Cross Talk Between the Inflammation and Coagulation Systems

J LYNNE WILLIAMS

ABBREVATIONS: APC = activated protein C; AT = antithrombin; C4bBP = C4b binding protein; DIC = disseminated intravascular coagulation; EC = endothelial cells; EPCR = endothelial cell protein C receptor; GAG = glycosaminoglycans; PAF = platelet activating factor; PAI-1 = plasminogen activator inhibitor-1; PARs= protease activated receptor; PC = protein C; PDGF = platelet derived growth factor; TAFI = thrombin activated fibrinolysis inhibitor; TFPI = tissue factor pathway inhibitor; TM = thrombomodulin; TNF = tumor necrosis factor; ULVWF = ultra-large multimers of von Willebrand factor.

INDEX TERMS: coagulation; cytokines; endothelial cells; hemostasis; inflammation; leukocytes.

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LEARNING OBJECTIVES

1. Identify the major components (cellular and inflammatory mediators) of inflammation.
2. Describe the functions of the inflammatory response.
3. List the major inflammatory cytokines.
4. Identify the significant effects of the inflammatory mediators on the coagulation and fibrinolytic systems.
5. Identify the significant effects of the coagulation and fibrinolytic systems on inflammation.

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Previously, blood coagulation and inflammation were thought to be completely different physiologic processes. It is now recognized, however, that these two systems are interrelated as part of the host defense mechanism.[1,2] In invertebrates, the functions of clotting and inflammation are mediated by a single cell system, the hemocyte.[3] In vertebrates, clotting and inflammation have diverged into the specialized functions of the platelets, phagocytic cells (neutrophils and macrophages), and several plasma protein systems (procoagulant proteins, complement proteins, proteins of the kinin system).

Several observations suggest an ongoing interaction between these two systems. There is significant structural homology between the proteins of the complement and coagulation cascades. Both cascades utilize serine proteases and are activated through a series of proteolytic cleavage reactions. Animals subjected to experimental sepsis have multi-organ failure associated with activation of blood coagulation (mediated primarily by inflammatory mechanisms). Inhibition of coagulation factor Xa activity inhibits fibrin formation but does not reduce mortality in this model system. However, inhibition of tissue factor/factor VIIa activity or infusion of activated protein C not only reduces fibrin formation, but also significantly reduces mortality.[4] Thus, there are interactions between the proteins of these two systems that go beyond simply the activation of fibrin formation.

Both coagulation and inflammation are essential parts of the host defensive response. These pathways have several connecting points through which they interact. Importantly, the interaction between coagulation and inflammation is bidirectional: both inflammation-induced coagulation as well as coagulation-induced inflammation occur.[5]

THE INFLAMMATORY RESPONSE

The inflammatory response refers to the biochemical and cellular processes that occur in vascularized tissue in response

to injury or infection.[6] It encompasses a complex series of events fundamental to the ability of the human body to protect itself against injurious and infectious agents. The inflammatory response requires the coordinated interaction of 1) local resident cells in the tissues (macrophages, mast cells, fibroblasts); 2) vascular endothelial cells; 3) plasma mediator systems; 4) leukocytes.[7]

The functions of the inflammatory response are: to neutralize or eliminate the offending agent (or to destroy the necrotic tissue); if this cannot be accomplished, the inflammatory response attempts to wall off and confine these agents to limit their effects on the host; to stimulate and enhance the adaptive immune response; and to promote healing.

The classic macroscopic hallmarks of inflammation have been recognized for more than 5000 years and include redness, swelling, heat, and pain. At the microscopic level, these can be explained by the dilation of blood vessels, which increases blood flow into the area, an increase in vascular permeability resulting in the outward leakage of plasma from the vessels into the tissues, and the adherence and emigration of leukocytes through the vascular wall into the surrounding tissues at the site of injury.

The major components of the inflammatory response are listed in Table 1. The process is regulated by a number of inflammatory mediator systems, listed in Table 2. Inflammatory mediators include endogenous (from cells or plasma) as well as exogenous (from bacteria or other pathogens) substances. As can be seen from the table, the list includes a number of components from the coagulation/hemostatic system.

