The platelet-activating factor signaling system and its regulators in syndromes of inflammation and thrombosis

Guy A. Zimmerman, MD; Thomas M. McIntyre, PhD; Stephen M. Prescott, MD; Diana M. Stafforini, PhD

Objectives: **To review the platelet-activating factor (PAF) signaling system, its regulation, and its dysregulation in acute inflammation and thrombosis and in syndromes that involve these cascades, including sepsis.**

Data Sources: **A summary of published literature from MEDLINE search files and published reviews.**

Data Extraction, Synthesis, and Summary: **PAF, a phospholipid signaling molecule, transmits outside-in signals to intracellular transduction systems and effector mechanisms in a variety of cell types, including key cells of the innate immune and hemostatic systems: neutrophils, monocytes, and platelets. Thus, the PAF signaling system is a point of convergence at which injurious stimuli can trigger and amplify both acute inflammatory and thrombotic cascades. The biological activities of PAF are regulated by several precise mechanisms that, together, constrain and control its action in physiologic inflammation. Unregulated synthesis of PAF or defects in the mechanisms that limit its biological activities have the potential to cause pathologic inflammation and thrombosis. In addition, nonenzymatic generation of oxidized phospholipids that are recognized by the PAF receptor can trigger inflammatory and thrombotic events. There is evidence that the PAF signaling system is dysregulated in sepsis, shock, and traumatic injury and that interruption or termination of its effector responses leads to beneficial outcomes. Plasma PAF acetylhydrolase, an enzyme that hydrolyzes PAF and structurally related oxidized phospholipids, yielding products that are no longer recognized by the PAF receptor, may be a particularly important signal terminator.**

Conclusion: **The PAF signaling system can trigger inflammatory and thrombotic cascades, amplify these cascades when acting with other mediators, and mediate molecular and cellular interactions (cross talk) between inflammation and thrombosis. Evidence from *in vitro* experiments, studies of experimental animals, and clinical observations in humans indicates that the PAF signaling system is important in sepsis and other syndromes of inflammatory injury and that therapeutic strategies to interrupt or terminate signaling via the PAF signaling system may be useful in these conditions. (Crit Care Med 2002; 30[Suppl.]:S294–S301)**

KEY WORDS: inflammation; platelet-activating factor; platelet-activating factor acetylhydrolase; platelet-activating factor–like lipid; platelet-activating factor receptor; sepsis; thrombosis

This focused review outlines key features of the platelet-activating factor (PAF) signaling system that are relevant to unregulated inflammation and thrombosis in sepsis, septic shock, and related syndromes of critical illness.

PAF SIGNALING SYSTEM

PAF. PAF is the central component in a system that has evolved to transmit juxtacrine and paracrine signals between cells (1, 2). In some cases, PAF may also have endocrine, autocrine, and intercrine signaling roles (3–6). A variety of cell types, many of which are central to the inflammatory and hemostatic systems, synthesize PAF. The enzymatic synthesis of PAF is highly regulated and most commonly involves a two-step mechanism that has been characterized in endothelial cells and other inflammatory and vascular cells; additional synthetic mechanisms operate in other tissues (5, 6).

PAF Receptor. The PAF receptor, a plasma membrane receptor that specifically recognizes PAF and related PAF-like lipids, is a central component in the PAF signaling system. The structural features of PAF and related lipids that confer their selective recognition by the PAF receptor have been defined (6, 7). The plasma membrane receptor for PAF is a member of the seven membrane–spanning domain G protein–linked superfamily that has been extensively studied in pharmacologic experiments (8). The receptors from several species have been cloned and characterized at the molecular level (5, 6, 8, 9). The murine PAF receptor has been deleted by homologous recombination and also overexpressed, yielding important biological and pathophysiologic insights into the PAF signaling system (9–13). Recently, a single nucleotide polymorphism in the human PAF receptor that has a significant frequency in the Japanese population was identified. The amino acid substitution, which occurs in the putative third cytoplasmic domain, partially impairs coupling of the receptor to intracellular signal transduction cascades (14). This may account for interindividual variation in responses to PAF, which occur in human subjects and inbred mouse strains (15–18). The PAF receptor undergoes homologous desensitization (19–22), a control mechanism that potentially limits its signaling actions. Homologous desensitization was also used to characterize specific actions of PAF before the development of highly selective competitive antagonists (23). Al-

