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Cross-talk between the complement and the kinin system in vascular permeability

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Abstract

The endothelium is a continuous physical barrier that regulates coagulation and selective passage of soluble molecules and circulating cells through the vessel wall into the tissue. Due to its anatomic localization, the endothelium may establish contact with components of the complement, the kinin and the coagulation systems which are the main, though not exclusive, inducers of vascular leakage. Although the complement and the kinin systems may act independently, increasing evidence suggest that there is a crosstalk that involve different components of both systems. Activation is required for the function of the two systems which are involved in pathological conditions such as hereditary and acquired angioedema (AE) and vasculitidis. The aim of this review is to discuss the contribution of complement and kinin systems to vascular leakage and the cross-talk between the two systems in the development of AE. This clinical condition is characterized by episodic and recurrent local edema of subcutaneous and submucosal tissues and is due to inherited or acquired C1-INH deficiency. Although the pathogenesis of the swelling in patients with AE was originally thought to be mediated by C2, ample evidence indicate bradykinin (BK) as the most effective mediator even though the possibility that both the complement and the kinin-forming systems may contribute to the edema has not been completely excluded. BK induces endothelial leakage interacting with B2 receptors but other molecules may be involved in the onset and maintenance of AE. In this review we shall discuss the role of B1 receptors and gC1qR/p33 in addition to that of B2 receptors in the onset of AE attacks and the importance of these receptors as new possible molecular targets for therapy.

Keywords

angioedema; complement; endothelium; kinins; C1-inhibitor; gC1qR/p33

1. Introduction

The vascular tree is covered on the inner surface by a continuous layer of endothelial cells which form a physical barrier between the circulating blood and the extravascular tissue

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preventing potential damage which may derive from undesired blood outflow. Besides playing a critical role in physically separating the intravascular from the extravascular sides of the vascular vessels, the endothelium exhibits other important functions to maintain vascular homeostasis. By expressing heparin sulphate, and by releasing thrombomodulin and tissue factor inhibitor, endothelial cells (ECs) expose an anti-thrombotic surface on the luminal side which controls blood flow preventing blood clot. The endothelium can also regulate the vascular tone contributing to induce changes in the blood flow in response to tissue demands. Several substance can be released by ECs which either promote vasodilation, such as nitric oxide and prostacyclin PGI₂, or induce vasoconstriction, as is the case of endothelin-1 and platelet-activating factor [1].

Another important function of the endothelium is to control passage of molecules and cells to the extravascular sites where they are needed for nutritional and defence purposes. This is made possible by its structural organization as a monolayer of cells joined together by tight junctions, a structure observed in the microcirculation of almost all tissues and organs with only a few exceptions.

Perturbation of ECs caused by physicochemical stimuli induces cell activation and has functional consequences resulting in cell egression into the extravascular sites and increased vascular permeability. These changes are usually associated with inflammatory processes which are responsible for systemic manifestations, such as sepsis or generalized vasculitides, or localized reactions. However, vascular leakage which manifests as non inflammatory edema may also occur in some pathological conditions in the absence of inflammation.

The soluble systems which are mainly, though not exclusively, involved in the induction of increased vascular permeability include the complement (C) and the kinin systems, which may have a direct permeabilizing effect through the release of biologically active products or may activate other cells which in turn release vasoactive molecules. Although the two systems may act independently to induce vascular leakage, increasing evidence suggests that there is a cross-talk which involves different components of these systems. Both systems require activation to function and are involved in triggering or exacerbating pathological conditions such as hereditary and acquired AE and vasculitidis.

The focus of this review is to discuss the contribution of C and kinin systems to vascular leakage and to highlight the cross-talk between the two systems in the development of AE.

2. Contribution of the complement system to the vascular leakage

The C system is an important component of innate immunity involved in host defence against microorganisms, clearance of immune complexes and removal of apoptotic cells and acts either alone or more often in collaboration with other components of both innate and acquired immune system. To accomplish these functions, the system must be activated to release biologically active products which may directly neutralize the target or favour the participation of other components of the immune system to complete the protective action. Complement activation may occur on the cell membrane and/or in the fluid phase and shares with the activation of the kinin system the characteristic feature to proceed in a cascade fashion leading to the release of a progressively increased number of biologically active products. These products are formed at various steps of the C sequence initiated by different components which trigger distinct activation pathways.

ECs are continuously exposed to C components present in circulating blood or at extravascular sites and establish with the C system multiple interactions which have important functional consequences. To begin with, the endothelium is an important source of C components and regulators at tissue level [2] and together with macrophages,

fibroblasts, and several other cell types, contributes to the pool of circulating plasma components which are not synthesized in the liver as is the case of C7 [3]. This terminal C component is not only secreted by ECs but also expressed on the cell surface as a means of protection against the damaging effect of SC5b-9 [4]. On the other hand, ECs represent a potential target of biologically active products which are formed as a result of C activation on the cell surface or in the fluid phase. Complement-fixing antibodies reacting with antigens expressed or bound to ECs can bind C1q and trigger the C sequence through the classical pathway. These antibodies can be found in patients with autoimmune diseases associated with vasculitides and in patients undergoing hyperacute graft rejection. Anti-phospholipid antibodies interacting with β 2GPI which binds to receptors expressed on ECs represent another example of C-fixing antibodies [5,6]. The lectin pathway can also be activated on cultured ECs exposed to oxidative stress [7] which leads to the exposure of sugar moieties -such as mannose, fucose or N-acetylglucosamine- usually expressed on the surface of bacterial pathogens or altered cells [8]. MBL binding and C activation through the lectin pathway has also been documented *in vivo* on rat myocardium [9] and mouse kidney [10] undergoing ischemia-reperfusion injury.

