ISSN- O: 2458 - 868X, ISSN–P: 2458 – 8687
National Library of Medicine - ID: 101731606

International Journal of Medical Science and Innovative Research (IJMSIR)
IJMSIR : A Medical Publication Hub
Available Online at: www.ijmsir.com
Volume – 6, Issue – 1, February – 2021 , Page No. : 96 - 106

Basics of Complement System – A Review on in its Activation Cascade

[1]Dr Aashi Fazeela M, PG Trainee, Deptt. of Oral Pathology & Microbiology, Government College of Dentistry, Indore
[2]Dr Jaya Joshi, Professor and Head, Deptt. of Oral Pathology & Microbiology, Government College of Dentistry, Indore.
[3]Dr Kundendu Arya Bishen, Assistant Professor, Deptt. of Oral Pathology & Microbiology, Government college of Dentistry, Indore
[4]Dr Ankit Goyal, PG Trainee, Deptt. of Oral Pathology & Microbiology, Government college of Dentistry, Indore
[5]Dr Ashish Verma, PG Trainee, Deptt. of Oral Pathology & Microbiology, Government college of Dentistry, Indore
Corresponding Author: Dr Aashi Fazeela M, PG Trainee, Deptt. of Oral Pathology & Microbiology, Government College of Dentistry, Indore

Citation this Article: Dr Aashi Fazeela M, Dr Jaya Joshi, Dr Kundendu Arya Bishen, Dr Ankit Goyal, Dr Ashish Verma, "Basics of Complement System – A Review on in its Activation Cascade", IJMSIR- February - 2021, Vol – 6, Issue - 1, P. No. 96 – 106.
Type of Publication: Review Article
Conflicts of Interest: Nil

Abstract

Complement system is an imperative part of innate immune system. It is composed of a group of plasma or membrane proteins that are found in a deactivated state within the body. On exposure to a particular stimulus that triggers this protein, a chain of reactions gets initiated. These reactions referred to as activation pathways are of three types: Classical pathway, Lectin binding pathway and Alternative pathway. In each step of these pathways, the complement proteins undergo conformational and chemical changes to bind to subsequent proteins ending with the formation of Membrane Activation Complex, the final product. The cleavage products formed as a result of chain reactions cross interacts with adaptive immune system and non-immune systems and influences it to carry out functions. Over activation of complement harm the autologous tissue and hence need to be closely monitored by regulatory proteins. These proteins act as a check point in each step of complement activation cascade thereby maintaining the physiologic state of host cells. Through this review an attempt is made to describe the basics of complement system; its activation and regulation.

Keywords: Complement system, Classical pathway, Lectin Binding pathway, Alternative Pathway, Receptors and Regulators.

Introduction

The immune system is main defensive armour of our body. It protects the body from pathogen or other harmful elements by generating immune response which are of two types, innate(inborn) and adaptive(acquired). Complement forms a major part of the innate immunity [1]. It is composed of large number of proteins including plasma proteins produced by the liver and membrane proteins expressed on cell surface. These proteins are found in a precursor form which gets

activated in a sequential manner to perform its functions. The activated proteins and their products bind to specific receptor (membrane expressed) to cause its effect [2].

On evolutionary terms, origin of complement is much more ancient than Adaptive immune system[4]. For a long time, complement was considered as a supportive part of innate immune system with only known function of recognition and elimination of pathogens [3]. In recent years the versatile role of complements has been better understood [5]. It interacts with other effector/ regulatory system thereby making it an important element in providing adaptive immunity, haemostasis, and neuroprotection**.** Role of complement in pathogenesis of many diseases have also been established [6]which sheds light into its cell- cell and cell- stroma interactions.

Unfortunately, due to complexity of different types of enzyme cascade reactions and their cross interactions, researchers consider this as one of the most uninteresting and incomprehensible topics in immunology [3]. Thereby a deep insight into it is frequently eluded.

A better understanding of the basics of complement forms the foundation for future studies. This review is an attempt to simplify the complement system, its path of activation, with special emphasis to its component and regulator proteins that keeps the body in its physiological state.

