Complements

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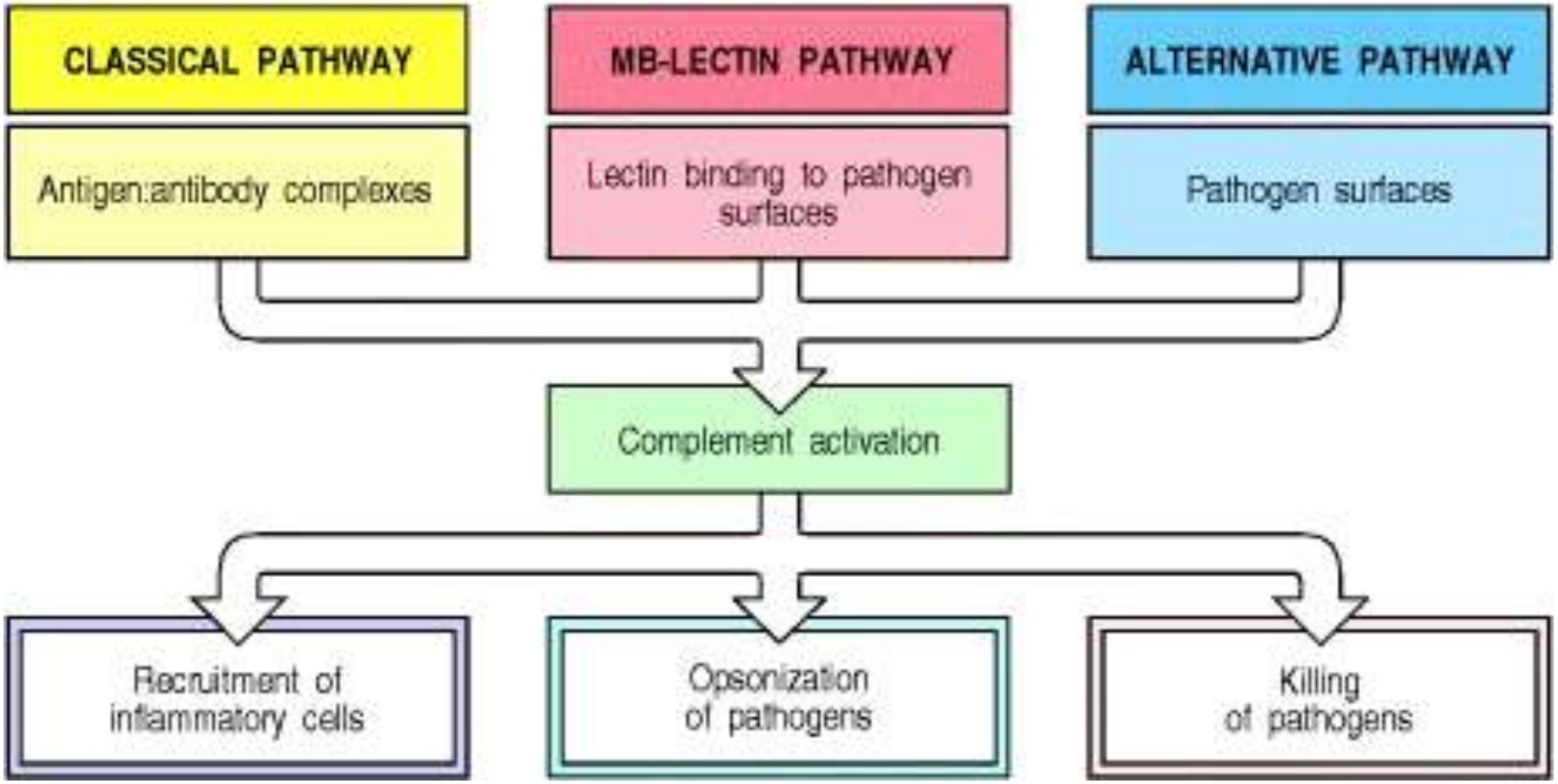
- The complement system consists of a number of small proteins found in the blood, generally synthesized by the liver, and normally circulating as inactive precursors (pro-proteins).
- When stimulated by one of several triggers, activation cascade is started and lead to functional effects.
- Over 25 proteins and protein fragments make up the complement system. They account for about 5% of the globulin fraction of blood serum.

