European Journal of Internal Medicine 24 (2013) 496–502

Contents lists available at ScienceDirect

European Journal of Internal Medicine

journal homepage: www.elsevier.com/locate/ejim

Review article

Complement activation in diseases presenting with thrombotic microangiopathy

Seppo Meri *

Department of Bacteriology and Immunology, Haartman Institute, PO Box 21, FI-00014 University of Helsinki, Helsinki, Finland

ARTICLE INFO

Available online 4 June 2013

Keywords:
Thrombosis
Microangiopathy
Complement regulation
Hemolytic uremic syndrome
Complement factor H
CD46

ABSTRACT

The complement system contains a great deal of biological "energy". This is demonstrated by the atypical hemolytic uremic syndrome (aHUS), which is a thrombotic microangiopathy (TMA) characterized by endothelial and blood cell damage and thrombotic vascular occlusions. Kidneys and often also other organs (brain, lungs and gastrointestinal tract) are affected. A principal pathophysiological feature in aHUS is a complement attack against endothelial cells and blood cells. This leads to platelet activation and aggregation, hemolysis, prothrombotic and inflammatory changes. The attacks can be triggered by infections, pregnancy, drugs or trauma. Complement-mediated aHUS is distinct from bacterial shiga-toxin (produced e.g. by *E. coli* O:157 or O:104 serotypes) induced "typical" HUS, thrombotic thrombocytopenic purpura (TTP) associated with ADAMTS13 (an adamalysin enzyme) dysfunction and from a recently described disease related to mutations in intracellular diacylglycerol kinase ε (DGKE). Mutations in proteins that regulate complement (factor H, factor I, MCP/CD46, thrombomodulin) or promote (C3, factor B) amplification of its alternative pathway or anti-factor H antibodies predispose to aHUS. The fundamental defect in aHUS is an excessive complement attack against cellular surfaces. This can be due to 1) an inability to regulate complement on self cell surfaces, 2) hyperactive C3 convertases or 3) complement activation and coagulation promoting changes on cell surfaces. The most common genetic cause is in factor H, where aHUS mutations disrupt its ability to recognize protective polyanions on surfaces where C3b has become attached. Most TMAs are thus characterized by misdirected complement activation affecting endothelial cell and platelet integrity.

1. Introduction

Thrombotic microangiopathies (TMAs) are diseases characterized by a tendency to develop vascular thrombi, endothelial cell damage, thrombocytopenia and hemolysis. They are systemic diseases with potential involvement of several organs, including the kidneys, brain, muscles, gastrointestinal system, skin and lungs. Common to all these diseases is a prothrombotic condition, which in many cases is due an underlying disturbance in the complement system. The complement and the coagulation systems have much in common, including surface-directed activation in cases where activation is not actively inhibited. Under normal circumstances, intact vascular and cellular surfaces are not only nonthrombogenic but they also resist complement activation. To a large extent this is due to a protective negatively charged layer of polyanionic molecules on viable, living cells. Molecularly they are composed of sulphated glycosaminoglycans, like heparan sulphate on endothelial cells, terminal sialic acids with hydroxyl groups and phospholipids with negatively charged head groups. This review analyses mechanisms, whereby complement can discriminate between nonactivating ("self") and activating ("nonself") surfaces, and why protection of self cells can fails and lead to severe diseases, like the atypical hemolytic uremic syndrome (aHUS).

2. Complement activation and regulation

The main physiological functions of complement are to initiate and mediate inflammatory reactions, participate in the clearance of endogenous waste products, label invading microbes for removal by phagocytes (opsonization) and to kill microbes with the membrane attack complex (MAC) [1]. Two main complement pathways, the classical and the alternative ones, are shown in Fig. 1. In the classical pathway, the C1 complex, via its C1q part binds to antigen-bound antibodies or to other targets, like bacterial surfaces, and activates, via C1r and C1s, the next components in the cascade, C2 and C4. Activation products form a C3 convertase enzyme C4bC2a that has proteolytic activity towards C3. In the alternative pathway (AP), an equivalent C3 convertase, C3bBb, is generated from activated C3 (C3b) and B (Bb). C3b-bound B is activated by factor D. C3b can be generated by several different mechanisms: (i) by a so called initiating C3 convertase that contains a spontaneously generated C3b-like C3 and Bb, by the (ii) classical or (iii) alternative pathway C3 convertases or by some (iv) proteolytic