Physiology of Complement System

The complement system is made up of a number of distinct serum (blood plasma) and membrane proteins in an inactivated state. To perform its function, complement proteins needs to get activated [2]. Activation occurs via 3 different pathways:

a) Classical pathway	Antibody dependent pathway Is initiated by formation of soluble antigen-antibody complex
b) Lectin Pathway	Antibody independent pathway It is initiated when Mannose/ N-acetyl glucosamine residues containing pathogen approaches.
c) Alternative pathway	Antibody independent pathway Stimulated by antigen directly eg. Bacterial cell surface components
d) Terminal pathway	Final common pathway

In all the three pathways, complement activation can be better understood under these 3 stages:

1. Formation of C3 convertase
2. Formation C5 convertase
3. Formation of Membrane Activation Complex (MAC)

The first two stages are unique for each pathway, leading to common terminal pathway ending with the formation of MAC.

A cascade of proteolytic cleavage precipitates with the initial activation of complement proteins. Here, each precursor of complement is cleaved into two fragments: Major/larger fragment (designated as ‘b’) and smaller fragment (as ‘a’**).** The larger ‘b’ fragment remains bound to its site of activation i.e cell membrane. It acts either as a receptor for activation of subsequent proteins in the chain or as an enzyme for cleavage of complement proteins [7-8].

The final effect of complement is brought about by interaction of MAC and intermediate proteins of activation cascade with their receptors present on different cell types and the overt activation of complement against host cells is kept under strict vigilance by the regulatory proteins.

Activation of Complement System

Classical pathway: Classical pathway of complement activation is triggered by Antigen- Antibody complex [8]. The sequential steps of activation cascade are as follows:

- When an antigen enters the body, antibody recognises and binds to it [9].
- This leads to conformational changes in the Fc portion of antibody causing exposure of binding site for C1, the initiator complement protein of Classical pathway [10].
- C1 is a hetero oligomeric complex (Figure 1a) consisting of one C1q, two C1r and two C1s molecules. Binding of C1 to IgG/IgM causes conformational changes in C1q leading to activation of C1r which in turn activates C1s [9].
- C1s enzymatically cleaves C4, the next component of complement. C4 (has 3 chains α, β, γ) is cleaved at a single site near the amino terminal of its alpha chain producing smaller C4a and larger C4b fragments. C4b remains bound to the membrane with an exposed reactive thioester group (receptor) whereas C4 a is released from complex (Figure 1b). C4b acts as a receptor for next component i.e C2 [9].
- C4b in presence of Mg^{2+} ions get attached to C2 which further gets cleaved by C1s within the complex(C1qrsC4b) cleaves C2 leading to formation of C2a and C2b (Figure 1c) [9].
- The cleaved product, C2a (an exception, where "a" is the larger fragment) remains attached to C4b, together forming the complex C3 convertase(C4b2a), whereas C2b diffuses away from the site [9] (Figure 1d).
- C2a binds non covalently to C3 and cleaves it into C3a and C3b [7].

a. C3b formed can have three possibilities: It can bind either to the host cell membrane (thereby acting as a component for alternative pathway) or can coat the microorganism (opsonisation) or can bind to C4b2a complex to continue the classical pathway activation cascade [12-13].

a. The enzyme thus formed is C4b2a3b complex which is referred to as C5 convertase. (Figure 1e)

- C5 the next component protein, binds non covalently to C3b in C4b2a3b complex and gets cleaved by C2a of complex. Two fragments C5a and C5b gets released, where C5b is the binding site for the subsequent protein C6.

The rest of the activation cascade is similar in all the three pathway of activation which is later described under Common Terminal Pathway.

Figure 1: Classical pathway of complement activation

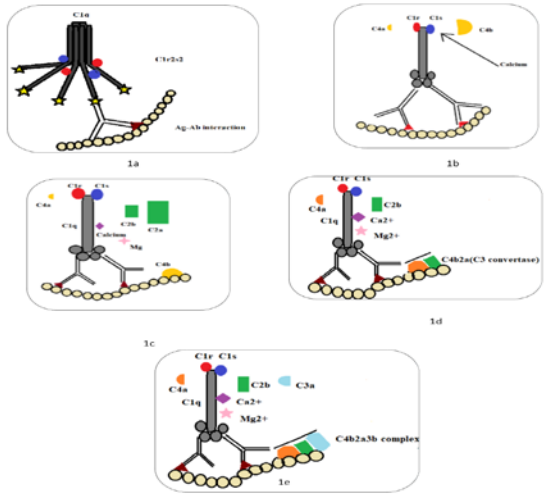


Figure 1: Shows the sequence of events happening in classical activation pathway (from activation of C1 to formation of c5 convertase) (1a) C1q binds to Ag-Ab complex in the membrane leading to activation of C1s via C1r. (1b) C4 gets cleaved by c1s to produce C4a and C4b; C4b remains bound to membrane whereas C4a is released (1c) C4b becomes active and exposes a reactive site where C2 binds to and gets cleaved by C1s into C2a and C2b (1d) C2a gets bound to c4b to produce C3 convertase(C4bC2a) (1e) C3 convertase cleaves C3 to C3a and C3b,C3b gets bound to the complex thereby producing C5 convertase (C4bC2aC3b)