Although the two pathways use different components in the early phase of the activation sequences including C1r and C1s, for the classical pathway, and MASPs, for the lectin pathway, they share C4 and C2 to form the C3 convertase which in turn activates C3 resulting in the split of this molecule into C3a and C3b [11].

Isolated C1q can interact with ECs by directly binding to receptors which recognize the collagen portion (cC1qR) [12] or the globular head (gC1qR) [13]. Since C1q tends to form a loose complex with C1r and C1s in both plasma and extravascular fluids, free C1q is made available mostly after activation of the classical pathway which results in the release of C1q from the complex following the dissociation of C1r and C1s by C1 inhibitor. Binding of C1q to C1qR stimulates ECs to express adhesion molecules [14] and to release the chemotactic factors IL-8, MCP-1 and IL-6 [15]. However, evidence that C1q may also promote EC leakage is lacking. C1s is the only early component which was previously shown to induce vascular permeability following intradermal injection into guinea pig [16]. This effect was later attributed to a split product of C2 generated by activated C1s [17]. However, subsequent studies failed to identify a C2-derived fragment with properties [18].

Cleavage of C3 and C5 by the C3 and C5 convertases respectively results in the release of the small peptides C3a and C5a which interact with receptors expressed on ECs [19,20]. Both C3a and C5a induce cytoskeletal changes on ECs [21] and up-regulate IL-8, IL-1 β and RANTES mRNA [20], but the two anaphylatoxins differ because C5a causes a decrease in IL-6 mRNA [20] and is chemotactic for dermal microvascular ECs [21].

Intradermal injection of C3a purified from an inflammatory exudate into rat skin was reported to enhance vascular permeability [22]. The involvement of C3 in vascular leakage was also confirmed in an *in vivo* model of immune complex-mediated disease in mice, as indicated by the marked decrease in the vascular permeability induced by immune complexes in C3 depleted mice [23]. C3a is likely to exert an indirect effect on vascular leakage since purified C3a fails to cause permeability of the EC monolayer [21]. This is in contrast with the ability of C5a to cause cell retraction, gap formation and consequent permeability of ECs evaluated by FITC-dextran passage [21].

The fragment C5a is released in inflammatory conditions associated with bacterial infections. A typical example is the infection caused by *Aeromonas sobria* which cleaves C5 through a serine protease with the release of C5a which induce histamine-mediated vascular leakage [24].

More recently, the potential role of C5aR in endotoxin-induced plasma leakage was documented in a mouse model following the observation that the administration of C5aR-siRNA was effective in preventing endothelial injury and vascular leakage [25].

Activation of ECs is also induced by the late C components from C5 to C9 involved in the assembly of the terminal C complex (TCC). The formation of this complex is initiated with the release of C5b from cleaved C5 (Figure 1), followed by progressive binding of the remaining late C components from C6 to C9 (Figure 1). This complex may cause cytolysis through a pore formation following insertion into the plasma membrane of target cells [26], or as a sublytic complex which fails to cause cell lysis despite its binding to the phospholipids bilayer of the cell membrane. The TCC can also assemble in the fluid phase where it accumulates as a cytolytically inactive complex due to the short half-life of its cell binding ability. The complex binds the soluble C regulators S protein and clusterin and circulates in plasma or is present in the extravascular fluid as SC5b-9 [27]. We have shown that this complex, despite its inability to cause cytolysis, promotes vascular leakage [27]. This effect was documented *in vitro* using a transwell system to evaluate the leakage of fluorescein-labeled BSA or the intravital microscopy to follow the extravasation of circulating FITC-BSA across mesenteric microvessels [27]. The permeabilizing activity of SC5b-9 was partially inhibited by the B2R antagonist (HOE-140), or the selective platelet activating factor (PAF) receptor antagonist (CV3988) and was completely neutralized by the mixture of the two antagonists suggesting that the effect of SC5b-9 is mediated by the formation of BK and the release of PAF.

Our observation was further supported by the finding of high levels of SC5b-9 in pleural fluids of patients affected by Dengue virus infections [28]. In these patients, who die mainly from vascular leakage and shock, the nonstructural viral protein 1 (NS1) activate C resulting in the release of anaphylotoxins and SC5b-9. Interestingly, the plasma levels of NS1 and SC5b-9 correlated with the disease severity suggesting that the anaphylotoxins and the terminal complex may contribute to the pathogenesis of the vascular permeability.

3. The role of gC1qR and the kinin-kallikrein system in the pathophysiology of vascular permeability

The classical pathway of C and the kallikrein-kinin system are two unique proinflammatory pathways which share several features. First, both pathways use functionally homologous “active zymogens” to trigger their activation. These so-called “active zymogens” in turn are generated when the inactive precursor enzymes undergo an autoactivation process and acquire enzymatic activity—by virtue of conformational change—when bound to a suitable surface [29]. In the kinin system this is represented by FXII [FXII (or Hageman Factor, HF)], which undergoes autoactivation upon binding to negatively charged surfaces [30]. The functional homologue of FXII in the C system is C1r, which is also able to autoactivate when C1 is bound to immune complexes [31,32]. In both cases, this autoactivation process is what triggers the downstream enzymatic interactions leading to the generation of vasoactive peptides which induce vascular permeability. Second, C1-INH, the most potent inhibitor of the classical pathway of C is not only the major regulator of the kinin system, but also contributes to the regulation of serine proteases of the clotting system [33]. It is therefore not surprising that an unabated activation of either pathway in the absence of C1-INH leads to the generation of factors which enhance vascular permeability, edema and inflammation.