From the Program in Human Molecular Biology and Genetics (GAZ, TMM), the Huntsman Cancer Institute (SMP, DMS), and the Departments of Internal Medicine (GAZ, TMM, SMP, DMS) and Pathology (TMM), University of Utah, Salt Lake City, UT.

Presented, in part, at the Margaux Conference on Critical Illness, Sedona, AZ, November 14–18, 2001.

Supported, in part, by the National Institutes of Health, the American Heart Association, and the Nora Eccles Treadwell Foundation.

Address requests for reprints to: Guy A. Zimmerman, MD, 15 North 2030 East, Building 533, Room 4220, University of Utah, Salt Lake City, UT 84112. E-mail: guy.zimmerman@hmbg.utah.edu

terations in expression of PAF receptor mRNA and protein occur in response to inflammatory and developmental stimuli in isolated cells and, presumably, *in vivo* (6, 24).

PAF Receptor-Induced Signaling. The PAF receptor is coupled via G proteins to intracellular signaling enzymes and pathways that regulate cytoplasmic calcium concentration, phosphatidylinositol turnover, cyclic AMP levels, and phosphorylation states of critical proteins (6, 9). The diversity of signaling cascades linked to the PAF receptor explains the varied and pleiotropic effector responses and functional changes in cells that are induced when it is engaged; this pattern of functional responses is cell specific and can be modulated by co-engagement of adhesion molecules or other surface receptors (2, 25, 26).

Many signaling events, and consequent functional responses, triggered by the PAF receptor occur in seconds to minutes and do not require new gene expression. However, ligation of the PAF receptor can also lead to nuclear signaling and transcriptional induction of genes involving nuclear factor-κB and other transcription factors (3, 27). In addition, we recently found that PAF induces activation of signal-dependent translation pathways in human platelets (28) and neutrophils (S Lindemann et al., unpublished observations). The latter observations demonstrate that the PAF receptor modulates new gene expression by signaling to key posttranscriptional checkpoints, in addition to activating transcription.

PAF IS A PIVOTAL MEDIATOR THAT LINKS THE HEMOSTATIC AND INNATE IMMUNE SYSTEMS

The intimate relationship between the hemostatic and innate immune systems has been recognized for many years, and new mechanisms of convergence between these systems continue to be discovered. Acute inflammation is a requisite response mediated by the innate immune system that is critical in defense against microbial infection and in wound surveillance and repair. Hemostasis is an equally critical homeostatic response to injury. The PAF receptor is constitutively expressed on human platelets and on key effector cells of the innate immune system—monocytes and polymorphonuclear leukocytes (PMNs; neutrophils)—establishing PAF as a signaling molecule with the capacity to trigger both thrombotic and acute inflammatory events (Fig. 1). These responses may be particularly important in sepsis, shock, and traumatic tissue injury, in which unregulated inflammation and pathologic thrombosis are central pathophysiologic mechanisms and interplay between the two systems may amplify systemic manifestations and tissue injury (29).

Platelet Aggregation. One of PAF's earliest identified activities was *in vitro* activation of platelets isolated from experimental animals, and this led to its trivial name (8). Intravenous infusion of PAF into baboons causes acute thrombocytopenia and neutropenia and was one of the earliest observations suggesting that it may have thrombotic and pro-inflammatory effects *in vivo* (30). PAF was subsequently shown in other species to trigger *in vivo* aggregation and accumulation of platelets and consequent changes in local blood flow at sites of experimental thrombosis and vascular injury (31). PAF also activates human platelets (32) and induces aggregation at nanomolar concentrations (33, 34). Platelet aggregation is mediated by inside-out signaling of integrin $\alpha_{IIb}\beta_3$ and consequent binding of fibrinogen (35), indicating that the PAF receptor is linked to this prothrombotic intracellular pathway. Engagement of integrin $\alpha_{IIb}\beta_3$ can then, in turn, mediate outside-in signaling and additional amplification responses (35).