The proteins that participates in classical and lectin pathways are similar. The structure and molecular weight of these proteins affect its affinity for different ligands hence determines its function. Therefore, it becomes essential to have an insight into the properties of each individual proteins. The details are assembled in Table 1.

Lectin Pathway: Lectin pathway is activated when a pathogen with mannose and N-acetyl glucosamine residues on their surface (seen on bacterial cell wall) approaches the host cell [9]. Mannan-binding lectin (MBL- also termed mannan-binding protein/MBP), the key component of lectin binding pathway [11],binds to these residues on the surface and leads to onset of lectin pathway.The structure and function of MBL is same as C1q (Classical pathway) – MBL is a tetrameric complex with globular binding regions and a collagenous stalk. This stalk in MBL is associated with a protease termed as MBL-associated serine protease (MASP) (similar to C1r and C1s of classical pathway) [14]. MBL-MASP complex is formed (similar to the one formed by C1r and C1s) which cleaves C4 and then C2 forming C3 convertase(C4b2a). The rest of the pathway is similar to classical pathway of complement activation [9].

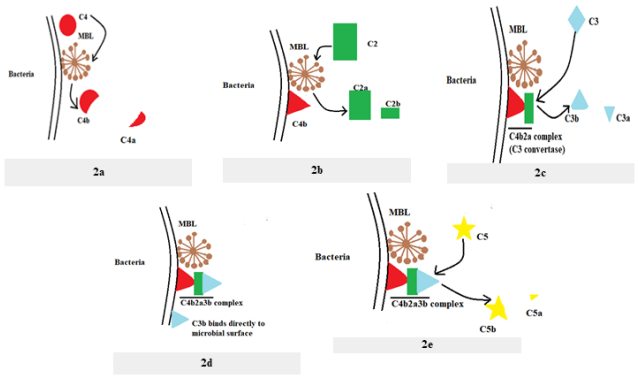


Figure 2: Shows the sequence of events happening in the lectin binding pathway (2a) MBL cleaves C4 into C4a and C4b; C4b remains attached to the membrane.(2b) MBL cleaves C2 intoC2a and C2b; C2a binds to C4b to form C3 convertase (2c) C3 convertase cleaves C3 into C3a and C3b (2d) C3b can directly bind to cell membrane as well as bind to complex forming C5 convertase(2e) C5 convertase cleaves C5 into C5a and C5

Alternative Pathway: Alternative pathway is different from other two as it is constantly activated at a slow rate within the body [15]. In host cells, this activation is controlled by the presence of certain regulatory proteins on their surface. When constituents that lack these regulators eg. a foreign cell, pathogen etc are present, a full-blown activation of alternative pathway is noted. The surface constituents that evoke the pathway may be lipopolysaccharide, fungal cell walls, or viral envelopes.

When a pathogen enters the host body, inflammation ensues leading to release of all the inflammatory mediators including complementary proteins. The C3 molecules of the complement gets activated by directly contacting antigen. C3 with the help of other three proteins, factor B(fB), factor D(fD) and properdin forms the main component in this pathway.Formation of C3 convertase can occur in two ways:

In first type:

1.C3 present in plasma in a deactivated form undergoes spontaneous hydrolysis and forms C3(H_2O) molecule.

2.C3(H_2O) has many characteristics of C3b and binds factor B in solution. This complex is subjected to cleavage by factor D.

3.As a result of cleavage, Bb and Ba are produced. Bb binds to C3(H_2O) to form fluid phase C3 convertase i.e [C3(H_2O)Bb] (Figure 3a) [15, 6].

4.C3 convertase converts C3 into C3a and C3b akin to that in classical and lectin pathway.

In second type

1.Spontaneous conversion of C3 (the key component of this pathway) to C3b occurs in body through the C3 convertase that is already present (from the previous loop of alternative pathway)

2.C3b binds Factor B and presents it for cleavage by Factor D thereby producing Ba and Bb [9].

