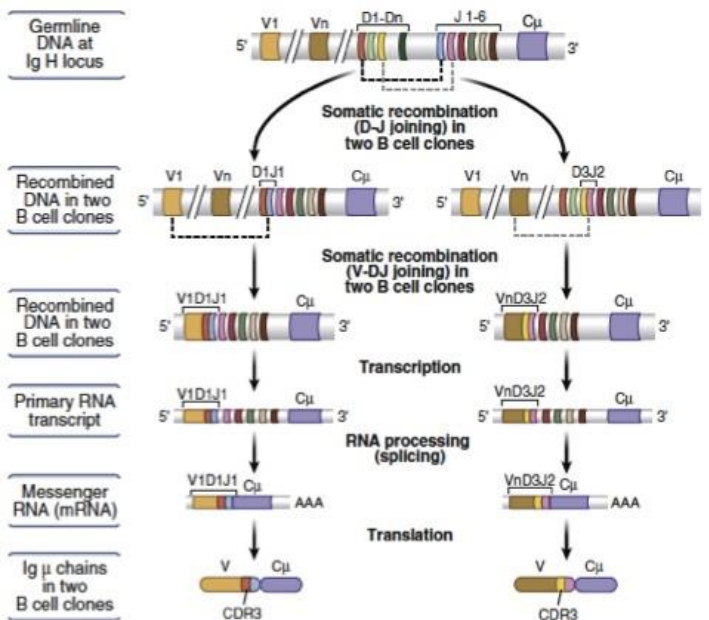
• Keep in mind: There is cytoplasmic expression before the surface expression.

• A frequently asked exam question: 😊

"what do immature/ mature B cells have on them?" Or "a group of cell have the following expression receptors are they mature? immature? Pro-B cell?"

Immature B cells have IgM

**Mature B cells have IgM+IgD



Mechanisms of V(D)J Recombination and Generation of Ig and TCR

Divers:

The somatic recombination of V and J, or of V, D, and J, gene segments is mediated by a lymphoid-specific enzyme, the VDJ recombinase (RAG-1 and RAG-2) proteins, and additional enzymes, most of which are not lymphocyte specific and are involved in repair of double-stranded DNA breaks introduced by the recombinase.

Diversity of antigen receptors is produced using different combinations of V, D, and J gene segments in different clones of lymphocytes (called combinatorial diversity) and even more by changes in nucleotide sequences introduced at the junctions of the recombining V, D, and J gene segments (called junctional diversity)

V= variable, C=constant

D= diverse, J= joining

What we mean by diversity is all possible outcomes of B and T cell receptors and it's the result of a molecular mechanism called Antigen receptor gene rearrangement during lymphocyte development.

- The gene segments that encode for the variable region (the domain/constant region, alpha chain, beta chain, gamma chain) are present on a chromosome.

-Immunoglobulin heavy chain is on chromosome 14

- K and V chains can be on chromosome 2 and 22

- Beta chain can be on chromosome 7

-Alpha chain is on chromosome 14

- Each one of these gene segments encodes for either heavy chain or light chain,, either alpha chain or beta chain

, they're constant domains and their number is limited also they have variable domains/ segments(D1/D2/D3)

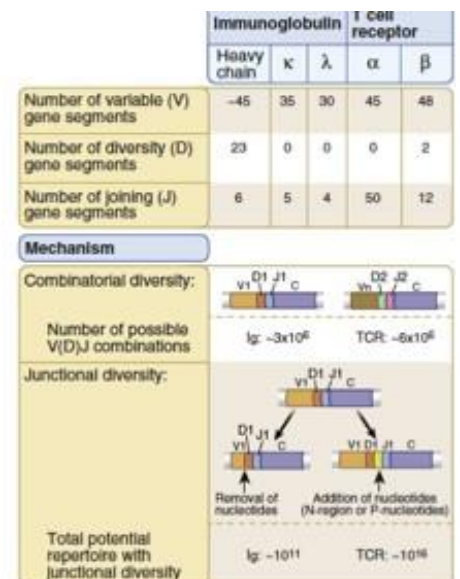
It depends on B or T cell

- They also have diverse regions(abbreviated short coding segments) ONLY presented on the immunoglobulin heavy chain and T receptor beta chain (very imp to know).

Between one segment and the other, there are short coding genes called Join (J).

The genes that encode for the variable domains range from 30-45 gene segments. This does give many different possibilities and diversity, because there are many different possible combinations of the V,D, and J segments. However, this diversity is still less than that given by junctional diversity (because you can add or remove as many nucleotides as possible).