Abbreviations: AP, alternative pathway; aHUS, atypical hemolytic syndrome; ADAMTS13, a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13; AP, alternative pathway; CCP, complement control protein domain; DDD, dense deposit disease; DGKE, diacylglycerol kinase ε; FH, factor H; PNH, paroxysmal nocturnal hemoglobinuria; STEC, Shiga-toxin producing *E. coli*; TMA, thrombotic microangiopathy; TTP, thrombotic thrombocytopenic purpura.

* Tel.: +358 50 5812462.
E-mail address: seppo.meri@helsinki.fi.

0953-6205/$ – see front matter © 2013 European Federation of Internal Medicine. Published by Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.ejim.2013.05.009

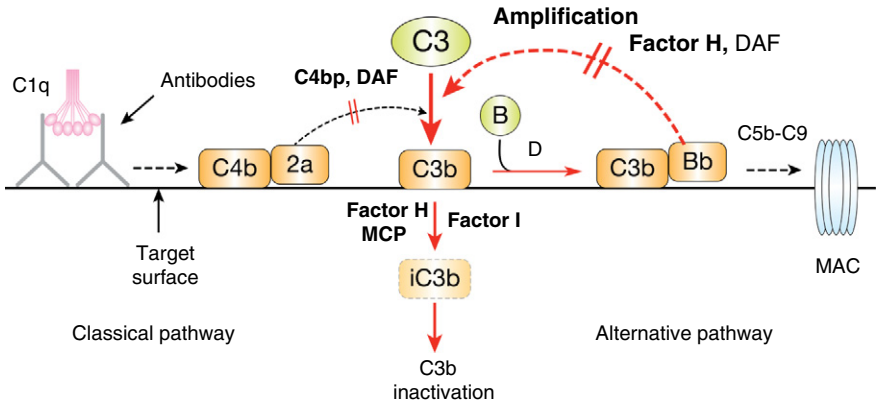

Fig. 1. The main complement activation pathways. C3 is activated by either the classical (C4b2a) or the alternative (C3bBb) pathway C3 convertase. The alternative pathway has an in-built amplification system, where C3 activation can promote the generation of new C3bBb convertases. Factor H is controlling the C3bBb activity and promoting C3b inactivation.

enzymes, like thrombin and plasmin. In addition, there is a third pathway, the lectin pathway, where a number of sensor molecules (mannan binding lectin, MBL and ficolins) recognize unmodified (mannose) or modified (acetylated) sugar moieties on surfaces. This leads to C2 and C4 activation similarly as in the classical pathway, but with different enzymes, MBL-associated serine proteases, or MASPs (primarily by MASP-2) [2].

The same enzymes that can cleave C3 (C4b2a and C3bBb) can also activate C5 to generate the potent anaphylatoxin C5a and C5b that can initiate formation of the membrane attack complex (MAC). C5a is the central molecule in generating inflammation. It can attract and activate leukocytes and platelets, increase permeability of blood vessels and contract smooth muscle cells. MAC is composed of C5b, C6, C7, C8 and multiple C9 molecules. It can generate holes on target membranes (red blood cells, bacterial outer membranes) and activate intracellular processes on nucleated cells via calcium-influx. The latter may lead to membrane changes, increased prothrombotic properties and, in extreme cases, to cell death. On human cells, however, the MAC attack is efficiently inhibited by protectin (CD59) that prevents C5b-8 catalyzed polymerization of C9 [3].