Bb binds to membrane bound C3b leading to formation of C3 convertase(C3bBb). Properdin is a regulatory protein that acts in this step. It binds to the convertase complex and prolongs its active life span [17].

3.C3 convertase cleaves C3 molecules into C3a &C3b, where C3b can either bind to cell membrane and form the receptor site for factor B or can form a complex with C3bBb to form C5 convertase(C3bBbC3b*)* (Figure 3b) [9].

Figure 3: Alternative pathway of complement activation

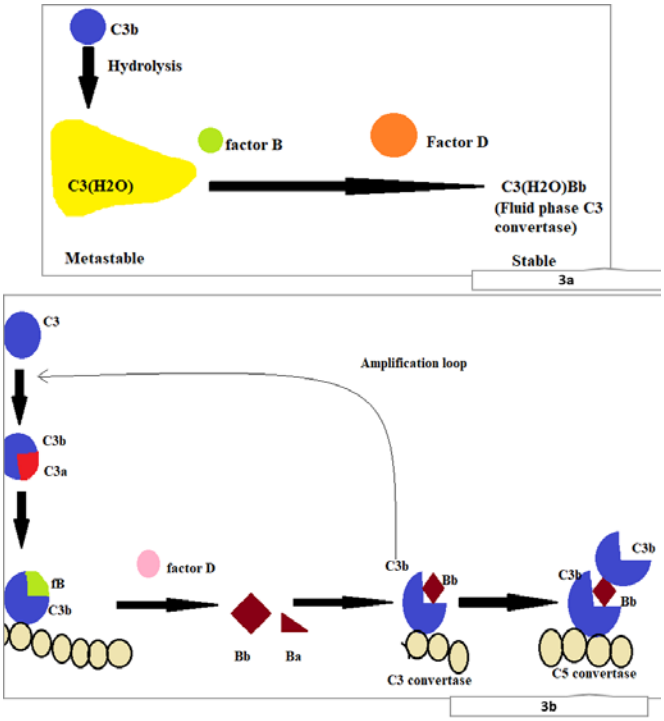


Figure 3: Shows the sequence of events happening in alternative activation pathway(3a) shows formation of fluid phase C3 convertase(3b) shows cleavage of C3 into C3a and C3b with the help of c3 convertase, C3b binds to membrane which is acted upon by fB and fD to produce C3 convertase (c3bBb), C5 convert (C3bBbC3b) is formed when another molecule of C3b binds to C3bBb.

In normal cells C3b is quickly inactivated by proteins on the surface of own cell walls, hence short lived. As bacteria or other foreign material lack these surface proteins C3b stays active and continues this pathway in an amplified manner leading to increased formation of intermediate products and MAC [18].The complement protein participating in alternative pathway, their structure, function and molecular weight are summarized in Table 1.

The Terminal Pathway (The Membrane Attack Pathway): This pathway is the last phase of the three-activation pathways. Here an amphipathic membrane inserted complex is formed by non-covalent association of C5b with four terminal C components (C6, C7, C8, C9). The sequence of events are as follows:

1) C5b formed via any of the three-complement pathway binds to C6 thereby stabilising the complex(C4b2a3b5b6) on the membrane.

2) Binding of C6 exposes a binding site in the complex for the subsequent protein i.e C7 [9].

3) Attachment of C7 leads to release of the C5bC6C7 complex from convertase complex(C4b2a3b). C5b67 complex is now present in the fluid phase.

4) During the formation of **C5b67** complex a transient lipid membrane binding site within it is exposed [9]. Through this the trimeric complex binds closely to the cell membrane thereby stabilising it.

5) C8, the next component, has α, β and γ chains. It uses β chain to bind to C7 resulting in the formation of the complex **C5b678**.C5b-8 gets deeply buried in the

cell membrane and forms small pores making the membrane leaky [19,9].

6) α chain of C8 in the complex binds to C9 leading to its conformational change from a globular, hydrophilic form to a linear amphipathic form. This helps the complex to traverse the membrane and increase leakiness.

7)Additional C9 molecules are also recruited to the C5b-8 complex leading to formation of an assembly of complex called MAC [9].