Thus the blood vessels and blood, in addition to being the transport mechanism for the distribution of nutrients and oxygen throughout the body (and the removal of metabolic waste products), are also the essential components of the body's natural immunity (defense) against foreign pathogens and defense against tissue injury. Following injury or infection, an inflammatory response is activated, the integrity of the endothelium is compromised, and phagocytic cells are activated. The endothelium, which is normally anticoagulant and antithrombotic in the resting state, changes to a procoagulant surface, and the balance of the hemostatic system shifts towards hypercoagulability.[7,8]

INFLAMMATION AND COAGULATION

It has long been recognized that systemic inflammation is associated with hypercoagulability, as evidenced by the common occurrence of disseminated intravascular coagulation (DIC) in severe sepsis.[9] More recently, the molecular basis of this association has been determined. Most of the hypercoagulable effects of inflammation are mediated via the inflammatory cytokines, including IL-1, IL-6 and tumor necrosis factor (TNF).[10,11]

The hypercoagulability associated with systemic inflammation is due primarily to an increase in procoagulant functions, an inhibition of fibrinolysis, and a downregulation of the three major physiologic anticoagulant systems (activated protein C/APC, antithrombin/AT, tissue factor pathway inhibitor/TFPI, Figure 1):

1. *Decreased activity of the protein C (PC) anticoagulant system* (Figure 2). This is due to decreased endothelial cell expression of thrombomodulin (TM), triggered by IL-6, IL-1 and TNF.[12,13] TM binds thrombin to activate PC to activated protein C (APC). These cytokines also decrease endothelial cell expression of the endothelial cell PC receptor (EPCR), which binds and localizes PC, facilitating its activation by TM/thrombin.[14] In addition, they trigger the hepatic "acute phase response" resulting in increased synthesis and secretion of several acute phase proteins, including C4b binding protein (C4bBP). C4bBP binds plasma protein S, reducing the quantity of "free" protein S. Free protein S is a cofactor for the APC degradation of the procoagulant proteins activated Factor Va and Factor VIIIa.

2. *Decreased activity of other coagulation inhibitors* (Figure 2). Both tissue factor pathway inhibitor (TFPI) and antithrombin (AT) are "negative acute phase response proteins" (meaning their synthesis and secretion is reduced during the acute phase response).[5] Inflammatory cytokines decrease EC expression of glycosaminoglycans (GAG), the endogenous cofactor for activation of AT activity.[15]

Table 1. Components of the inflammatory response

Cells	Plasma protein systems
PMNs (neutrophils)	Complement proteins
Monocytes/macrophages	Coagulation proteins
Eosinophils	Kinin system proteins
Basophils/mast cells	
Platelets	

3. *Increased procoagulant activity of the endothelium.* Inflammatory cytokines upregulate endothelial expression of tissue factor, the procoagulant protein required for factor VIIa activity,[11] and down regulate endothelial expression of heparin sulfate/glycosaminoglycans, the cofactors for the anticoagulant activity of antithrombin.[15]
4. *Increase in plasma procoagulants.* Plasma fibrinogen levels rise, as fibrinogen is another acute phase protein.[16] Inflammatory mediators induce the endothelial release of ultra-large multimers of von Willebrand factor (ULVWF)—which are hemostatically more effective in inducing platelet activation responses.[17] They also inhibit the cleavage of ULVWF by ADAMTS13. Inflammatory mediators upregulate TF expression on circulating monocytes, shedding of monocyte-derived microparticles, and initiating systemic coagulation[2] (Figure 3).
5. *Decreased activity of the fibrinolytic system*. Inflammatory cytokines stimulate endothelial cells to increase production of plasminogen activator inhibitor-1 (PAI-1).[18,5]
6. *Increased platelet count and platelet reactivity*. IL-6 promotes platelet production and is also associated with an increased platelet reactivity (as the newly formed platelets are activated at lower concentrations of agonists).[19]

Table 2. Inflammatory mediator systems

Mediator system	Major actions
Vasoactive amines:	Induction of adhesion, ↑P-, E-selectin
Histamine, serotonin	Vasodilation; ↑vascular permeability
Vasoactive peptides:	
Kinins (bradykinin)	Vasodilation, ↑vascular permeability Endothelial contraction; pain
C3a, C5a	↑vascular permeability Chemotaxis; activation of mast cells
Other complement components:	
C3b	Opsonization (assist phagocytosis)
C56789	Cytolysis
Coagulation/fibrinolytic:	
Fibrinopeptides A, B (FPA,FPB)	↑vascular permeability; chemotactic
Fibrin degradation products (FDP)	↑vascular permeability; chemotactic
Plasmin	Generates FDPs; activates C3, factor XIIa, prekallikrein, releases kinins
Thrombin	Releases FPA, FPB; ↑P-, E-selectin; Activate endothelial cells, platelets; Chemotactic; induce cytokine release
Phospholipids:	
Prostaglandins, leukotrienes	Vasodilation; ↑vascular permeability; Chemotaxis
Platelet activating factor (PAF)	↑vascular permeability; cell activation
Reactive oxygen intermediates:	
O_2^-, H_2O_2, HO·	Tissue damage (cytolysis); activation of complement; generate chemotactic lipids
Lysosomal granule contents	Tissue damage (proteolysis); matrix degradation; generate oxidant reactions
Cytokines/chemokines	Cell activation; induction of adhesion, chemotaxis, fever, acute-phase response