Notice how junctional diversity gives a much larger potential repertoire



Combinatorial diversity: in lymphocyte mother cells and their offspring **RAG** enzyme^{1,2}(recombinase) re-organizes the segments, one segment from your mother, the other from your father, and the next one from your father... and so on, so the result is a totally new and different DNA.

-This occurs randomly in the cell.

-At the same time, between one segment (either variable, diverse, joining, or constant) there's **TDT** (terminal deoxynucleotidyl transferase) that adds nucleotides without the need for a template, there's also exonuclease that removes nucleotides (it can add 10 and then remove 3, add 2 and remove 7...) this is called **junctional diversity**.

•So imagine the possibilities! Reorganizing segments gives us combinatorial diversity, and the pinpoints that are done by the TDT give us junctional diversity.

-So the repertoire of this B/T cell receptor to cover the cognate antigen is very properly

- -if we add/remove nucleotides without templates doesn't this result in mutations?

: there are DNA repair mechanisms working in lymphocytes, but they work randomly if there is a mutation.

- for example, a known mutation in the RAG enzyme results in severe combined immunodeficiency syndrome (lack of B as well as T cells).

-If there's a partial activity it's called Omenn syndrome.

• mRNA can undergo splicing which is an important step to form IgD on B cell receptors.

• Receptor gene rearrangement (recombining) starts from the middle of the diverse region (so if we were talking about the heavy chain in an antibody or beta chain in T cell receptor it starts from the D segment(diverse segment)that's present only in Beta in T and Heavy chain in B then it gets the variable gene then the joining(at the level of DNA) and finally the constant region will be added at the level of mRNA, post transcription modification) This is basically what antigen receptor gene rearrangement is.

• Remember: in the development steps between each step and the other there is proliferation things don't go smoothly always for example if something is not intact happened and not functional then this will signal for apoptosis, if it is normal, it will give survival signal(it will proliferate).

• 95-99% of T cells that go to the thymus will die, after positive/ negative selection it will give non-self reactive T cell and still this is not enough there still might be self reactive T cells against self antigens, so there is something called peripheral tolerance mechanism will work on periphery after activation.

- B cell receptors will undergo somatic hypermutation and affinity maturation, this happens after encountering the antigen (these are the steps during lymphocyte development until they are naïve mature (and haven't been activated by an antigen yet))

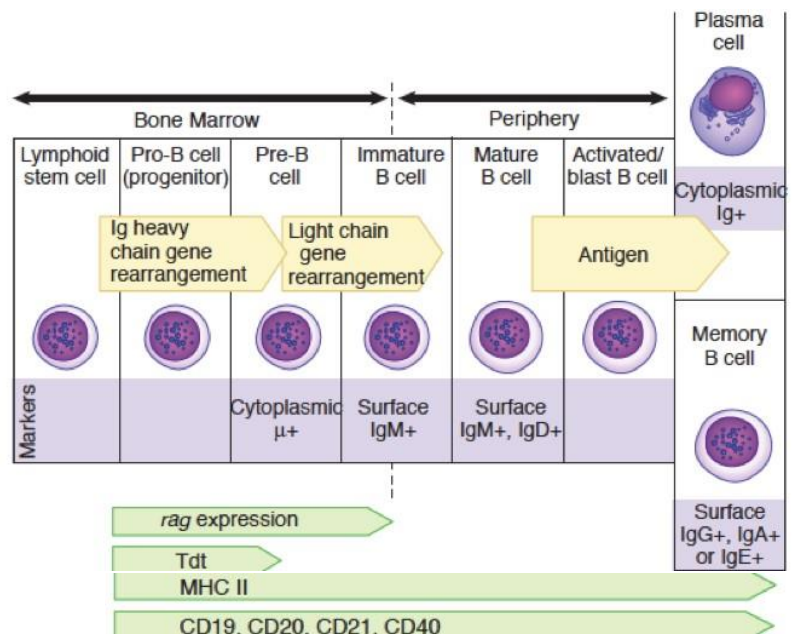
*Maturation and Selection of B Lymphocytes:

-B cell maturation occurs mostly in the bone marrow, but some maturation may also occur in the spleen.