Figure 4: Terminal pathway of complement activation

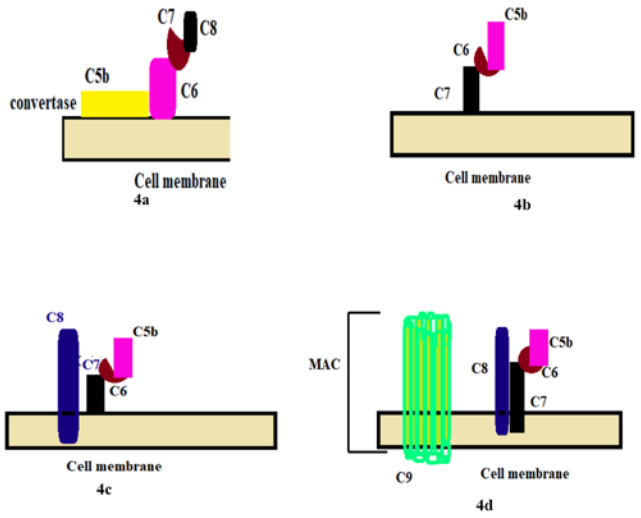


Figure 4: Shows the sequence of events happening in the terminal pathway (4a) shows the formation of C567 complex (4b) attachment of C7 causes detachment of complex from C5 onto liquid phase (4c) C8 in the complex of C678 deeply buries itself to the cell membrane(4d) shows the final assembly of MAC

Complement protein of Terminal pathway, their structure, function and molecular weight are summarized in **Table 1:**

Receptors of The Complement Components

The effect of the mediators released during complement activation is generated only when it binds to specific receptors. Complement Receptors (CR) recognizes pathogens, immune complexes and cell debris that are opsonized by C3 fragments (opsonin). These receptors present on cell surfaces (phagocytes) generate pro-inflammatory response or tolerogenic suppression when bound to the complement fragments. CR has three different structural organizations: Complement Control Protein (CCP)modules- CR1 and CR2, Integrin family members- CR3 and CR4, Immunoglobulin superfamily member- CRIg [15].

CR1: CR1 is the receptor for the cleavage product C3b and C4b and is seen on erythrocytes, PMNL,Mononuclear Phagocytes, B cells, T cells and mesangial phagocytes [2].The receptor on activation in erythrocytes helps to bind and process the immune complex in vivo; in PMNL & Mononuclear phagocytes it promotes phagocytosis; whereas in B cells it acts as **processing molecule**(converts C3b to inactivated C3b(iC3b)) and sends contrasting signals to **CR2** to down regulate B cell response against C3b coated antigens [16].

CR2: Following the generation of C3b, this molecule is degraded by **Factor I** and cofactor (**CR1) into iC3b and C3dg** and then into **C3d** through nonspecific proteases. The latter three interacts with the receptor **CR2/CD21** which is seen on B cells, epithelial cells, follicular dendritic cells(FDC),Thymocytes and subset of peripheral T cells [2]. These receptors once activated, promotes B cell activation by the C3 fragment opsonised antigen and traps immune complexes on Follicular Dendritic Cells (FDC) within lymphoid tissues. This becomes an immunologic memory of the host.

CR3 and CR4: CR3 and CR4 are the receptor of iC3b, seen on Macrophages, Mononuclear Phagocytes, PMNLs, FDC.On activation CR3 and CR4 receptor binds to iC3b [2].

CR5:CR5 is expressed on Kupffer cells. With activation it causes clearance of complement coated particles and immune complexes in circulation [2].

Additional receptors

C5aR, CD88: This recognizes C5a which has pro-inflammatory property like leukocyte chemotaxis, aggregation of neutrophils and platelets, release of mast cell mediators and generation of leukotrienes, cytokines, and reactive oxygen metabolites. It also engages cross talks with Fc receptor of IgG.

C3aR: It binds to C3a and expressed on neutrophils, monocytes/macrophages, mast cells, hepatocytes, bronchial/alveolar epithelial cells, vascular endothelium, and astrocytes. On activation, it plays key roles in inflammatory disorders.

Receptors of MAC: As MAC has no specific receptor, it gets directly inserted into the cell membrane. This insertion can be either transient(non-lethal) or permanent/matured(lethal). When transient, growth factors and cytokines are released either directly through MAC pore(eg: Interleukin-1) or through canionical pathways produced in response to MAC. These leaked molecules act on its receptors and bring about its effect by promoting cell proliferation, inflammation, and thrombosis. As transient MAC performs immune functions via transmitting biological molecules, the matured MAC produced perform its immune function by direct lysis of target cells [29-30].

