

***Title of Lecture: complement system and MHC***

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Revision
We said that there are 3 types of complement pathways,
1- The classical pathway : in this pathway complement #1 will start the reaction and all the nine complement proteins are involved in the classical complement pathway
2- The alternative pathway , 3- Lectin pathway “to be discussed here”

- The alternative pathway
activation of the classical pathway is by a complex made between “IgM ,IgG” and the antigen “Ag-Ab complex” on the other hand activation of the alternative pathway is done by spontaneous convergent of C3 to C3b.
Note: normally “without any microorganism present in the body” there is hydrolysation of C3 to (C3a and C3b) but in a very low concentration, but if there is a microorganism present in the body “or antigen” the amount of C3b will be amplified.
Amplification: certain molecules are produced for this process, one of these molecules is “factor B” which binds to “C3b” the result is “C3bB”
Then “factor D” will be involved to divide “C3b**B**” to “C3b**B**a” and “C3b**B**b” (factor **B** has been divided to “**B**a and **B**b”)
“C3bBb” is a convertase enzyme which cleaves more C3 molecules to “C3a and C3b” and that’s how amplification happens.
 Note: the first C3b results from physiologic hydrolysis “and may be called iC3b” , but the second C3b results from activation of C3 convertase enzyme “also known as C3bBb”
If there is no microorganism present in the body “C3bBb” will be cleaved, and its amplifying effect will be stopped by “Factor I and Factor H”.
But if there is microorganism present in the body the “C3bBb” will attach to its membrane and this will protect it from the effect of “Factor I and Factor H”
Then when there is sufficient amount of “C3bBb” C5 will be activated, which will divide into “C5a and C5b”, “C5b” will start the attack on the microorganism, and “C5a” which is a small molecule will be released to do some biological functions.
“C3a, C5a” (the molecules that have not been discussed above) are antitoxins and have biological functions (they increase permeability of the cell, they have receptors on the mast cells) .. will be discussed later.

After “C5b” is produced, it will attach to the cell membrane and trigger C6, C7, and C8 to attach to the cell membrane, then this complex “C5b, C6, C7, and C8” will activate C9, which will be polymerized and works as a “needle” in the wall of the microorganism, and this will cause lysis to the cell, because the internal osmotic pressure of the cell is much more higher than the external osmotic pressure.

So in comparison: the end result of the classical and alternative pathways is the same, but they differ in the initiation mechanism.

- Lectin pathway “mannose binding lectin pathway”

MBL “mannose binding lectin” structure is homologous to C1 “which is the first complement protein used in the classical pathway”.
MBL is found on macrophages and other cells.
MBL acts as C1 so after it C4 will come then C2 then C3 … etc. (like the classical pathway)
so the end result will be like the previous two pathways but the initiation is different.

Complement proteins have different receptors on different cells “CR”
CR1 is found on RBC’s (so in case of hemolytic anemia this receptor will increase in the blood)
CR2 is found on B-lymphocytes
CR3 is found on macrophages
these receptors are important in activation of antigen reactions and are important as phagocytic molecules.

Biological functions of the complement system
in classical pathway C2 is divided into “C2a” and “C2B” (NOTE: when the C is divided into “a and b” the “a” type is always smaller than the “b” –for example when C3 is divided into C3a and C3b, C3a will be smaller than C3b- except in C2 “here C2a is larger than C2b”)
C2a is kinin like molecule “kinin is part of the coagulation cascade” so C2a works nearly the same as kinin, so it increases the permeability of the vessels and edema will result in this specific area.
C3a, C4a, C5a are chemotactic factors, anaphylatoxins (means that they cause release of granules from granulated cells; like mast cells)

If there is insufficient amount of CR3 on the macrophages they will not perform phagicytosis efficiently.
C3b and C4b are responsible for removal of immune complexes (when antigen interacts with antibody a complex will result but these complexes need to be removed from the body by the macrophages, C3b and C4b are needed by the macrophage to remove these complexes)

In summary:
- complements activate the macrophages and natural killer cells “C3b”
- anaphylatoxins “C3a and C5a”
- chemotctic factors for neutrophils and macrophages “C5a”
- membrane attack complex “from C5b – C9”
-removal of immune complexes “C3b, C4b, iC3b”

Regulation of the complement system is very important, without regulation complement system can be catastrophic.
In the classical pathway “C1” should be regulated because this pathway begins with it, so we have C1 inhibitor.
C3b is spontaneously hydrolyzed in the plasma, then it’s either amplified or removed so we can regulate this thing. (I think he’s talking about the Alternative pathway in this point)
C3 convertase could also be inhibited, by plasma factor 1, Decay accelerating factor “DAF”, and membrane cofactor protein.
Membrane attacking complex can also be regulated by some enzymes “present in the slides”.