The three essential proteins of the plasma kallikrein-kinin system are: FXII, prekallikrein (PK), and high molecular weight kininogen (HK), and together with a fourth protein FXI form the core of what is called the contact activation or intrinsic pathway of blood

coagulation [34,35]. During the inflammatory response or atherogenesis, the endothelial surface is able to provide the molecular platform requisite for the assembly and activation of the kallikrein-kinin pathway which eventuates in the generation of a powerful peptide called bradykinin (BK) [34]. Activation of the kinin-generating pathway is therefore triggered when cell surface-bound FXII undergoes autoactivation to generate factor XIIa. Factor XIIa then converts PK to kallikrein and the latter is then able to digest surface-bound HK to generate BK [34–38] (Figure 2). The other substrate of Factor XIIa in plasma is surface bound factor XI (FXI), which it cleaves to generate FXIa thus setting in motion the self-amplifying activation of proteins involved in the intrinsic coagulation cascade [34,35]. One of the most important products of the assembly and interactions of the contact activation proteins is the formation of the nonapeptide BK, which is a cleavage product of the initiating step of the cascade [34,38] (6,10). Another pathway by which BK is generated starts intracellularly. First, conversion of prekallikrein to tissue kallikrein takes place inside the cell and the kallikrein generated in this manner is then secreted into the pericellular milieu. Second, the secreted kallikrein digests low molecular weight kininogen (LK) to generate the decapeptide, lysyl-BK (Lys⁰-BK) or kallidin [34,36]. Finally, kallidin is converted to the nonapeptide BK by the zinc-dependent enzyme called aminopeptidase [39]. BK which is produced by either pathway is then degraded—in a sequential manner—to lower-molecular-weight peptides by three kininases. These enzymes are carboxypeptidase N, angiotensin-converting enzyme (ACE), and aminopeptidase P. [39–41]. The major plasma enzyme is carboxypeptidase N, which removes the C-terminal arginine from BK to yield an octapeptide, des-arg⁹-BK [40,41]. The second kininase in plasma is kininase II, which is identical to ACE and is found mainly along the pulmonary vascular endothelial cell surface. This kininase removes the dipeptide phe-arg from the C-terminus of BK to yield a heptapeptide and a second cleavage removes ser-pro to leave a pentapeptide [42].

BK is a powerful endothelium-dependent peptide, which in addition to its well-established function in vasodilation, enhancement of vascular permeability and smooth muscle constriction, has also been implicated in various pathophysiological processes including hypotension, tumor angiogenesis and metastasis, as well as pain [36,38]. More importantly, BK is thought to play a critical role in the onset of the rare disease, hereditary AE. BK action and signaling is mediated by two seven-transmembrane domain containing G-protein-coupled receptors named B1 receptor (B1R) and B2R. While the B2R is ubiquitously and constitutively expressed in healthy tissues, B1R is induced and synthesized *de novo* as a consequence of tissue injury or inflammation [43,44]. Furthermore, while the B2R displays high affinity for BK and Lys-BK peptide agonists, the B1 receptor displays highest affinity to des-Arg⁹-BK and des-Arg⁹-kallidin. Following initial stimulation of B2 receptors, the action of BK is further enhanced by secondary production of vasodilators such as nitric oxide (NO), and prostacyclins, which exacerbate the inflammatory process [38,43]. Interestingly, an undesired side effect angiotensin converting enzyme inhibitors (ACE inhibitors), a class of drugs used to treat patients with hypertension and congestive heart failure, is the inhibition of BK degradation. This in turn elevates plasma levels of BK thereby causing hypotension and vascular permeability [43,44].

Because simultaneous and unregulated activation of both the C and kallikrein-kinin systems along the endothelial surface is largely responsible for the BK-mediated vascular permeability which leads to edema, identification and characterization of the molecule or molecules which facilitate the assembly and activation of the kinin generating pathway has been the focus of intense research for the past 20 years. To date, three endothelial cell-binding sites for HK and factor XII have been described. These include the 33-kDa cell surface receptor for the first component of C C1q (gC1qR/p33) [45–47], cytokeratin-1 [48,49], and the urokinase plasminogen activator receptor (u-PAR) [50]. Although the three proteins bind HK and/or FXII in a zinc-dependent manner and are postulated to form a

multi-protein receptor complex with each contributing to the recruitment and assembly of the kinin generating pathway proteins [51], gC1qR/p33, which binds specifically to domain 5 in the light chain of HK [47,52], may play a central role by virtue of its high affinity for HK. Whereas HK binds to isolated gC1qR/p33 with a K_d of 0.8 ± 0.7 nM ($k_{on} = 12.3 \pm 5.0 \times 10^4$ M⁻¹s⁻¹; $k_{off} = 0.8 \pm 0.5 \times 10^{-4}$ M⁻¹s⁻¹) [53], the binding of HK to endothelial cell surface expressed gC1qR is 9 ± 2 nM, and this binding is completely inhibited by mAb 74.5.2, which recognizes the C-terminal half of gC1qR/p33 corresponding to residues 204–218 [46,54]. Conversely, domain 5 of HK, located at the N-terminus of the light chain, is rich in histidine and arginine residues and contains the site for interaction with gC1qR/p33. In fact a 20 amino acid peptide termed HKH20 and corresponding to domain 5 has been synthesized and shown to inhibit the interaction of HK with intact endothelial cells [47,52].

