Complement System

Complement refers to a system of factors in normal serum that are activated characteristically by antigen antibody interaction and subsequently mediate a number of biologically significant consequences.

Buchner(1889) was the first to observe that the bactericidal effect of serum was destroyed by heating at 55 ^{0}c for one hour.Pfieffer (1894) discovered that the *Cholera vibrios* were lysed when injected intraperitoneally into specifically immunized guinea pigs (bacteriolysis *in vivo* or Pfieffers phenomenon).Bordet (1895) established that immune bacteriolysis and hemolysis required two factors i.e.,the heat stable antibody and the heat labile factor which is called *alexine.*This term has been replaced by the present name-complement,coined by Ehrlich.(because this factor complemented the action of antibody.

General Properties of Complement

1. Complement ordinarily does not bind to free antigen or antibody, but only to antigen-antibody complex.
2. Complement is present in the sera of all mammals, birds, amphibians etc., and is a nonspecific serological reagent in that complement from one species can read with antibodies from other species.
3. Complement constitute about 5 % of normal serum proteins and is increased as a result of immunization.
4. Complement as a whole is heat labile,its cytolytic property undergo spontaneous denaturation slowly at room temperature and being destroyed in 30 minutes at 56 ^{0}c.The serum deprived of complement activity by heating at 56 ^{0}c for 30 minutes is said to be 'inactivated'.
5. The complement binding site is located on the Fc piece of the Ig molecule(CH2 domain onIgG,CH4 domain on IgM).All classes of Ig do not fix complement. Only IgM,IgG 3,1 and 2 fix complement, but not IgG4,IgA,IgD or IgE.
6. Complement fixation is influenced by the class of immunoglobulins and not by the nature of antigens.

Components of complement

There are nine central components of the cascade (C1 to C9), multiple activation products (such as C3a and C3b), regulators and inhibitors (e.g. Factor H and C4BP), proteases and newly assembled enzymes (e.g. C4b2a and Factor B), or effector molecule receptors (such as C3aR and C5aR).

Complement activation

A. Classical Pathway:

The chain of events in which C components react in a specific sequence following activationof C1 and typically culminate in immune cytolysis is known as the classical pathway.

1. The first step is the binding of C1 to the antigen antibody complex(represented by EA).
The recognition unit of C1 is Cq, which reacts with the Fc piece of bound IgM or IgG.C1q has six combining sites .Effective activation occurs only when C1q is attached to immunoglobulins by atleast two of its binding sites. Therefore one molecule of IgM 0r two molecules of IgG can initiate the process.C1q binding, in the presence of calcium ions leads to sequential activation of C1r and s.

2. Activated C1s is an esterase(C1s esterase),one molecule of which can cleave several molecules of C4.C4 is split into C4a (anaphylatoxin) and C4b which binds to cell membranes along with C1.

3.C14b in the presence of magnesium ions cleaves C2 into C2a,which remains linked to cell bound C4b,and C2b which is released into fluid phase .C4b2a has enzymatic activity and is referred to as the classical pathway C3 convertase.

4. C3 convertase splits C3 into two fragments-C3a (anaphyllatoxin) and C3b which remains cell bound along with C4b2a to form a trimolecular complex C4b2a3b which has enzymatic activity and is called C5 convertase.

5. **Membrane attack phase of complement activity** begins at this stage with-C5 convertase cleaving C5 into C5a(anaphyllatoxin which is released into the medium)**,** and C5b continues with the cascade.
C6 and C7 then join together and a trimolecular complex C567 is formed, part of which binds to the cell membrane and prepares it for lysis by C8 and C9-which join the reaction subsequently.
Most of C567 escape and serve to amplify the reaction by adsorbing on to unsensitised 'bystander cells' and rendering them susceptible to lysis by C8 and C9. The unbound C567 has chemotactic activity.
The mechanism of complement mediated cytolysis is the production of 'holes', approximately 100A^0A in diameter on the cell membrane. This disrupts the osmotic integrity of the membrane ,leading to the release of the cell contents.

Although the classical pathway is generally activated by the antigen antibody complexes or aggregated immunoglobulin, activation may also be due to other stimuli, such as DNA, C-reactive protein, trypsin like enzymes or some retroviruses.