Deficiencies of the complement proteins cause diseases.
- Deficiency in early complements “C1,C2, and C4” will cause autoimmune diseases (because if there is any changes in human cells the immune cells have to remove them “by phagocytosis” (which happens in the presence of the complement system), so if there is deficiency in the complement system these abnormal human cells will remain in the body and autoimmunity will occur against them).
Also deficiency in the complement system can lead to fatal biogenic infections. (ordinary bacterial invasion will lead to severe infection and lead to death because of the absence of the complement system).
NOTE: early complements “C1,C2,C4”, middle complements “C3,C5”, late complements “C6, C7, C9”
- Congenital deficiency may be present in C2 which will lead to deactivation of the classical complement pathway.
- C1 inhibitor deficiency causes hereditary angioedema (the lymph nodes will enlarge and there will be a lot of edema, the patient may die from asphyxia (suffocation).
- Paroxysmal Nocturnal hemoglobiuria is an immune problem due to the decay factor.

Measuring complements in the lab
- by Ag-Ab reactions
- by precipitation reactions
we can measure C4 quantitatively, and we can measure CH50 (to look for the whole system of complements)
CH50 is calculated through Ag-Ab complexes, we take sheep blood with antibodies (here the antigen is RBC’s of the sheep and the antibody is IgM), and we take serum from the human being “which contains the complement system”.
Then we dilute the human serum and we put these different solutions in different containers (1/2 diluted, ¼ diluted, 1/16 diluted, 1/64 diluted … etc).
If the complement system is functional it should lyse the RBC’s of the sheep at certain dilution.
The solution of the diluted serum which will cause lysis to 50% of the sheep’s RBC’s will be the CH50
(If 50% of the RBC’s has been lysed at solution that is 1/64 diluted then this is the CH50), and this can tell us if the complement system is present in normal, high or low levels.
If the result were abnormal “high or low” we should examine each complement protein “C1, C2, C3 … etc” alone to see where the problem is.

In addition to autoimmune diseases complement system is important in rheumatic diseases, SLE.

Complement system is important in:
-innate immunity
-opsonization
-lysis
-chemotaxis
-cell activation.
In adaptive immunity complement system is important for:
-augmentation of antibody response
-promotion of T-cell response
-elimination of self reacting lymphocytes
-enhancement of immunological memory
-clearance of Ag-Ab complexes
-apoptosis.

-Major Histocompatibility Complex. (MHC)

<<< Major histocompatibility complex (MHC), group of genes that code for proteins found on the surfaces of cells that help the [immune system](http://www.britannica.com/EBchecked/topic/283636/immune-system) recognize foreign substances. MHC proteins are found in all higher vertebrates. In human beings the complex is also called the human leukocyte antigen (HLA) system.
There are two major types of MHC protein molecules—class I and class II. Class I MHC molecules span the membrane of almost every [cell](http://www.britannica.com/EBchecked/topic/101396/cell) in an organism, while class II molecules are restricted to cells of the [immune system](http://www.britannica.com/EBchecked/topic/283636/immune-system) called macrophages and lymphocytes. In humans these molecules are encoded by several genes all clustered in the same region on [chromosome](http://www.britannica.com/EBchecked/topic/116055/chromosome) 6. Each [gene](http://www.britannica.com/EBchecked/topic/228226/gene) has an unusually large number of alleles (alternate forms of a gene that produce alternate forms of the protein). As a result, it is very rare for two individuals to have the same set of MHC molecules, which are collectively called a [tissue type](http://www.britannica.com/EBchecked/topic/597057/tissue-typing). The MHC also contains a variety of genes that code for other proteins—such as [complement](http://www.britannica.com/EBchecked/topic/129861/complement) proteins, cytokines (chemical messengers), and enzymes—that are called class III MHC molecules.>>> from internet for hopefully better understanding