Regulation of Complement System

To function in a precise manner, Complement is controlled by certain regulatory protein that acts as a switch to either increase or decrease its expression. The human body is equipped with a large array of both fluid-phase and membrane bound complement inhibitors for protection of host tissue from autologous damage. The complement is positively as well as negatively regulated in the body. An intricate network of cytokines determines the expression of these regulatory molecules.Positive regulation is through Properdin which acts to prolong the duration of activation of C3 convertase [2].

Negative regulation is via many proteins including:

In Plasma

a) C1 inhibitor- Binds to activated C1 and removes C1s and C1r from the complex

b) Factor I-Inactivates convertases by cleaving C3b and C4b.

c) Factor H-Catalyses fI associated cleavage of C3b in the alternative pathway

d) C4bp- Causes cleavage of C4b in Classical pathway convertase

e) S protein (Vitronectin)- Targets the hydrophobic membrane binding site in C5b-7 complex when it is in fluid phase

f) Clusterin-Targets the hydrophobic membrane binding site in C5b-7 complex when it is in fluid

In Membrane

a) DAF (Decay Accelerating Factor)- Binds to and breaks convertase

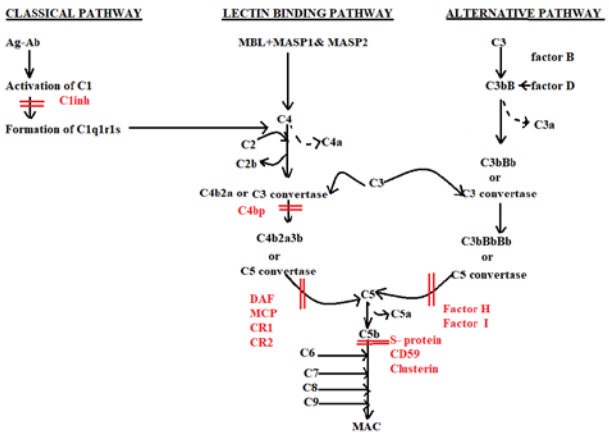
b) MCP (Membrane Cofactor Protein)- Binds to convertase and acts as co factor for the action by Fi

c) C Receptor 1: Causes inactivation by causing dissociation of convertase as well as acting as a co factor for action of fI.

d) CD 59: Binds to C8 in the complex and prevents assembly of C9

The intersection of various regulators of complement in different pathways are summarised in *Figure 5*.

Figure 5: Different pathways of complement activation and levels of action of the corresponding regulators.



Important Roles of Complement System

When the complement is activated and regulated normally, it is able to accomplish its functions. Important roles performed by complement includes:

a) Compliment as a First Line of Defence Against Pathogens:

Complement system helps the innate immunity in host defence through three effector pathways:

1. Lysis/Direct killing: When pathogen enters the body, MAC [20] punches minute pores on its cell surface bringing about osmotic imbalance between the foreign (bacterial) cell and surrounding environment. It leads to lysis/direct killing of affected cells/pathogens [21-22].

2. Opsonization and Phagocytosis: In pathogen elimination, main role of complement is indirect. C3b is an opsonin and gets deposited on the surface of pathogen. This helps the phagocytic cells to recognize, ingest and destroy the antigen [23].

3. Inflammation:C3a, C5a produced during activation cascade acts as potent anaphylatoxins. C3aR and C5aR (trans-membranous G protein coupled receptors) [24-25] are the receptors for these and are predominantly expressed by cells of myeloid origin granulocytes, monocytes/ macrophages, mast cells, and some dendritic cells [26]. These interact with their receptors to produce functional response via causing inflammation.

b) Protection of the Host Cells Against Complement Attack: Complement system follows a rule, it attacks everything that is not protected specifically. Host cells are protected as they express complement regulatory molecules on their surface or recruit it from plasma [3,23].The absence of Complement regulatory proteins marks cell as foreign or altered leading to increased accumulation of C3b (opsonin) on cell surface bringing about excessive complement activation and finally its elimination [3].

c) Immunologically Silent Phagocytosis of Apoptotic cells: The complement plays a major role in providing immunologically silent apoptosis. During apoptosis cells undergo molecular alteration and expression of certain “eat me” signals on their surface. These signals are recognized by phagocytes leading to execution of cell without an immunologic response [3,23].

d) Cell Homeostasis: Cells in native state secrete complement thereby generating its cleavage products such as C3a and C5a in the microenvironment [7]. These products are important to maintain the intracellular homeostasis, especially of T cell [28].