1. Bone marrow progenitors committed to the B cell lineage proliferate, giving rise to many precursors of B cells, called pro-B cells. (Commitment = pro-B cells). Pro-B cells do not express any receptors.
2. Then, it starts the expression of genes for BCRs. The Ig heavy-chain locus rearranges first, and only cells that are able to make an Ig μ (IgM) heavy-chain protein are selected to survive and become pre-B cells. The assembled pre-BCR serves essential functions in the maturation of B cells. The IgM-expressing B lymphocyte is the immature B cell.
3. The mature B cell expresses both IgM and IgD. How does this occur? The mRNA for the heavy chain gets spliced. The result of this splicing is that the mRNA once transcribed gives IgM and IgD. This cell is now able to respond to antigens in peripheral lymphoid tissues.
4. Now selection must occur. Positive selection occurs for B cells that express intact, functional BCRs. So developing B cells are positively selected, based mainly on the expression of complete antigen receptors and not on the recognition specificity of these cells. Negative selection occurs for B cells that react strongly with self-antigens.

Cells committed to becoming B cells (pro-B cell) so this common lymphoid progenitor will transform into pro cell and will start antigen receptor gene rearrangement, we're talking about B cell receptor so we will start with the heavy chain

- If we were talking about T cells, then we start with the beta chain



In the pre-B cell, there is a cytoplasmic new chain (cytoplasmic expression, translation + transcription occurred) through positive/negative selection will either become a memory lymphocyte or effector lymphocyte

Effective=> IgM,, Memory=> not IgM

- *Cell surface IgM is immature B cell*

- *IgM, IgD=> mature B cell*

-RAG enzyme starts at the pro stage (where antigen receptor gene rearrangement occurs and continues until the **Immature** stage)

-the surface markers :

- CD3=> T CELLS

- CD19, CD20, CD40,CD21=> B cells

- MHC class 2(APC) starts and stays with the cell until it dies

Maturation and selection of T lymphocyte

-T cell progenitors migrate from the bone marrow to the thymus, where the entire process of maturation occurs.

- -Double negative means they have neither CD4 nor CD8 on them.

- The least developed progenitors in the thymus are called pro-T cells or double-negative T cells(or double-negative thymocytes) because they do not express CD4 or CD8.

- TCR β gene recombination, mediated by the VDJ recombinase, occurs in some of These double-negative cells.

- If VDJ recombination is successful in one of the two inherited loci and a TCR β chain protein is synthesized, it is expressed on the cell surface in association with an invariant protein called pre-T α , to form the pre-TCR complex of pre-T cells.

- If the recombination in one of the two inherited loci is not successful, recombination will take place in the other locus. If that fails too and a complete TCR β chain is not produced in a pro-T cell, the cell dies.

-When it comes out of the bone marrow and it's a committed lymphocyte to become a T cell they will become Pro-T cells , they get to the cortex of the thymus and we call them double negative T cells

The pre-TCR complex delivers intracellular signals once it is assembled, similar to the signals from the pre-BCR complex in developing B cells.

- These signals promote survival, proliferation, and TCR α gene recombination and inhibit VDJ recombination at the second TCR β -chain locus (allelic exclusion).
- Failure to express the α chain and the complete TCR again results in death of the cell.
- The surviving cells express the complete $\alpha\beta$ TCR and both the CD4 and CD8 coreceptors; these cells are called double-positive T cells (or double positive thymocytes).

At the same time, at the cortex of the thymus starts antigen receptor gene rearrangement and it starts at the beta chain, when they reach the medulla they are mature

•

They were double negative and then they become double positive (CD4+CD8 positive) then they will become single positive and after that either positive selection or negative selection, the cell that has CD4 interacts with MHC class 2 keeps CD4 and CD8 is gone

In double positive:

No encountering with an antigen=> inactive

Keep in mind they haven't encounter antigens yet primary lymphoid organs are sterile (they learn from self Ag)

If a cell with CD8 interacts with MHC class 1 then CD4 will be gone (will become single positive)