- Complement was discovered many years ago as to ‘complement’ the antibacterial activity of antibody, hence the name.
- Although first discovered as an effector arm of the antibody response (Adaptive), complement can also be activated early in infection in the absence of antibodies (innate) .

- In the case of the complement system, the precursor are widely distributed throughout body fluids and tissues without adverse effect. At sites of infection, however, they are activated locally and trigger a series of potent inflammatory events
- There are three distinct pathways through which complement can be activated on pathogen surfaces. These pathways depend on different molecules for their initiation, but they converge to generate the same set of effector molecules


CLASSICAL PATHWAY
MB-LECTIN PATHWAY
ALTERNATIVE PATHWAY
Antigen:antibody complexes
Lectin binding to pathogen surfaces
Pathogen surfaces
Complement activation
Recruitment of inflammatory cells
Opsonization of pathogens
Killing of pathogens

Complement pathways activation

- Classical pathway; recognize antibody binding microbe as viruses or bacteria (IGG1, IGG3, IGA and IGM) it is arm of humoral immunity
- Alternative; recognize LPS or endotoxins of microbe (part of innate response)
- Lectin pathway. The lectin is a protein bind carbohydrates on microbe (mannose)

Complements

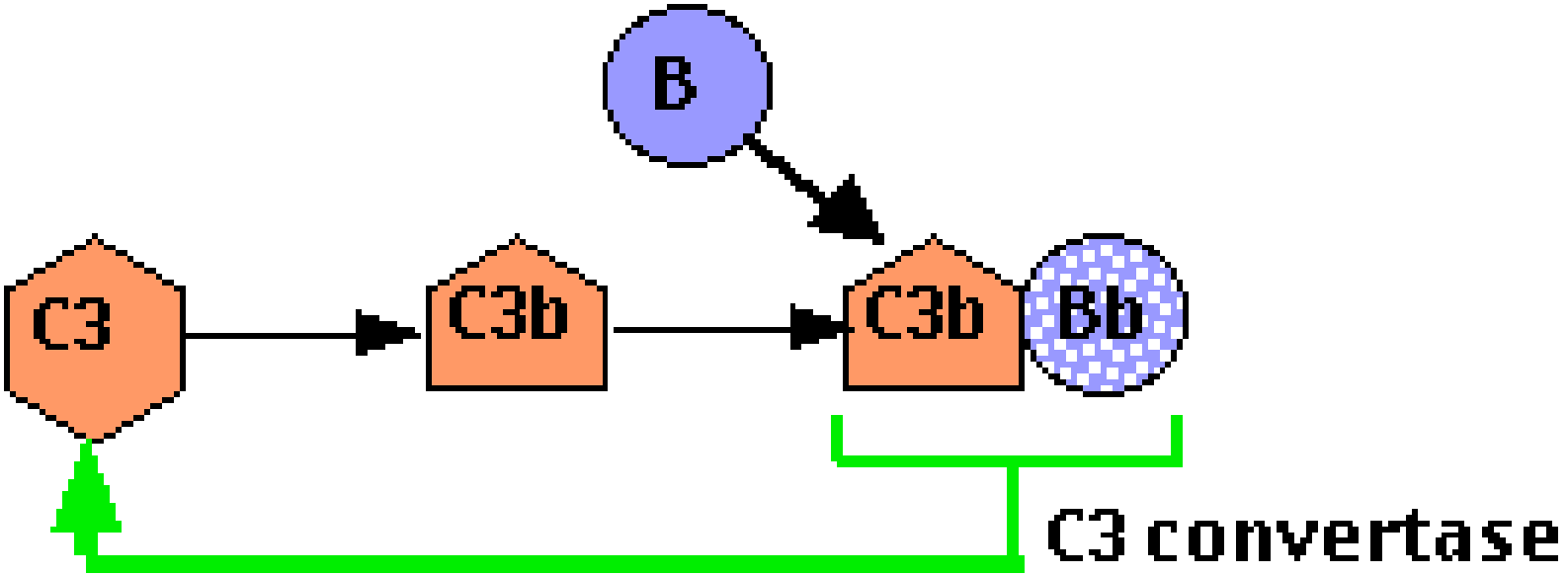
- The **classical pathway**, so called because it was discovered first, uses a plasma protein called C1q to detect antibodies bound to the surface of a microbe or other structure. Once C1q binds to the Fc portion of the antibodies, two associated serine proteases, called C1r and C1s, become active and initiate a proteolytic cascade involving other complement proteins (C2, C4) to make C3 convertase. The classical pathway is one of the major effector mechanisms of the humoral arm of adaptive immune responses.