Conclusion

“Complement is a Double-edged sword”. A slight dysregulation can have hazardous effects. Apart from being a vital component of innate immune system, complement also acts as a bridge between the innate and adaptive immunity as well as other non-immune systems (such as coagulative system) and in pathogenesis of many disorders. This review is to introduce the reader onto the beautiful world of complement system, its complex pathway of activation, the effects of its activation and ultimately its regulation.

"You can't build a great building on a weak foundation. You must have a solid foundation if you are going to have a strong super structure"- Gordon B Henckley. It is essential to understand the basics of this intricate cascade of protein system to take on further studies related to it.

References

1. Rus H, Cudrici C, Niculescu F. The role of the complement system in innate immunity. Immunol Res. 2005;33(2):103–12.
2. Holers, V Michael. "Complement and its receptors: new insights into human disease." Annual review of immunology 2014;32: 433-59.
3. Merle NS, Noe R, Halbwachs-Mecarelli L, Fremeaux-Bacchi V, Roumenina LT. Complement system part II: Role in immunity. Front Immunol. 2015;6:1–26.
4. Evolution of the complement system Masaru Nonaka. Curr Opin Immunol. 2001;13(1):69–73.
5. Dunkelberger JR, Song WC. Complement and its role in innate and adaptive immune responses. Cell Res. 2010;20(1):34–50.
6. Zhang R, Liu Q, Li T, Liao Q, Zhao Y. Role of the complement system in the tumor microenvironment. Cancer Cell Int [Internet]. 2019;19(1):1–12.
7. Lubbers R, van Essen MF, van Kooten C, Trouw LA. Production of complement components by cells of the immune system. Clin Exp Immunol. 2017;188(2):183–94.
8. Manuscript A. Ricklin D, Hajishengallis G, Yang K, Lambris JD, Complement: a key system for immune surveillance and homeostasis. Nat Immunol. 2010;11(9):785–797.
9. Morgan BP. Chapter 10 Activation and control of the complement system. Princ Med Biol. 1996;6:171–196.
10. Tulamo R, Frösen J, Junnikkala S, Paetau A, Kangasniemi M, Peláez J, et al. Complement system becomes activated by the classical pathway in intracranial aneurysm walls. Lab Investig. 2010;90(2):168–79.
11. Auriti C, Prencipe G, Moriondo M, Bersani I, Bertaina C, Mondì V, et al. Mannose-Binding Lectin: Biologic Characteristics and Role in the Susceptibility to Infections and Ischemia-Reperfusion Related Injury in Critically Ill Neonates." Journal of immunology research vol. 2017 (2017)
12. Walport MJ. Complement. First of two parts. N Engl J Med [Internet]. 2001;344(14):1058–66.
13. Walport MJ. Advances in Immunology BETWEEN INNATE AND ADAPTIVE. English J. 2001;344(15):1140–4.
14. Matsushita M, Fujita T. Activation of the classical complement pathway by marmose-binding protein in association with a novel C1s-like serine protease. J Exp Med. 1992;176(6):1497–502.
15. Brown EJ. Complement receptors, adhesion, and phagocytosis. Infect Agents Dis. 1992;1(2):63–70.
16. Tuveson DA, Ahearn JM, Matsumoto AK, Fearon DT. Molecular interactions of complement receptors on B lymphocytes: A CR1/CR2 complex distinct from the CR2/CD19 complex. J Exp Med. 1991;173(5):1083–9.
17. Weiler JM, Daha MR, Austen KF, Fearon DT. Control of the amplification convertase of complement by the plasma protein j1lH (amplification convertase inhibitor/C3b inactivator/properdin/C3 nephritic factor/alternative complement pathway). Immunology. 1976;73(9):3268–72.