Since C3 activation is a major step in the complement cascade it needs to be strictly controlled. The classical pathway C3 convertase, C4b2a, is regulated by a fluid phase inhibitor C4b binding protein (C4bp) [4,5]. In the alternative pathway, because of its tendency for positive feedback activation, i.e. amplification of its own activation, various control mechanisms are critical. Because C3b, the activation product of C3 cleavage by C3bBb, can generate new C3 convertases that again generate new C3b molecules, the alternative pathway (AP) efficiently coats unprotected target surfaces (nonself) with C3b molecules. Covalent attachment of large amounts of C3b can occur within a few minutes. This is inhibited both in the fluid phase and on surfaces by factor H (beta-1 H globulin, FH) (Fig. 1).

Cell surfaces are protected against complement attack at the C3 level by specific membrane inhibitors. They include complement receptor 1 (CR1, CD35), the membrane cofactor protein (MCP, CD46), decay accelerating factor (DAF, CD55) and CRIg [6–8]. CR1 is a very long complement receptor and regulator containing 30 so called complement control protein domains (CCPs; also called short consensus repeat domains, SCRs), units of approximately 60 amino acids each, folded into globular structures by two internal disulphide bridges. Because of this CR1 is thought to act as a catcher and transporter of C3b or C4b containing immune complexes and microbes rather than as an endogenous inhibitor on cells (erythrocytes, leukocytes, podocytes) that carry it [9]. By promoting the inactivation of C3b to iC3b CR1 would help the release of the C3b binding complexes to macrophages in the spleen and liver that carry the iC3b receptor, CR3 (the CD11b/18 integrin) [10]. CRIg is an immunoglobulin domain containing receptor for C3 activation products that is expressed selectively by the liver Kupffer cells. Like CR1 it is involved in the uptake of microbes and particles coated with C3b (and to some extent iC3b) [7].

CD46 is an important and widely distributed complement inhibitor with four extracellular domains [11]. The two most membrane proximal domains (CCP3 and 4) have been shown to have the C3b binding sites [12]. With regard to the development of aHUS this is important because MCP-mutations linked to aHUS (if not leading to loss of expression) are often located on one of these two domains [13]. Key factors why mutations in CD46/MCP predispose to aHUS are that MCP (i) is expressed on blood end endothelial cell surfaces and (ii) acts, similarly to factor H, as a cofactor for C3b (and additionally for C4b) cleavage. Thus, the activity is restricted to cell surfaces, and mutations that compromise its protective activity increase the susceptibility of the cells to complement attack.

3. The dual role of complement in inflammation and disease

Opsonic and phlogistic activities of complement activation can have a positive or negative impact on our health. Because of its clean-up functions in helping waste removal by the reticuloendothelial system the proper activity of the complement system is important for body homeostasis [14]. Complement deficiencies predispose to infections, particularly to those caused by meningococci or pneumococci, and to systemic immunoinflammatory diseases, where the removal of cell debris is compromised [15] (Table 1). An example of the latter is systemic lupus erythematosus, where some forms are related to deficiencies in the early classical pathway (C1q, C2 and C4). These components are involved in the removal of immune aggregates, chromatin and membrane fragments from injured, ischemic,

Table 1
Examples of diseases associated with complement activation. The diseases are divided into those, where complement dysfunction has a primary role or an important role in the generation of tissue injury. Also, a subdivision to hematological and nonhematological diseases is provided.

Hematological	Nonhematological
Complement has a primary role	
Paroxysmal nocturnal hemoglobinuria (PNH)	Dense deposit disease(DDD)
Hemolytic uremic syndrome (aHUS and STEC-HUS)	Age-related macular degeneration (AMD)
Secondary tissue damage and inflammation caused by complement	
Catastrophic antiphospholipid antibody syndrome (CAPS)	Myocardial infarction
Cold agglutinin disease (CAD)	Sepsis, ARDS
Autoimmune hemolytic anemia (AIHA)	Systemic lupus erythematosus
Thrombotic thrombocytopenic purpura (TTP)	HELLP syndrome
	Rheumatoid arthritis
	Antibody-mediated rejection