The release of BK and related peptides and their interaction with B1R or B2R often lead to pathological conditions usually characterized by increased vascular permeability. One of the effects of diabetes is increased retinal vascular permeability due to loss of retinal endothelial tight junctions. This pathology is mediated by the carbonic anhydrase-I induced alkalization of the vitreous which promote the enzymatic activity of kallikrein and the generation of activated factor XII. This activation promote the formation of kinins which are finally responsible for increase of vascular leakage [55]. The same permeabilizing effect of kinins released after stimulation by carbonic anhydrase-I is presumably responsible for intracerebral haemorrhage and edema [55]. Some experimental evidences underline the important role of B2R more than B1R in the blood-brain disruption and brain edema in ischemic-stroke [56]. However, the relevant role of B1R has been shown in other pathological conditions such as endotoxic shock [57] and maintenance of intra-articular edema in arthritis [58]. B1R and B2R are also involved in vascular leakage seen in tumors and infections. In particular antagonist of B2R are able to suppress vascular permeability in solid tumors and reduce accumulation of ascitic fluid [59]. Bacterial surfaces can directly activate HK leading to the release of BK which can be transformed by bacterial proteases to B1 agonists and in this way contribute to bacterial dissemination [59].

4. Angioedema as a model of crosstalk between complement and kinin systems

The data showing that the vascular leakage induced by SC5b-9 is mediated through the release of BK clearly indicate that the C and the kinin systems are very closely linked in the stimulation of the endothelium [27]. However it has been shown that other molecules - such as gC1qR/p33 [60] and C1 inhibitor (C1-INH) [61] - belonging to the C system are involved in the regulation and activation of the kinin system.

The multimolecular receptor complex comprising of gC1qR/p33, uPAR and CK-1 present on EC surface are able to bind the trimolecular complex formed by prekallikrein (PK), HK and FXII [60,62]. Once bound, PK is cleaved by a cell surface serine protease (prolylcarboxypeptidase/PRCP) releasing active kallikrein [63] which generates activated factor XII leading to an amplification loop responsible for a continuous activation of kallikrein.. The vasoactive peptide BK is released from HK as a result of cleavage by kallikrein [60]. These data suggest that the gC1qR/p33 plays a pivotal role in triggering and modulating the coagulation/kinin cascade, being responsible for the generation of the potent proinflammatory peptides BK and related kinins.

C1-INH belongs to a family of serine protease inhibitors called serpins which together constitute 20% of all plasma proteins [64]. This serpin is a glycoprotein of 478 amino acid residues encoded by a single copy gene on chromosome 11 and synthesized by the liver, fibroblasts, monocytes, macrophages, ECs and other cell types [65]. C1-INH is the sole

known inhibitor of C1r and C1s proteases of the first C component (C1), and moreover inactivates the complex formed by MBL and MASPs [66], and inhibits the alternative pathway [67]. In addition, C1-INH is also able to control the serine proteases of the clotting and the kinin systems including Factors XIIa and XIa, kallikrein, plasmin, and tissue plasminogen activator activated by injury to blood vessels and by some bacterial toxins [65].

The inherited and acquired C1-INH deficiency is associated with the onset of AE characterized by episodic and recurrent local edema of subcutaneous and submucosal tissues. The hereditary form of angioedema (HAE) is an autosomal dominant disorder due to a decreased serum level of C1-INH (type I HAE) or to a secretion of a non functional protein (type II HAE). There is a third type of HAE, usually estrogen-dependent and occurring only in women, characterized by the classical swelling and normal function of C1-INH. The acquired form of AE (AAE) are associated with the high consumption of C1-INH (type I) or to the presence of auto-antibodies (type II) or induced by treatment with ACE inhibitors. Although the pathogenesis of the swelling in patients with AE was originally thought to be mediated by C and more specifically by C2-derived kinin [17], ample evidence now exists that the swelling involves primarily the kinin-forming pathway [68]. Nevertheless, the possibility that activation of both C and kinin-forming systems may contribute to the edema has not been completely excluded. Activated plasma kallikrein, with the contribution of factor XII, initiates the kinin cascade (Figure 2), cleaving HK to generate BK. Nussberger et al. [69] documented increased plasma levels of BK in patients with AE. FXIIa is also able to activate C1 and plasmin, leading to the cleavage of C2 into a kinin-like fragment (C2 kinin) [70]. BK, and possibly the C2 fragment, can cause enhanced permeability of post-capillary venules most likely mediated by B1R and B2R localized on the endothelial cell surface [43]. This effect is responsible for the development of edema and the movement of fluid from the intravascular space to the perivascular tissues. A significant activation of the terminal pathway of the C sequence was observed during the attack phase of hereditary AE as documented by the increased levels of SC5b-9 measured in patients during the attack phase [71]. These data together with the *in vitro* and *in vivo* observations indicating a clear permeabilizing activity of SC5b-9 [27] suggest the existence of a cross-talk between the C and the kinin systems in induction of vascular leakage during the acute episodes of AE.

The involvement of B2R in the onset of hereditary AE symptoms is supported by the beneficial effects observed in patients treated with HOE-140 (Icatibant), a selective antagonist of B2R [72]. However, despite the immediate relief obtained with this drug, complete resolution of symptoms requires several hours raising the hypothesis that other molecules and receptors are involved in the maintenance of AE. The two receptors B1R and gC1qR/p33 appear to be the most likely candidates, though their contribution to the release of kinins and the onset of the swelling remains to be fully elucidated. We have recently used *in vitro* and *in vivo* permeability assays to analyse attack phase plasma (APL) samples from C1-INH deficient patients for their ability to cause endothelial leakage [73]. APL induced a FITC-BSA leakage on human adult dermal microvascular ECs and on ECs isolated from the human umbilical vein (HUVECs) which was delayed (30 minutes) as opposed to the rapid action of BK (5 minutes) while the remission plasma (RPL) elicited a modest effect as compared to control plasma. These data were also confirmed by *in vivo* experiments showing that topical application of APL on rat mesenteric microvessels induced a significant increase in vascular leakage, while RPL was ineffective. The receptor for the globular head of C1q was found to participate in this process as the permeabilizing effect of APL on EC monolayer was completely abrogated by a monoclonal antibody recognizing an epitope on gC1qR involved in the interaction with high-molecular weight kininogen. The contribution of B1R to APL-induced BSA leakage was suggested by the decrease in EC permeability obtained with B1R antagonists, which was only partial, as was that obtained with the B2R