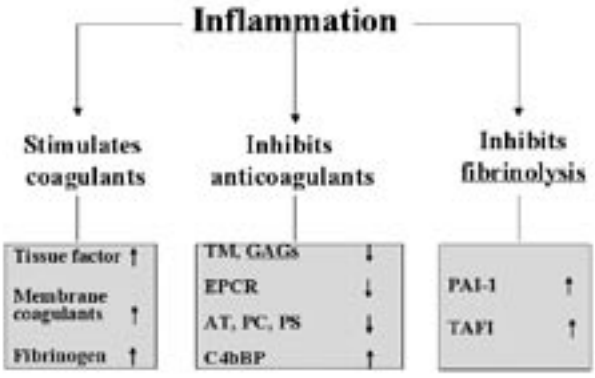
INFLUENCE OF COAGULATION ON INFLAMMATION

Plasma procoagulant enzymes stimulate and augment the inflammatory response. Coagulation inhibitor systems have anti-inflammatory activities in addition to their anti-coagulant functions:

1. *Thrombin, factors Xa and VIIa,* in addition to their roles in activating coagulation protein zymogens, can interact with specific cell receptors and activate intracellular signaling pathways that mediate inflammatory responses. These enzymes bind protease activated receptors (PARs) on cell membranes and induce a cellular response (Figure 4).[2,20,21]

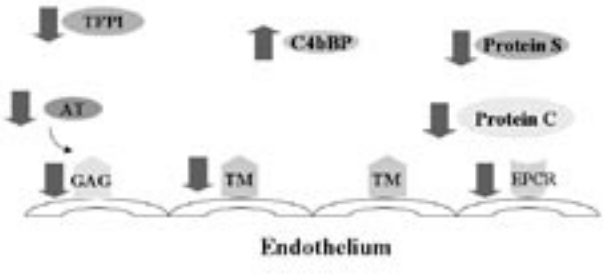
2. *Thrombin* stimulates endothelial cells (EC), promoting vasoconstriction and increased vascular permeability; promotes adhesion and transendothelial migration of leukocytes; stimulates EC and fibroblasts to produce inflammatory cytokines; activates platelets, inducing the release of platelet-derived inflammatory mediators.[22,23]

Figure 1. Impact of inflammatory mediators on coagulation



TM = Thrombomodulin; GAGs = Glycosaminoglycans; EPCR = Endothelial cell protein C receptor; AT = Antithrombin; PC = Protein C; PS = Protein S; C4bBP = Complement component 4b binding protein; PAI-1 = Plasminogen activator inhibitor-1; TAFI = Thrombin activatable fibrinolysis inhibitor

Figure 2. Effects of inflammation on coagulation inhibitor systems



TFPI = Tissue factor pathway inhibitor; AT = Antithrombin; GAG = Glycosaminoglycan; TM = Thrombomodulin; C4bBP = Complement component 4b binding protein; EPCR = Endothelial cell protein C receptor

3. *Factors VIIa and Xa* promote leukocyte adhesion and migration and the synthesis and secretion of inflammatory cytokines.[22,24]

4. *Activated protein C (APC)* can bind to a PAR, and generate signals that inhibit the inflammatory response (Figure 5).[22]

5. *AT, APC, and TFPI* all interact with inflammatory cells and decrease the production of inflammatory mediators, and leukocyte activation in inflammation (Figure 5).[2,5]

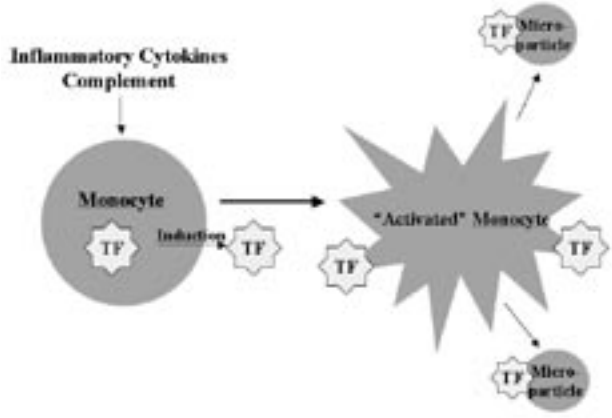
6. *Thrombin activated fibrinolysis inhibitor (TAFI)*, in addition to inhibiting fibrinolysis, inactivates proinflammatory mediators including C3a, C5a, and bradykinin (Figure 5).[2,5,25]

7. *Platelets* are a rich source of inflammatory mediators; when activated, they secrete a number of factors that promote vascular reactions, leukocyte chemotaxis, endothelial cell activation, and other aspects of the inflammatory response including serotonin, platelet activating factor (PAF), and platelet derived growth factor (PDGF).[1,26]

CLINICAL IMPLICATIONS OF "CROSS TALK" BETWEEN INFLAMMATION AND COAGULATION

The processes of coagulation, thrombosis, and inflammation do not happen in isolation. There is a clear interaction

Figure 3. Upregulation of tissue factor on monocytes by inflammatory cytokines



Monocytes are stimulated by cytokines or complement components, triggering tissue factor (TF) synthesis, monocyte shape change and the release of tissue factor-rich microparticles.

between all three, in which thrombosis and coagulation can act as triggers for inflammation, and severe or systemic inflammatory responses can trigger coagulation.