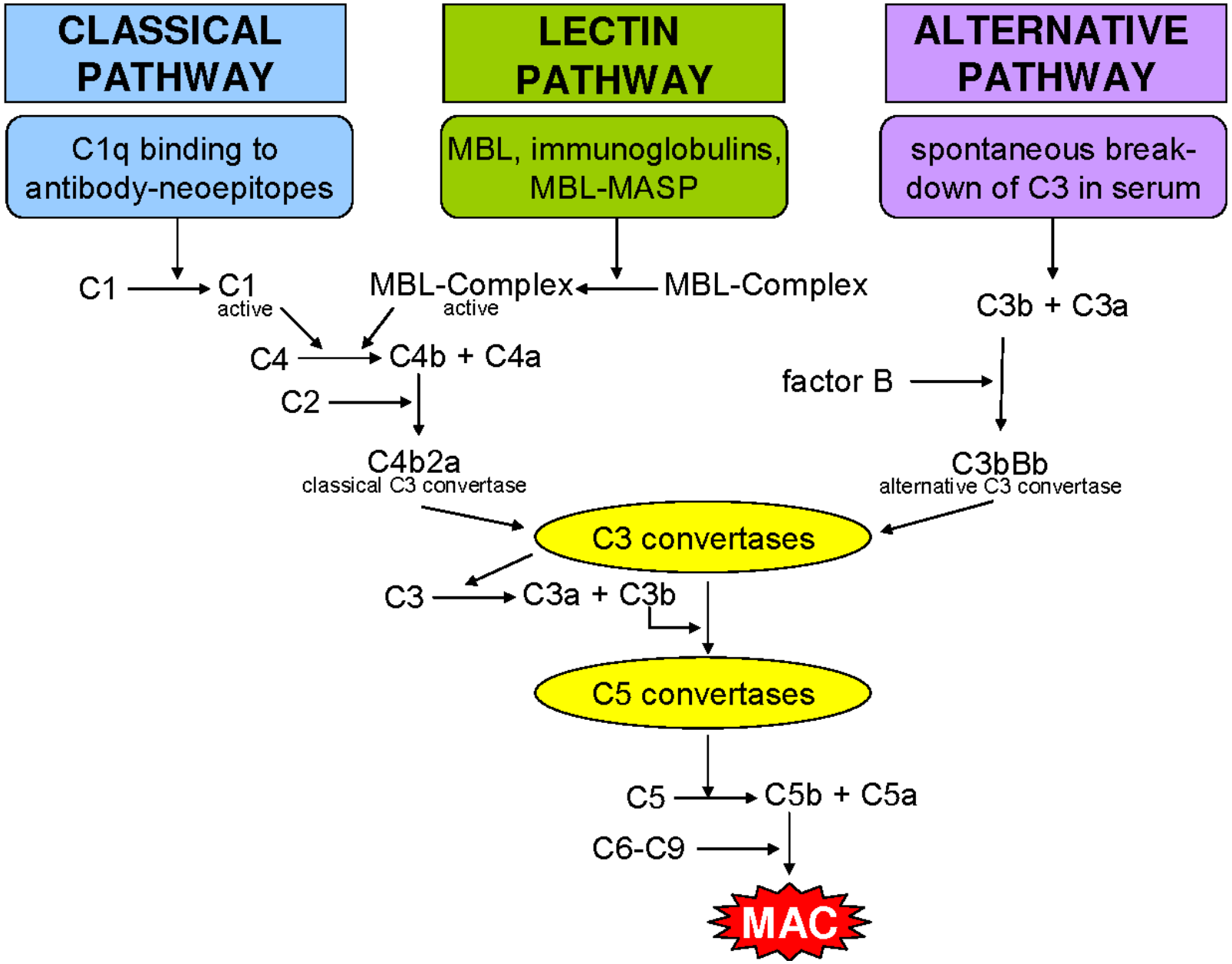
Classical pathway

- C1 exists in blood serum as a molecular complex containing:
  - C1q
  - C1r
  - C1s
- The IGM and IGG that bound by antigen, contain a binding site for C1q. (A single molecule of IgM is enough to initiate the pathway. IgG is far less efficient, requiring many molecules to do so.)
- Binding of C1q activates **C1s** and **C1r**.
- Activated C1s (a protease) cleaves two serum proteins:
  - **C4** is cleaved into a large fragment
    - **C4b**, which binds covalently to surface of antigen (opsonisation) and
    - **C4a,** smaller, inactive, which diffuses away.
  - **C2** is cleaved into
    - **C2b**, which binds to a site on **C4b**,
    - **C2a** a smaller, inactive, fragment of which diffuses away.
  - The complex of C4b2a is called "**C3 convertase**" because it catalyzes the cleavage of **C3**.

Complement

- **The alternative pathway**, is triggered when a complement protein called C3 simultaneously degraded to C3b that recognizes certain microbial surface structures, such as bacterial LPS.
- C3b undergoes its post-cleavage conformational change, a binding site for a plasma protein called Factor B is also exposed. Factor B then binds to the C3b protein that is now covalently tethered to the surface of a microbial or host cell. Bound factor B is in turn cleaved by a plasma serine protease called Factor D, releasing a small fragment called Ba and generating a larger fragment called Bb that remains attached to C3b.
- The C3bBb complex is the alternative pathway **C3 convertase**,


B
C3
C3b
C3b
Bb
C3 convertase


CLASSICAL PATHWAY
LECTIN PATHWAY
ALTERNATIVE PATHWAY
C1q binding to antibody-neoepitopes
MBL, immunoglobulins, MBL-MASP
spontaneous break-down of C3 in serum
C1
C1 active
MBL-Complex active
MBL-Complex
C3b + C3a
C4
C4b + C4a
C2
factor B
C4b2a
classical C3 convertase
C3bBb
alternative C3 convertase
C3 convertases
C3
C3a + C3b
C5 convertases
C5
C5b + C5a
C6-C9
MAC