MHC is a molecule “protein” found on all nucleated cells in the body “RBC’s don’t have them” , it’s inherited, it’ll not change throughout life “unlike T-cell and B-cell receptors which change throughout life”.
MHC’s must be present in order for T-cell to recognize the antigen.
 MHC was discovered in the 1980’s by three scientists, they said that MHC is genetically determined cell membrane structure and they are important in regulation of the immune system, they studied them on mice first then they studied them in the human body. Their function is to present antigen fragments (epitopes) to the T-cells” and they’re important in immune recognition “in the adaptive immune system”, and they are important in transplantation.
MHC are controlled by a gene present on chromosome 6 in the human being, they’re polymorphic “they have many types”, they’re also co-dominant “both alleles will be expressed on the cell membrane”.

MHC consists of two classes: (I’m not sure about this info)
class 1 : which has three subtypes (a,b,c) “each subtype controlled by a gene”
class 2 : which has three subtypes (dp,dq,dr) “each subtype controlled by a gene”.

 Each gene of the previous subtypes will express a protein “molecules”, and since each molecule is controlled by two alleles “maternal and paternal” there should be 12 molecules at least on the cell membrane, these molecules can be homogeneous or heterogeneous.

The gene that encodes class 2 is around 3600 kilobase, which is a large gene.

The inheritance of MHC happens by taking one allele from the father and the other allele from the mother.

Class 1 is composed of two polypeptides alpha and beta “the alpha polypeptide is produced by the cell itself but beta polypeptide is coming from outside” the alpha polypeptide has different subtypes “alpha-1, alpha-2 … etc”.
In class 1 alpha-1 and alpha-2 make the groove where the epitope will react. (The beta polypeptide is not expressed by MHC gene as we said it comes from outside and only bind to class one to make it in the activated form “beta polypeptide is actually part o the super-family of the immunoglobulin and produced in the liver and is called <beta microglobulin>”), alpha-3 polypeptide forms a tail that passes through the cell.

Class 2 is composed of two polypeptide chains which are different from each other, we have alpha chain and beta chain, and the antigen binding site consist of alpha-1 and beta-1, here alpha-2 and beta-2 forms a tail that passes through the cell, in class 2 there is no beta microglobulin.

Class 1 MHC is found on cytotoxic T-cells (CD8), and class 2 MHC is found on T-helper cells (CD4), while other nucleated cells in the body have both types.

The groove that binds to the antigen on the MHC class 1 can hold from 8-11 amino acids, while class 2 can hold 10-30 amino acids, which means that the groove in class 2 is much larger than class 1.

 Class 2 MHC is very much concentrated on antegin presenting cells “macrophages” and on B-cells,
and on the thymus epethilium (they are responsible for teaching the T-cells when they come to the thymus during development).
Gamma-interferon is produced by T-cells, and causes increase in production of class 2 MHC on these cells.

Cell activation determines how much MHC is found on the cell
class 1 is important in regulation of antiviral immune response, while class 2 is important in regulation of intracellular infections.
RBC’s can’t be attacked by viruses because there is no MHC on their surface.

We said that the epitope binding groove on the MCH molecule in class 1 can hold up to 11 amino acids, but actually when an epitope “which consist of amino acids” interacts with the binding groove not all the 11 amino acids sites will be occupied “only two of them will interact; for example amino acid number 5 and amino acid number 9, and when different epitope comes it interacts in different way, it can interact by amino acid number 6 and number 7 for example” so there are many ways of how the epitope can interact with the MHC and this explains why small number of different types of MHC “12 types” can interact with a huge variety of antigen types.

Class 1 is expressed on all nucleated cells, while class 2 is expressed primarily on the immune cells.
In humans MHC is called HLA.
We have proximately 2100 different types of HLA in people.

If we said that in certain population, 5% of the people have type A1 MHC, and 10% of the population have B1 MHC, if we want to find out how many people have B1 and A1 MHC together, we should multiply 10% with 5% “5%\*10%=0.5%”
but this is not the case in MHC calculations, because the actual result if we did tests on people it will be completely different than the number we got in the mathematical calculation, and this phenomena is called linkage disequilibrium and haplotypes. (it’s important in finding what is called matching zone).

Also the prevalence of HLA types is different from one population to another “certain HLA type may be found more in black people for example, or Chinese … etc”