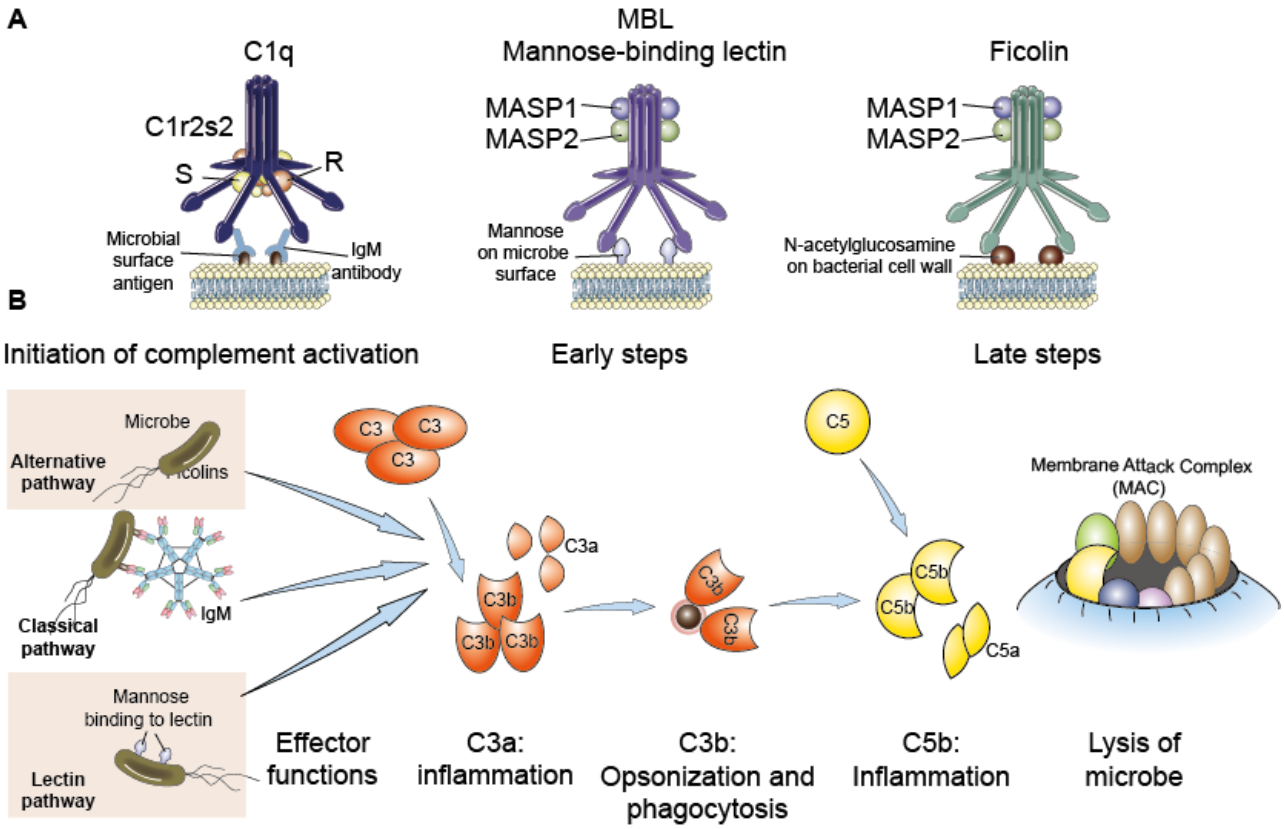

Figure. C1, Classical, mannose-binding lectin, and Alternate (A).
Three Pathways of complement activation (B)
https://www.c-di.com/complement-system.htm

B. Alternate Complement Pathway:

The central process in the complement cascade is activation of C3.The activation of C3 **without prior participation of C142 (as in case of classical pathway)** is known as the **alternate pathway**.

The first step in alternative pathway is the binding of C3b to an activator. Bound C3b which is protected from inactivation by serum proteins factors H and I, interacts with serum protein –Factor B(C3 proactivator) to form a magnesium dependant complex C3bB.'This complex is cleaved by an other serum protein factor D (C3 proactivator convertase into two fragments Ba and Bb. Fragment Ba is released into the medium.

Fragment Bb remains bound to C3b to form esterase C3b,Bb complex (alternative pathway C3 convertase).

The properdin (Factor P) stabilizes the C3 convertase, which hydrolyzes C3 leading to further steps in the cascade, as in the classical pathway.

C. Mannose-binding Lectin (MBL) Pathway

Some bacteria can activate the complement system without having antibodies and endotoxin. This occurs through the MBL pathway which is activated when circulating lectin (MBL) binds to mannose residues on glycoproteins or carbohydrates on the surface of microorganisms. Microorganisms inducing the MBL pathway are bacteria, such as *Salmonella, Listeria,* and *Neisseria* strains, some fungi, and some viruses including HIV-1. **MBL is an acute-phase protein** and its concentration increases during inflammation. The lectin recognizes and binds the carbohydrate of the target cell which then activates complements.

REGULATION OF C ACTIVATION: Unchecked complement activity can cause not only exhaustion of the complement system but also serious damage to the tissues. Several inbuilt control mechanisms regulate the complement cascade at different steps.

Control of Complement System Activation

The cascade reaction should be controlled one. This control is exerted by proteins which activate key components either by binding to the active sites of the enzymes or by further cleaving active proteins to give inactive products (e.g., cleavage of C3b by factor-I).

C1 Inhibitor	Glycoprotein present in normal serum inhibits C1r and C1s binding to active sites
Factor I	Cleaves C3b and C4b
Factor H	Prevents factor B from binding to C3b. Dissociates Bb from active complex C3bBb makes the C3b susceptible to cleavage by factor I.
C4 Binding Protein	Binds tightly to C4b and enhances in degradation
Factor P/ Properdin	Stabilizes the alternative pathway C3 convertase when it is bound to activating surfaces
Cobra venom	Is equivalent to C3, C5 convertase (i.e., Complex formed by (COVF+B Factor). And is resistant to the action of inactivators of the alternative pathway

Biological Effects of Complement

1.Complement mediates immunological membrane damage-results into bacteriolysis, cytolysis.
2. Neutralization of some viruses requires the participation of complement.
   e.g., Herpes virus neutralization by IgM requires binding of C1,C4 and C3.
3. Facilitates opsonization.
4. Complement participate in type-II, type-III hypersensitivity reactions.
   Type II - Cytotoxic hypersensitivity
   - Leading to destruction of erythrocytes following Incompatible blood transfusion.
   Type III – Immune complex hypersensitivity. Diseases e.g., serum sickness & Arthus reaction.
5. Complement plays important role in pathogenesis of autoimmune disease and immune adherence.
6. C3 & C6 participate in coagulation process.

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