antagonist. Both receptors seem to be involved since complete inhibition of leakage was obtained by the combined treatment with B1R and B2R antagonists. However, B1R, unlike B2R, is not constitutively present on the surface of ECs and requires cell activation to be expressed. This effect can be induced by inflammatory stimuli as revealed by the marked increase in the vascular leakage induced by APL from patients with hereditary or acquired AE on ECs exposed to the proinflammatory cytokine IL-1 β . The ability of the protein trafficking inhibitor brefeldin-A to reduce the BSA leakage induced by APL provides further evidence that the newly synthesized B1Rs are involved in the APL-mediated increased permeability of ECs.

In conclusion, these data suggest that the interaction between the HK-prekallikren-FXII trimolecular complex and gC1qR/p33 leading to the formation of BK and des-Arg⁹-BK, and the expression of B1R (Figure 3) represent critical steps in the development of AE and that the blockade of both B1R and B2R receptors or gC1q/p33 may provide novel therapeutic tools to better control the acute attacks in patients with AE.

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References

1. Cines DB, Pollak ES, Buck CA, Loscalzo J, Zimmerman GA, McEver RP, et al. Endothelial cells in physiology and in the pathophysiology of vascular disorders. *Blood*. 1998; 91:3527–3561. [PubMed: 9572988]
2. Laufer J, Katz Y, Passwell JH. Extrahepatic synthesis of complement proteins in inflammation. *Mol Immunol*. 2001; 38:221–229. [PubMed: 11532283]
3. Langedeggen H, Pausa M, Johnson E, Casarsa C, Tedesco F. The endothelium is an extrahepatic site of synthesis of the seventh component of the complement system. *Clin Exp Immunol*. 2000; 121:69–76. [PubMed: 10886241]
4. Bossi F, Rizzi L, Bulla R, Debeus A, Tripodo C, Picotti P, et al. C7 is expressed on endothelial cells as a trap for the assembling terminal complement complex and may exert anti-inflammatory function. *Blood*. 2009; 113:3640–3648. [PubMed: 19179470]
5. Fischetti F, Durigutto P, Pellis V, Debeus A, Macor P, Bulla R, et al. Thrombus formation induced by antibodies to beta2-glycoprotein I is complement dependent and requires a priming factor. *Blood*. 2005; 106:2340–2346. [PubMed: 15956288]
6. Girardi G, Bulla R, Salmon JE, Tedesco F. The complement system in the pathophysiology of pregnancy. *Mol Immunol*. 2006; 43:68–77. [PubMed: 16023727]
7. Collard CD, Montalto MC, Reenstra WR, Buras JA, Stahl GL. Endothelial oxidative stress activates the lectin complement pathway: role of cytokeratin 1. *Am J Pathol*. 2001; 159:1045–1054. [PubMed: 11549596]
8. Ricklin D, Hajishengallis G, Yang K, Lambris JD. Complement: a key system for immune surveillance and homeostasis. *Nat Immunol*. 11:785–797. [PubMed: 20720586]
9. Collard CD, Vakeva A, Morrissey MA, Agah A, Rollins SA, Reenstra WR, et al. Complement activation after oxidative stress: role of the lectin complement pathway. *Am J Pathol*. 2000; 156:1549–1556. [PubMed: 10793066]
10. Moller-Kristensen M, Wang W, Ruseva M, Thiel S, Nielsen S, Takahashi K, et al. Mannan-binding lectin recognizes structures on ischaemic reperused mouse kidneys and is implicated in tissue injury. *Scand J Immunol*. 2005; 61:426–434. [PubMed: 15882434]
11. Wallis R, Mitchell DA, Schmid R, Schwaebler WJ, Keeble AH. Paths reunited: Initiation of the classical and lectin pathways of complement activation. *Immunobiology*. 215:1–11. [PubMed: 19783065]