18. Schmidt CQ, Lambris JD, Ricklin D. Protection of host cells by complement regulators. Immunol Rev. 2016;274(1):152–71.
19. Tamura N, Shimada A, Chang S. Further evidence for immune cytolysis by antibody and the first eight components of complement in the absence of C9. Immunology. 1972;22(1):131–40.
20. Bubeck D. The making of a macromolecular machine: Assembly of the membrane attack complex. Biochemistry. 2014;53(12):1908–15.
21. Campbell DG, Gagnon J, Reid KB, Williams AF. Rat brain Thy-1 glycoprotein. The amino acid sequence, disulphide bonds and an unusual hydrophobic region. Biochem J. 1981;195(1):15–30.
22. Morgan BP, Campbell AK. The recovery of human polymorphonuclear leucocytes from sublytic complement attack is mediated by changes in intracellular free calcium. Biochem J. 1985;231(1):205–8.
23. Merle NS, Church SE, Fremeaux-Bacchi V, Roumenina LT. Complement system part I - molecular mechanisms of activation and regulation. Front Immunol. 2015;6:1–30.
24. Klos A, Tenner AJ, Johswich KO, Ager RR, Reis ES, Köhl J. The role of the anaphylatoxins in health and disease. Mol Immunol. 2009;46(14):2753–66.
25. Klos A, Wende E, Wareham KJ, Monk PN. International union of basic and clinical pharmacology. LXXXVII. complement peptide C5a, C4a, and C3a receptors. Pharmacol Rev. 2013;65(1):500–43.
26. Noris M, Remuzzi G. Overview of complement activation and regulation. Semin Nephrol [Internet]. 2013;33(6):479–92. Available from: http://dx.doi.org/10.1016/j.semnephrol.2013.08.001
27. Kroemer G, Galluzzi L, Vandenabeele P, Abrams J, Alnemri ES, Baehrecke EH, et al. Classification of cell death: Recommendations of the Nomenclature Committee on Cell Death 2009. Cell Death Differ. 2009;16(1):3–11.
28. Liszewski MK, Kolev M, Le Friec G, Leung M, Bertram PG, Fara AF, et al. Intracellular Complement Activation Sustains T Cell Homeostasis and Mediates Effector Differentiation. Immunity [Internet]. 2013;39(6):1143–57.
29. Ghosh P, Sahoo R, Vaidya A, Chorev M, Halperin JA. Role of complement and complement regulatory proteins in the complications of diabetes. Endocr Rev. 2015;36(3):272–88.
30. Benzaquen LR, Nicholson-Weller A, Halperin JA. Terminal complement proteins C5b-9 release basic fibroblast growth factor and platelet-derived growth factor from endothelial cells. J Exp Med. 1994;179(3):985–92.

Legends Table

Table 1: (Modified from Morgan BP. Chapter 10 Activation and control of the complement system. Princ Med Biol. 1996;6(C):171–96

Complement Proteins

Complement Proteins Participating In Classical Pathway			
Component	**Structure**	**Molecular wt**	**Function**
• C1 (chromosome 12)	Large heterooligomeric complex, consisting of: - one molecule of C1q - two molecules each of C1r and C1s	460 kDa 80 kDa each	- C1q binds to Fc region of aggregated Ig G and causes conformational change leading to activation of other complexes(C1r) - C1s and C1r provides enzymatic action to C1 complex. - C1s in the complex cleaves and activate C4
• C4 (in chromosome 6)	Three disulphide bonded chains i.e α, β. γ	400 kDa	- C1s cleaves C4 to C4a and C4b (exposes reactive thioester group). - C4a acts as mediator for inflammation whereas membrane bound C4b provides receptor for next component, C2
• C2	Single chain	102 kDa	- C2 attaches to the binding site of C4b in presence of Mg ion and gets cleaved by C1s. - C2a, the larger component, gets further attached to C4b and forms C4b2a complex
• C3	Two chains α β	 110KDa 75KDa	- C3a: mediates inflammation - C3b: membrane binding protein and opsonin (**C3 is common both in classical and alternative pathway)**
Complement Proteins Participating In Alternative Pathway			
• Factor B (fB)	Single chain	93kDa	Binds C3b in a Mg dependent manner.
• Factor D (fD)	Single chain	26kDa	Cleaves fB at a single site exposing a serine protease domain in the fragment Bb
• Properdin	Glycoprotein made up of oligomers	Each monomer of 53 KDa	Binds and stabilises the C3 convertase complex
Complement Proteins Participating In Terminal Pathway			
• C5	Two chains	115kDa and 75kDa	C5a- peptide mediators of inflammation C5b- Membrane Attack Protein
• C6	Single chain plasma protein	120kDa	Binding of C6 stabilises C5b and exposes binding site for C7
• C7	Single chain	110kDa	Attachment of C7 causes release of complex into fluid phase
• C8	Three chains:α β γ	65kDa 65kDa 22kDa	βchain in C8 binds to C5b67 to form C5b-8 complex that is deeply embedded to the membrane. It leads to pore formation, thereby making the membrane leaky.
• C9	Single chain	69kDa	It binds to α chain of C8 in the complex leading to conformational changes and exacerbation of leakiness of the membrane.