Cytokine Synthesis by Platelets. Rapid aggregation, release of preformed mediators, and synthesis of eicosanoids are responses of platelets that have been recognized for many years. In addition to these "traditional" activities, our recent experiments indicate that platelets have a previously unrecognized synthetic repertoire that can be activated by PAF and other agonists. Freshly isolated human platelets carry many constitutive messenger RNAs (mRNAs) that were transcribed at the nucleated megakaryocyte stage. This had been recognized earlier, but more recently, we further documented the presence of constitutive mRNAs by using interrogation of arrayed complementary DNA libraries (28, 36). In response to appropriate activating signals, some of these mRNAs are translated to their corresponding proteins in a highly regulated manner (28, 36, 37). Intracellular compartmentalization and cytoskeletal association of critical translation control factors is an important mechanism (38).

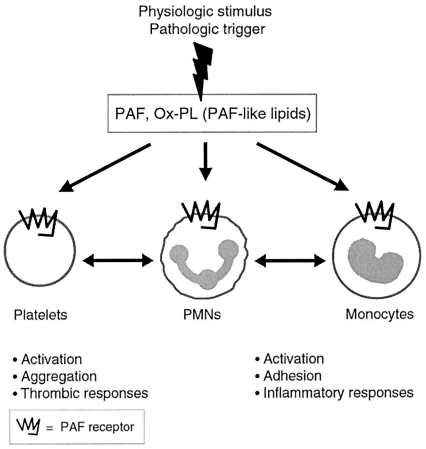


Figure 1. The platelet-activating factor (*PAF*) signaling system is a point of convergence and amplification of the thrombotic and inflammatory cascades. PAF and oxidatively modified phospholipids (*Ox-PLs*) are recognized by a signal-transducing receptor (PAF receptor) that is constitutively expressed by key cells of the hemostatic and innate immune systems: human platelets, neutrophils, and monocytes. Pathologic inflammatory and thrombotic responses can be triggered if mechanisms that generate PAF or other ligands recognized by the PAF receptor are dysregulated or impaired. Signaling through the PAF receptor can also indirectly induce procoagulant events. Because the PAF signaling system can induce, amplify, and mediate cross talk between hemostasis and inflammation, it is a point of convergence that may be critical when these cascades are induced in pathologic syndromes. *PMNs*, polymorphonuclear leukocytes.

mRNA for interleukin (IL)-1β is one of the transcripts that is translated in a rapid and sustained fashion. Signal-dependent translation of pro–IL-1β and processing of this precursor to the mature protein can be induced by PAF, thrombin, and certain other agonists (28). Newly synthesized IL-1β is released from activated platelets in microvesicles and accumulates in the fibrin matrix in a model of platelet-fibrin clot formation, suggesting that the platelet-fibrin thrombi may be reservoirs for cytokines and other inflammatory signaling molecules. In addition, IL-1β released from stimulated platelets induces human endothelial cells to become adhesive for PMNs (28), a response previously shown to be dependent on new gene expression and synthesis of E selectin and chemokines (39, 40). Thus, this sequence of events represents a new mechanism linking the thrombotic and inflammatory cascades (28) and adds to the ways in which PAF can mediate interplay between these systems (41). Because IL-1 induces

expression of a variety of genes in endothelial cells (42–44), additional functional alterations in the endothelium, including procoagulant and pro-inflammatory phenotypic changes, may be stimulated by its synthesis and release from activated platelets (28). In addition, IL-1 stimulation of endothelial cells induces genes that are involved in the switch from neutrophil accumulation to mononuclear leukocyte trafficking and chronic inflammation (43, 44).