An early outcome of the understanding of the interaction between these processes was the use of the laboratory test, high sensitivity C-reactive protein (hsCRP), as a marker for atherosclerosis and coronary artery disease.[27,28] It is now recognized that an elevated hsCRP may be an early, pre-clinical symptom indication of atherosclerotic inflammatory disease and impending acute thrombotic events. Similarly, studies are underway to evaluate the usefulness of serum IL-6 assays as an indication of the same.

A better understanding of the inflammation/coagulation interface has prompted ongoing research on the interaction of coagulation and inflammation in a variety of disease processes, including inflammatory bowel disease, rheumatoid arthritis, and malignancy. An intriguing study suggesting the importance of the contribution of inflammation to atherothrombosis reported that patients with rheumatoid arthritis exhibited increased risk of myocardial infarction.[29] Additional clinical implications of the interaction of these two systems in other disease processes is likely.

An important outcome of the recognition that inflammation and coagulation are intricately related processes is the use of natural anticoagulants as a treatment in acute inflammatory diseases, such as severe sepsis. Activated protein C has been shown to significantly decrease and morbidity and mortality associated with severe sepsis in an animal model,[4] as well as in early human clinical trials.[30] As our understanding of the molecular mechanisms underlying the close relationship between these two systems progresses, it is likely that new potential targets for therapies will be evaluated, capable of modulating excessive activation of both systems simultaneously.

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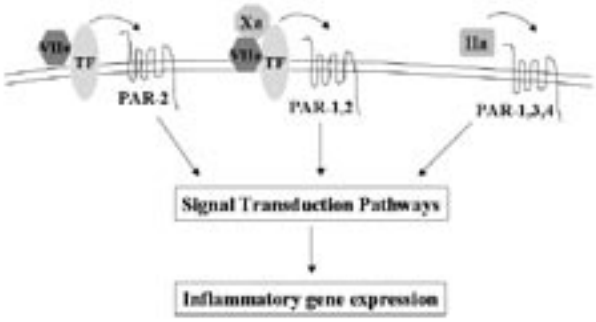
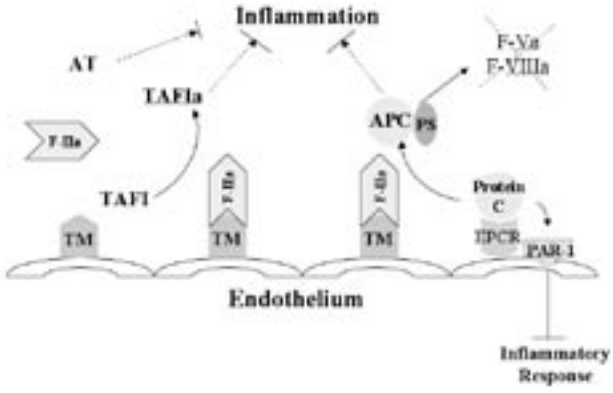
Figure 4. Coagulation proteases and inflammation



Activated coagulation proteases (factor IIa or thrombin, factor VIIa, factor Xa) not only interact with coagulation protein zymogens, but also with specific cell receptors (PARs) to induce signaling pathways that mediate inflammatory responses. PARs are protease activated receptors (of which there are four types, PAR 1 to 4) which, upon cleavage by the coagulation protease, initiates transmembrane signaling, and the induction of genes necessary for the inflammatory response (e.g. synthesis of inflammatory cytokines, cell adhesion molecules, activation of inflammatory responses in macrophages).

TF = Tissue factor; PAR = Protease activated receptor; VIIa = activated factor VII; Xa = activated Factor X; IIa = thrombin (activated factor II)

Figure 5. Anti-inflammatory actions of anticoagulant proteins



The natural anticoagulant proteins also have anti-inflammatory actions. These include antithrombin, activated TAFI, activated Protein C.

TM = thrombomodulin; F-IIa = activated factor II or thrombin; TAFI = Thrombin activatable fibrinolysis inhibitor; AT = Antithrombin; APC = Activated protein C; EPCR = Endothelial cell protein C receptor; PAR-1 = Protease activated receptor-1; F Va = activated factor V; F VIIIa = activated factor VIII

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