12. Peerschke EI, Malhotra R, Ghebrehwet B, Reid KB, Willis AC, Sim RB. Isolation of a human endothelial cell C1q receptor (C1qR). *J Leukoc Biol.* 1993; 53:179–184. [PubMed: 8445329]
13. Peerschke EI, Smyth SS, Teng EI, Dalzell M, Ghebrehwet B. Human umbilical vein endothelial cells possess binding sites for the globular domain of C1q. *J Immunol.* 1996; 157:4154–4158. [PubMed: 8892652]
14. Lozada C, Levin RI, Huie M, Hirschhorn R, Naime D, Whitlow M, et al. Identification of C1q as the heat-labile serum cofactor required for immune complexes to stimulate endothelial expression of the adhesion molecules E-selectin and intercellular and vascular cell adhesion molecules 1. *Proc Natl Acad Sci U S A.* 1995; 92:8378–8382. [PubMed: 7545301]
15. van den Berg RH, Faber-Krol MC, Sim RB, Daha MR. The first subcomponent of complement, C1q, triggers the production of IL-8, IL-6, and monocyte chemoattractant peptide-1 by human umbilical vein endothelial cells. *J Immunol.* 1998; 161:6924–6930. [PubMed: 9862726]
16. Ratnoff OD, Lepow IH. Complement as a Mediator of Inflammation. Enhancement of Vascular Permeability by Purified Human C'1 Esterase. *J Exp Med.* 1963; 118:681–698. [PubMed: 14087615]
17. Strang CJ, Auerbach HS, Rosen FS. C1s-induced vascular permeability in C2-deficient guinea pigs. *J Immunol.* 1986; 137:631–635. [PubMed: 3487577]
18. Fields T, Ghebrehwet B, Kaplan AP. Kinin formation in hereditary angioedema plasma: evidence against kinin derivation from C2 and in support of "spontaneous" formation of bradykinin. *J Allergy Clin Immunol.* 1983; 72:54–60. [PubMed: 6222104]
19. Van Beek J, Bernaudin M, Petit E, Gasque P, Nouvelot A, MacKenzie ET, et al. Expression of receptors for complement anaphylatoxins C3a and C5a following permanent focal cerebral ischemia in the mouse. *Exp Neurol.* 2000; 161:373–382. [PubMed: 10683302]
20. Monsinjon T, Gasque P, Chan P, Ischenko A, Brady JJ, Fontaine MC. Regulation by complement C3a and C5a anaphylatoxins of cytokine production in human umbilical vein endothelial cells. *FASEB J.* 2003; 17:1003–1014. [PubMed: 12773483]
21. Schraufstatter IU, Trieu K, Sikora L, Sriramaraio P, DiScipio R. Complement c3a and c5a induce different signal transduction cascades in endothelial cells. *J Immunol.* 2002; 169:2102–2110. [PubMed: 12165538]
22. Miyama K, Takano K, Atsumi I, Nakagawa H. Identification of C3a and N-truncated C3a as vascular permeability-enhancing factors from the exudate of chronic phase of carrageenan-induced inflammation in rats. *Biol Pharm Bull.* 2002; 25:648–651. [PubMed: 12033507]
23. Lister KJ, James WG, Hickey MJ. Immune complexes mediate rapid alterations in microvascular permeability: roles for neutrophils, complement, and platelets. *Microcirculation.* 2007; 14:709–722. [PubMed: 17885996]
24. Nitta H, Imamura T, Wada Y, Irie A, Kobayashi H, Okamoto K, et al. Production of C5a by ASP, a serine protease released from *Aeromonas sobria*. *J Immunol.* 2008; 181:3602–3608. [PubMed: 18714034]
25. Liu ZM, Zhu SM, Qin XJ, Cheng ZD, Liu MY, Zhang HM, et al. Silencing of C5a receptor gene with siRNA for protection from Gram-negative bacterial lipopolysaccharide-induced vascular permeability. *Mol Immunol.* 47:1325–1333. [PubMed: 20138669]
26. Tedesco, F.; Bulla, R.; Fischetti, F. Terminal Complement Complex: Regulation of Formation and Pathophysiological Functions. In: DSzebeni, J., editor. *The Complement System*. KAP; 2004. p. 97-127.
27. Bossi F, Fischetti F, Pellis V, Bulla R, Ferrero E, Mollnes TE, et al. Platelet-activating factor and kinin-dependent vascular leakage as a novel functional activity of the soluble terminal complement complex. *J Immunol.* 2004; 173:6921–6927. [PubMed: 15557188]
28. Avirutnan P, Punyadee N, Noisakran S, Komoltri C, Thiemmecca S, Auethavornanan K, et al. Vascular leakage in severe dengue virus infections: a potential role for the nonstructural viral protein NS1 and complement. *J Infect Dis.* 2006; 193:1078–1088. [PubMed: 16544248]
29. Lachmann PJ, Hughes-Jones NC. Initiation of complement activation. *Springer Semin Immunopathol.* 1984; 7:143–162. [PubMed: 6495149]