Lectin pathway

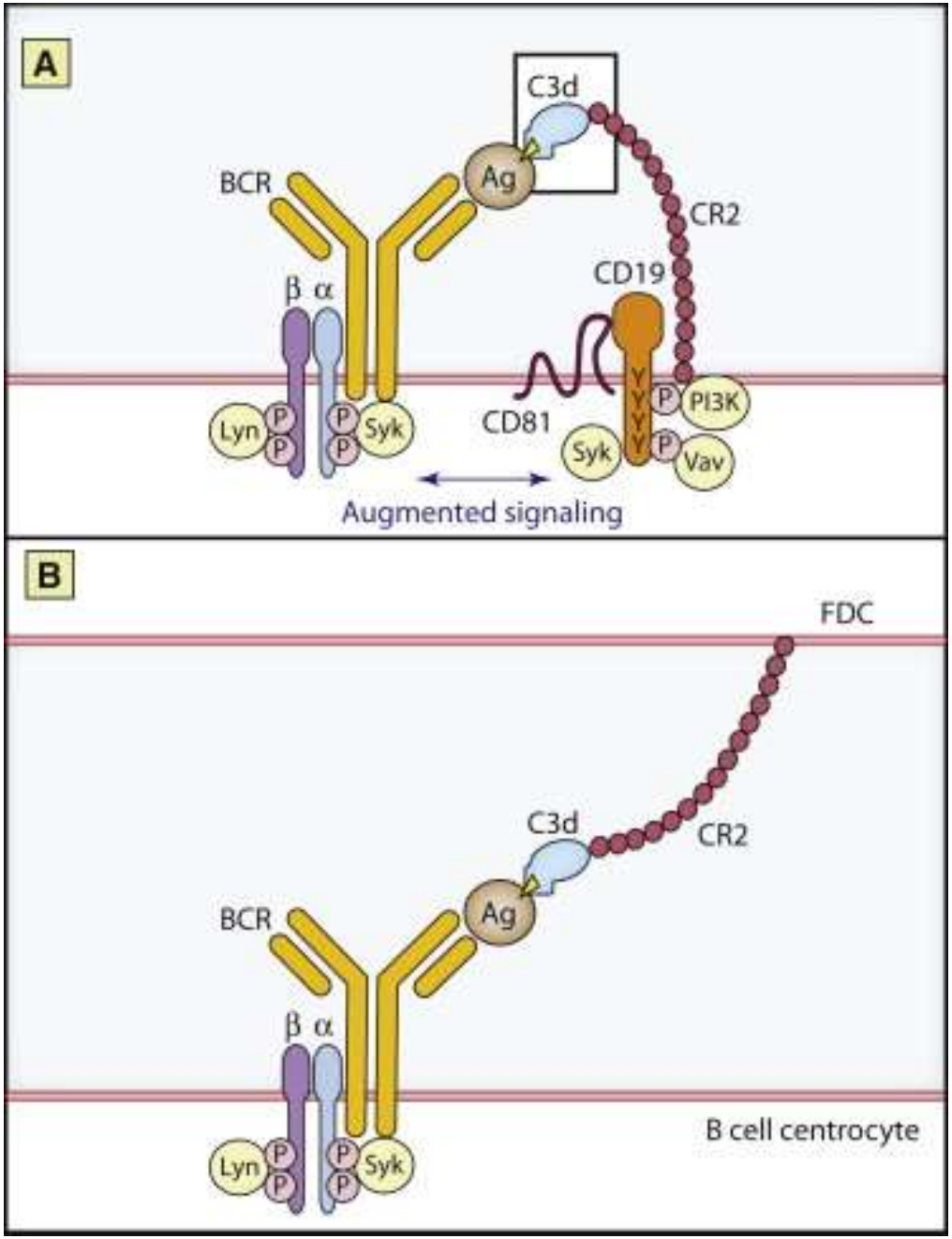
- Lectin pathway. The lectin (proteins macromolecules in blood that are highly specific to mannose on pathogen) pathway is homologous to the classical pathway, but with mannose-binding lectin (MBL) instead of C1q, and in the absence of antibody
- This pathway is activated by binding of lectin to mannose residues on the pathogen surface, which can then split C4 into C4a and C4b and C2 into C2a and C2b the rest pathway is similar to classical

C3

- Recognition of microbes by any of the three complement pathways results in sequential recruitment and assembly of additional complement proteins
- **C3** is the most abundant protein of the complement system. Because of its abundance and its **ability to activate itself** (as described later), it greatly magnifies the response.
  - **C3 convertase** cuts **C3** into major fragments:
    - **C3b**, which binds covalently to glycoproteins scattered across the microbial cell surface. Macrophages and neutrophils have receptors for **C3b** and can bind the C3b-coated cell or particle preparatory to phagocytosis. This effect qualifies C3b as an **opsonin**.
    - **C3a** This small fragment is released into the surrounding fluids. It can bind to receptors on basophils and mast cells triggering them to release their vasoactive contents (e.g., histamine). Because of the role of these materials in anaphylaxis and inflammation, C3a is called an **anaphylatoxins**.
    - C3d:

C3d link innate to humoral immunity

- antigen gets “tagged” with the appropriate C3d product via the classical or lectin complement pathways.
- C3d bind both the antigen receptor on B cell (IGM) and complement receptor CR2 (CD21)on B cells, which forms with CD19 a coreceptor on B cells during antigen-induced activation regulating humoral immunity and enhancing signaling through the B cell Ag receptor
- CR2 on B cell enhance entrance and infection of B cells by by Epstein–Barr virus