Selection of Mature T Cells

for a single positive cell high avidity (affinity) => negative selection

Low or medium avidity (affinity) => positive selection

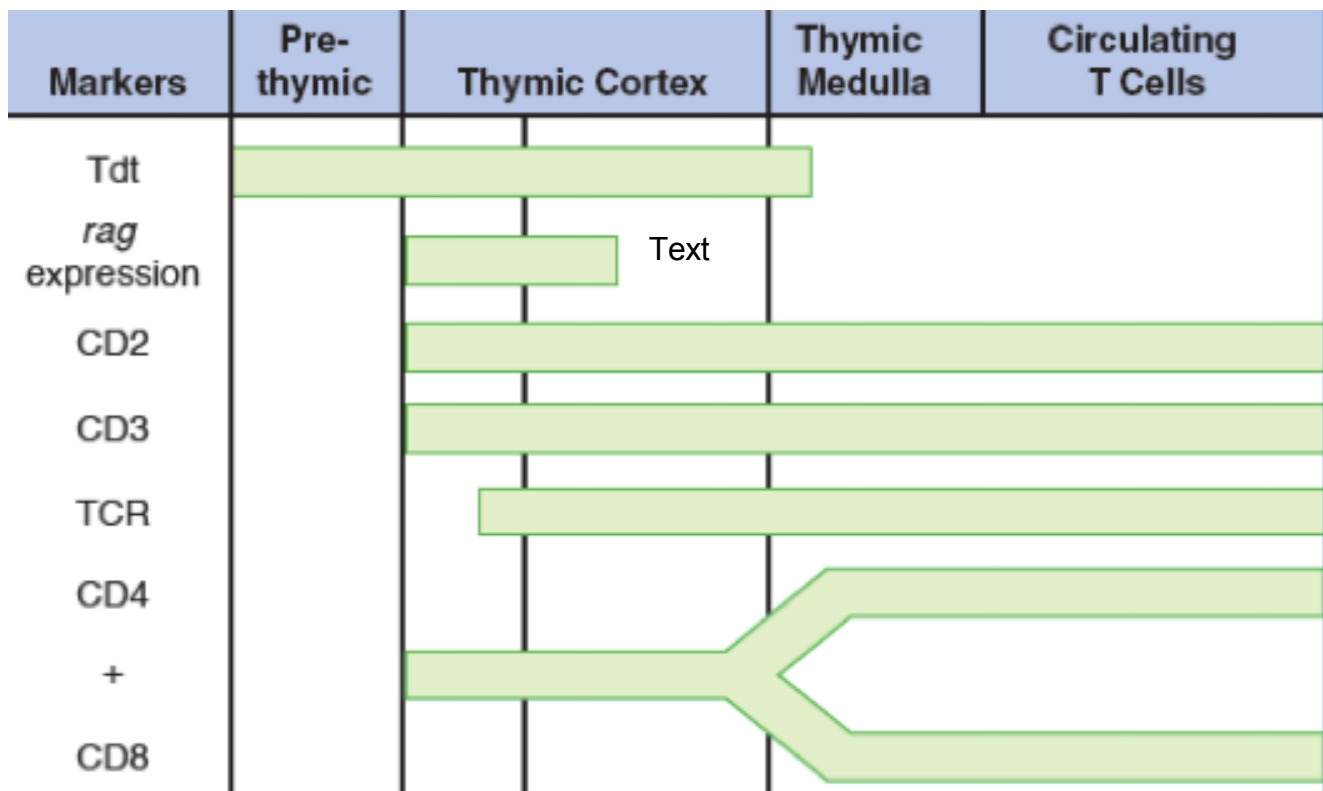
Let's compare:

the TDT starts at the heavy chain (receptor gene rearrangement) but stops at the beginning of the light chain Here it continues till the end of selection

- If the TCR of a T cell recognizes an MHC molecule in the thymus, which must be a self MHC molecule displaying a self peptide, and if the interaction is of low or moderate affinity, this T cell is selected to survive (positive selection).
- During this process, T cells whose TCRs recognize class I MHC–peptide complexes preserve the expression of CD8, the coreceptor that binds to class I MHC, and lose expression of CD4, the coreceptor specific for class II MHC molecules and the other way around. single-positive T cells.
- Immature, double-positive T cells whose receptors strongly recognize MHC-peptide complexes in the thymus undergo apoptosis. This is the process of negative selection.

-At the same time, RAG expression (Combinatorial diversity) will define in the cortex so they will go from PRO to PRE T cells

CD2+CD3 are surface markers for T cells.



They were double negative then double positive then will be single positive (either CD4 or CD8)

(وآخر دعوانهم أن الحمد لله رب العالمين)
لا تنسوني من صالح دعائكم
الله يرضى عنكم ويوفقكم

V3

التعديل باللون الاحمر والاصفر
فقط

-RAG enzyme starts at the pro stage (where antigen receptor gene rearrangement occurs and continues until the **Immature** stage)

TCR α -chain and β -chain loci each contain multiple variable region (V) gene segments, numbering about **30 - 45** and one or a few constant region (C) genes.

• **IgM, IgD** => *mature B cell*

Immature B cells have IgM

****Mature B cells have IgM+IgD**