30. Silverberg M, Dunn JT, Garen L, Kaplan AP. Autoactivation of human Hageman factor. Demonstration utilizing a synthetic substrate. *J Biol Chem.* 1980; 255:7281–7286. [PubMed: 7391081]
31. Ziccardi RJ. A new role for C-1-inhibitor in homeostasis: control of activation of the first component of human complement. *J Immunol.* 1982; 128:2505–2508. [PubMed: 7077078]
32. Kasahara Y, Odai H, Takahashi K, Nagasawa S, Koyama J. Autocatalytic activation of C1r subcomponent of the first component of human complement. *J Biochem.* 1985; 97:365–372. [PubMed: 2987200]
33. Ratnoff OD, Pensky J, Ogston D, Naff GB. The inhibition of plasmin, plasma kallikrein, plasma permeability factor, and the C'1r subcomponent of the first component of complement by serum C'1 esterase inhibitor. *J Exp Med.* 1969; 129:315–331. [PubMed: 4178758]
34. Kaplan, AP. Mechanisms of bradykinin generation. In: Greaves, MW.; Kaplan, AP., editors. *Urticaria and angioedema.* Marcel Decker Inc; New York: 2009. p. 73-90.
35. Vigler M, Lee SK, Palti R, Williams JC, Kaminsky AJ, Posner MA, et al. Biomechanical comparison of techniques to reduce the bulk of lacerated flexor tendon ends within digital sheaths of the porcine forelimb. *J Hand Surg Am.* 2009; 34:1653–1658. [PubMed: 19762164]
36. Regoli D, Barabe J. Pharmacology of bradykinin and related kinins. *Pharmacol Rev.* 1980; 32:1–46. [PubMed: 7015371]
37. Mori K, Nagasawa S. Studies on human high molecular weight (HMW) kininogen. II. Structural change of HMW kininogen by the action of human plasma kallikrein. *J Biochem.* 1981; 89:1465–1473. [PubMed: 6168636]
38. Bhoola KD, Figueroa CD, Worthy K. Bioregulation of kinins: kallikreins, kininogens, and kininases. *Pharmacol Rev.* 1992; 44:1–80. [PubMed: 1313585]
39. Margolius HS. Tissue kallikreins structure, regulation, and participation in mammalian physiology and disease. *Clin Rev Allergy Immunol.* 1998; 16:337–349. [PubMed: 9926284]
40. Erdos EG, Sloane EM. An enzyme in human blood plasma that inactivates bradykinin and kallidins. *Biochem Pharmacol.* 1962; 11:585–592. [PubMed: 13890520]
41. Sheikh IA, Kaplan AP. Studies of the digestion of bradykinin, lysyl bradykinin, and kinin-degradation products by carboxypeptidases A, B, and N. *Biochem Pharmacol.* 1986; 35:1957–1963. [PubMed: 3718539]
42. Yang HY, Erdos EG. Second kininase in human blood plasma. *Nature.* 1967; 215:1402–1403. [PubMed: 6055465]
43. D'Orleans-Juste P, de Nucci G, Vane JR. Kinins act on B1 or B2 receptors to release conjointly endothelium-derived relaxing factor and prostacyclin from bovine aortic endothelial cells. *Br J Pharmacol.* 1989; 96:920–926. [PubMed: 2545298]
44. Duchene J, Lecomte F, Ahmed S, Cayla C, Pesquero J, Bader M, et al. A novel inflammatory pathway involved in leukocyte recruitment: role for the kinin B1 receptor and the chemokine CXCL5. *J Immunol.* 2007; 179:4849–4856. [PubMed: 17878384]
45. Ghebrehwet B, Lim BL, Peerschke EI, Willis AC, Reid KB. Isolation, cDNA cloning, and overexpression of a 33-kD cell surface glycoprotein that binds to the globular "heads" of C1q. *J Exp Med.* 1994; 179:1809–1821. [PubMed: 8195709]
46. Joseph K, Ghebrehwet B, Peerschke EI, Reid KB, Kaplan AP. Identification of the zinc-dependent endothelial cell binding protein for high molecular weight kininogen and factor XII: identity with the receptor that binds to the globular "heads" of C1q (gC1q-R). *Proc Natl Acad Sci U S A.* 1996; 93:8552–8557. [PubMed: 8710908]
47. Herwald H, Dedio J, Kellner R, Loos M, Muller-Esterl W. Isolation and characterization of the kininogen-binding protein p33 from endothelial cells. Identity with the gC1q receptor. *J Biol Chem.* 1996; 271:13040–13047. [PubMed: 8662673]
48. Hasan AA, Zisman T, Schmaier AH. Identification of cytokeratin 1 as a binding protein and presentation receptor for kininogens on endothelial cells. *Proc Natl Acad Sci U S A.* 1998; 95:3615–3620. [PubMed: 9520414]
49. Joseph K, Ghebrehwet B, Kaplan AP. Cytokeratin 1 and gC1qR mediate high molecular weight kininogen binding to endothelial cells. *Clin Immunol.* 1999; 92:246–255. [PubMed: 10479529]

50. Colman RW, Pixley RA, Najamunnisa S, Yan W, Wang J, Mazar A, et al. Binding of high molecular weight kininogen to human endothelial cells is mediated via a site within domains 2 and 3 of the urokinase receptor. *J Clin Invest.* 1997; 100:1481–1487. [PubMed: 9294114]
51. Joseph K, Tholanikunnel BG, Ghebrehwet B, Kaplan AP. Interaction of high molecular weight kininogen binding proteins on endothelial cells. *Thromb Haemost.* 2004; 91:61–70. [PubMed: 14691569]
52. Hasan AA, Cines DB, Herwald H, Schmaier AH, Muller-Esterl W. Mapping the cell binding site on high molecular weight kininogen domain 5. *J Biol Chem.* 1995; 270:19256–19261. [PubMed: 7642598]
53. Pixley RJ, Espinola RG, Ghebrehwet B, Joseph K, Kao A, Cines DB, et al. Interaction of High Molecular Weight Kininogen with endothelial cell binding proteins uPAR, gC1qR and Cytokeratin 1 determined by plasmon resonance (BiaCore). *Thrombosis and Hemostasis.* 2011
54. Ghebrehwet B, Lu PD, Zhang W, Lim BL, Eggleton P, Leigh LE, et al. Identification of functional domains on gC1Q-R, a cell surface protein that binds to the globular "heads" of C1Q, using monoclonal antibodies and synthetic peptides. *Hybridoma.* 1996; 15:333–342. [PubMed: 8913782]
55. Gao BB, Clermont A, Rook S, Fonda SJ, Srinivasan VJ, Wojtkowski M, et al. Extracellular carbonic anhydrase mediates hemorrhagic retinal and cerebral vascular permeability through prekallikrein activation. *Nat Med.* 2007; 13:181–188. [PubMed: 17259996]
56. Su J, Cui M, Tang Y, Zhou H, Liu L, Dong Q. Blockade of bradykinin B2 receptor more effectively reduces postischemic blood-brain barrier disruption and cytokines release than B1 receptor inhibition. *Biochem Biophys Res Commun.* 2009; 388:205–211. [PubMed: 19647718]
57. Merino VF, Todiras M, Campos LA, Saul V, Popova E, Baltatu OC, et al. Increased susceptibility to endotoxic shock in transgenic rats with endothelial overexpression of kinin B(1) receptors. *J Mol Med.* 2008; 86:791–798. [PubMed: 18425495]
58. Cruwys SC, Garrett NE, Perkins MN, Blake DR, Kidd BL. The role of bradykinin B1 receptors in the maintenance of intra-articular plasma extravasation in chronic antigen-induced arthritis. *Br J Pharmacol.* 1994; 113:940–944. [PubMed: 7858889]
59. Maeda H, Wu J, Okamoto T, Maruo K, Akaike T. Kallikrein-kinin in infection and cancer. *Immunopharmacology.* 1999; 43:115–128. [PubMed: 10596842]
60. Shariat-Madar Z, Mahdi F, Schmaier AH. Assembly and activation of the plasma kallikrein/kinin system: a new interpretation. *Int Immunopharmacol.* 2002; 2:1841–1849. [PubMed: 12489798]
61. Davis AE 3rd. The pathophysiology of hereditary angioedema. *Clin Immunol.* 2005; 114:3–9. [PubMed: 15596403]
62. Mahdi F, Madar ZS, Figueroa CD, Schmaier AH. Factor XII interacts with the multiprotein assembly of urokinase plasminogen activator receptor, gC1qR, and cytokeratin 1 on endothelial cell membranes. *Blood.* 2002; 99:3585–3596. [PubMed: 11986212]
63. Shariat-Madar Z, Mahdi F, Schmaier AH. Identification and characterization of prolylcarboxypeptidase as an endothelial cell prekallikrein activator. *J Biol Chem.* 2002; 277:17962–17969. [PubMed: 11830581]
64. Beltrani, VS. Angioedema: some new thoughts regarding idiopathic angioedema. In: Greaves, MV.; Kaplan, AP., editors. *Urticaria and angioedema.* Marcel Dekker, inc; New York: 2004. p. 421-439.
65. Davis, AE, 3rd. C1 inhibitor gene and hereditary angioedema. Volanakis, E.; Frank, M., editors. *The Human Complement System in Health and Disease;* New York: 1998. p. 455-480.
66. Petersen SV, Thiel S, Jensen L, Vorup-Jensen T, Koch C, Jensenius JC. Control of the classical and the MBL pathway of complement activation. *Mol Immunol.* 2000; 37:803–811. [PubMed: 11257302]
67. Jiang H, Wagner E, Zhang H, Frank MM. Complement 1 inhibitor is a regulator of the alternative complement pathway. *J Exp Med.* 2001; 194:1609–1616. [PubMed: 11733575]
68. Kaplan, AP. C1 inhibitor deficiency. In: Greaves, MV.; Kaplan, AP., editors. *Urticaria and angioedema.* Marcel Dekker inc; New York: 2004. p. 303-320.
69. Nussberger J, Cugno M, Amstutz C, Cicardi M, Pellacani A, Agostoni A. Plasma bradykinin in angio-oedema. *Lancet.* 1998; 351:1693–1697. [PubMed: 9734886]