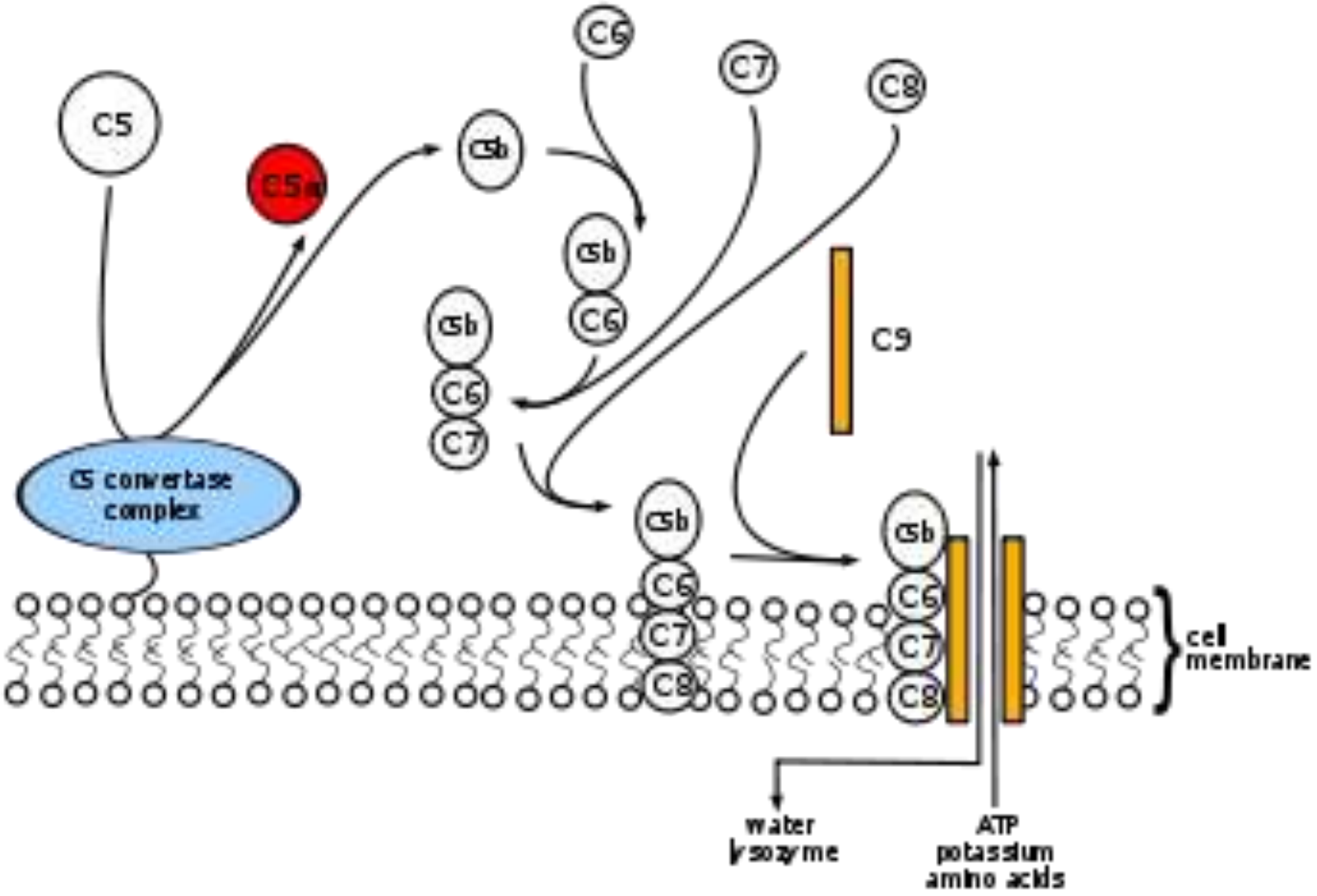

A
C3d
Ag
BCR
CR2
CD19
β α
CD81
Lyn
P
Syk
PI3K
Vav
Augmented signaling
B
FDC
C3d
CR2
Ag
BCR
B cell centrocyte

Membrane attack complex (MAC

-**C5 convertase formed by joining C3 convertase to C3b**

- Cleavage of C5 by the (C3bBb3b)and other complements (C4b•2a.3b)(which is thus a "C5 convertase".) produces:
- **C5a**, which is released into the fluid surroundings where it
  - is a potent anaphylatoxin
  - is a chemotactic attractant
- **C5b**, which serves as the anchor for the assembly of a single molecule each of
  - C6;
  - C7, and
  - C8.

- **The Membrane Attack Complex (MAC)**
  - The resulting complex **C5b•6•7•8** guides the polymerization of as many as 18 molecules of **C9** into a tube inserted into the lipid bilayer of the plasma membrane. This tube forms a channel allowing the passage of ions and small molecules. Water enters the cell by osmosis and the cell lyses.


C5
C5a
C5b
C6
C7
C8
C9
C5b
C6
C5b
C6
C7
C5 convertase complex
C5b
C6
C7
C8
C5b
C6
C7
C8
cel membrane
water
lysozyme
ATP
potassium
amino acids

Summary of complement functions

- **Opsonization** by C3b, C4b and C5b targets foreign particles for phagocytosis.
- **Chemotaxis** by C5a, C4a and C3a attracts phagocytic cells to the site of damage.
- This is aided by the **increased permeability (anaphylatoxins)** they cause smooth muscle contraction, vasodilation, histamine release from mast cells, and enhanced vascular permeability.mediated by **C3a, C5a, C4a**.
- C3b ; are also important for **solubilizing** antigen-antibody complexes and elimination from the body (by binding the immune complex to CR1 on erythrocyte). otherwise aggregation of the complexes lead to immune complex disorder (SLE, diabetes mellitus, RA)
- **Lysis** of target cells (C5b-9).
- Promoting B cell activation and antibody formation. Breakdown of C3b generates a fragment (C3d) that binds to antigens enhancing their uptake by B cells.

Complement receptors

- Type 1 receptor(CR1); bind C3b, and C4b. Expressed in erythrocytes, macrophages, neutrophil
  - Do opsonization and Induce phagocytosis (with antibody)
  - Help to remove immune complexes from blood to liver and spleen (erythrocytes)
- Type 2 receptor (CR2), bind C3d, and, expressed on B lymphocytes and DC
  - With other proteins enhance B cell response to antigen
  - Receptor for epstein barr virus on B cells
- Type 3 and 4 receptors on phagocytes
  - bind opsonizing C3b and lead to phagocytosis. Found on macrophages and neutrophils

Regulation of complement activity

- The explosive potential of the complement system requires that it be kept under tight control. At least 12 proteins are known that do this. Three examples:
- **Factor H and Decay-accelerating factor (DAC),** removes Bb from the alternative pathway C3 convertase.
- **Factor I** inactivates C3b.
- **C1 inhibitor** (**C1INH**) binds to sites on activated C1r and C1s shutting down their proteolytic activity.
- **CD59** on normal tissue cells which inhibit association of C9 with C5b-8

Disorders of the complement system

- With so many proteins involved, it is not surprising that inherited deficiencies of one or another are sometimes encountered in humans. Four examples:
  - **C3**. An inherited deficiency of C3 predisposes the person to frequent bouts of bacterial infections mainly gram negative bacteria.
  - **C2 , C1, C3 or C4**. immune complex disorders are the main problem with a deficiency of C2 , C1 or C4. This emphasizes the important role of the complement system in clearing away antigen-antibody complexes. A deficiency of C2 is frequently found in patients with lupus erythematosus (SLE).
  - **C9**. most people who cannot make C9 have no problem with bacterial infections. Laboratory studies suggest that the **C5b•6•7•8** complex by itself is able to lyze bacteria although not as efficiently as C9.
  - **C1INH**. A deficiency of C1INH produces **hereditary angioedema**. The massive release of anaphylatoxins (C3a, C5a) may cause dangerous swelling (edema) of the airways, as well as of the skin and intestine.
  - **CD59** deficiency in its expression lead to inadequate control of MAC assembly results in intravascular red cell lyses called **paroxysmal nocturnal haemoglobinuria**