70. Donaldson VH, Rosen FS, Bing DH. Role of the second component of complement (C2) and plasmin in kinin release in hereditary angioneurotic edema (H.A.N.E) plasma. *Trans Assoc Am Physicians*. 1977; 90:174–183. [PubMed: 148134]
71. Nielsen EW, Johansen HT, Hogasen K, Wuillemin W, Hack CE, Mollnes TE. Activation of the complement, coagulation, fibrinolytic and kallikrein-kinin systems during attacks of hereditary angioedema. *Immunopharmacology*. 1996; 33:359–360. [PubMed: 8856186]
72. Cicardi M, Banerji A, Bracho F, Malbran A, Rosenkranz B, Riedl M, et al. Icatibant, a new bradykinin-receptor antagonist, in hereditary angioedema. *N Engl J Med*. 363:532–541. [PubMed: 20818888]
73. Bossi F, Fischetti F, Regoli D, Durigutto P, Frossi B, Gobeil F Jr, et al. Novel pathogenic mechanism and therapeutic approaches to angioedema associated with C1 inhibitor deficiency. *J Allergy Clin Immunol*. 2009; 124:1303–1310. e1304. [PubMed: 19796797]

Highlights

- Complement and kinin systems play a role in hereditary and acquired angioedema.
- C1-inhibitor controls activation of both kinin and complement systems.
- Bradykinin contributes to angioedema attacks interacting with B2 receptors.
- Interaction of high molecular weight kininogen with gC1qR induces bradykinin release.
- B1 and gC1q receptors represent novel therapeutic targets for angioedema attacks.

FIGURE 1.

C1-Inhibitor is a key regulator of the complement cascade inhibiting the enzymatic activity of C1r and C1s in the classical pathway, blocking MASPs in the lectin pathway. In addition, C1-Inhibitor binds to C3b, interfering with the formation of the C3 convertase of the alternative pathway .

FIGURE 2.

Schematic representation of the assembly of the kallikrein-kinin system on the endothelial cell surface. In this model, the multi-protein receptor complex is represented as a tandem of urokinase plasminogen activator receptor (uPAR)-Cytokeratin 1 (CK-1) and gC1qR-CK-1 complex. The designation is based on the affinity the proteins have for each other. Binding of FXII to the uPAR-CK-1 complex generates the initial enzyme through autoactivation of FXII to FXIIa. FXIIa in turn converts PK to kallikrein within the PK/HK complex bound to the gC1qR moiety of the gC1qR-CK-11 complex. Kallikrein then cleaves HK to generate bradykinin (BK). Generation of BK induces vascular permeability through signalling via B2 receptor (BK-B2 receptor) constitutively expressed on ECs.

FIGURE 3.

Mechanisms of activation of the kinin system on the surface of endothelial cells during angioedema attacks. The trimolecular complex formed by Factor XII, prekallikrein and high molecular kininogen (FXII-PK-HK) interacts with the endothelial cell membrane through a binding site formed by gC1qR/p33, urokinase plasminogen activator receptor (uPAR) and cytokeratin 1 (CK-1). The consequent activation of kallikrein (K) by FXIIa and prolylcarboxypeptidase (PRCP) leads to BK generation. BK may stimulate B2R or may be cleaved by carboxypeptidase M (CPM), on the cell membrane, or CPN, in the soluble phase, forming des-Arg⁹-BK (DABK). This is the agonist for B1R which is an inducible receptor expressed on the membrane following stimulation by proinflammatory molecules.