PAF-Induced PMN Responses. The activation responses of myeloid leukocytes that are triggered by signals delivered through the PAF receptor are even more diverse than those in platelets. When PAF is rapidly produced and displayed on the surfaces of human endothelial cells that have been stimulated with thrombin and certain other agonists, it acts as a juxtacrine signal for spatially regulated activation and adhesion of neutrophils (1, 2, 23, 45). This provided the first evidence that human endothelium can synthesize and locally express signaling molecules for leukocytes (1, 23, 33). PAF has the potential to trigger a variety of responses in PMNs, whether it is locally displayed by endothelial cells in physiologic inflammation or synthesized by endothelium or other cells in a dysregulated fashion in pathologic syndromes (5, 41, 46). Activation of PMNs via the PAF receptor induces inside-out signaling of β_2 (CD11/CD18) integrins with consequent adhesiveness and aggregation, priming for enhanced inflammatory responses, polarization and directional migration, degranulation, and oxygen radical generation. Each of these effector activities is rapidly triggered with constitutive signal transduction systems (8). Priming for augmented release of granular factors may be a critical neutrophil response to PAF (13, 25) because granular enzymes, such as elastase, mediate inflammatory tissue injury and also have the capacity to induce coagulation when they are locally released (41, 47). It is likely that one or more of these PAF-mediated activation responses is particularly important in syndromes of inflammatory tissue damage, such as acute lung injury (13). In addition to these effector responses, which do not require new gene expression, PAF induces transcriptional events in PMNs; furthermore, it is a remarkably potent stimulus for signal-dependent translation of a subset of mRNAs in human neutrophils (S Lindemann et al., unpublished observations) (48). Thus, previously unrecognized activation responses of neutrophils continue to be identified, and some are induced by PAF.

PAF-Induced Monocyte Responses. Human monocytes also bear the PAF receptor on their plasma membranes and are activated when it is ligated (8). A critical function of these cells, which are ubiquitous in inflammatory syndromes ranging from sepsis to atherosclerosis (27, 49, 50), is synthesis of chemokines, cytokines, tissue factor, and other mediators. In addition to the PAF receptor, monocytes express P selectin glycoprotein ligand-1, which is a ligand for P selectin that mediates adhesive interactions with stimulated endothelial cells and platelets (Fig. 2). Human monocytes adherent to P selectin respond to PAF with enhanced translocation of nuclear factor-κB to the nucleus and dramatically increased synthesis of monocyte chemoattractant protein-1, IL-8, tumor necrosis factor-α, and other inflammatory gene products (27). Nuclear signaling and altered gene expression is prominent when monocytes establish stable adhesive interactions with thrombin-stimulated platelets (51), a cell-cell interaction that occurs in thrombosis and a variety of vascular syndromes (52).

PAF Synthesis by Critical Cells of the Hemostatic and Inflammatory Systems. An additional feature that indicates that PAF is an important mediator at points of convergence between the hemostatic and acute inflammatory cascades is its synthesis by critical cells that are involved in these systems. As outlined above, human endothelial cells rapidly synthesize PAF and use it as a juxtacrine signal for neutrophils (1, 2, 4–6, 23, 33, 41, 45). In this context, it acts coordinately with P selectin, which is rapidly translocated to the surfaces of inflamed endothelial cells from intracellular storage granules (Fig. 2) (2, 25, 45). These observations have been confirmed by many laboratories, and there is evidence that PAF at the cell surface can perform its signaling function *in vitro* and *in vivo* under flow conditions when leukocytes are subjected to shear forces (45). Notably, PAF synthesis by endothelial cells is stimulated by another pivotal hemostatic and inflammatory agonist: thrombin (23, 33, 53). A variety of other inflammatory mediators, oxidants, and bacterial toxins that induce thrombosis and inflammation in pathologic syndromes also induce PAF synthesis (8, 41, 45, 46). In addition, PAF is synthesized by adherent, activated platelets. A tethering and juxtacrine signaling system at the platelet surface analogous to that on the plasma membranes of endothelial cells mediates rolling and tight

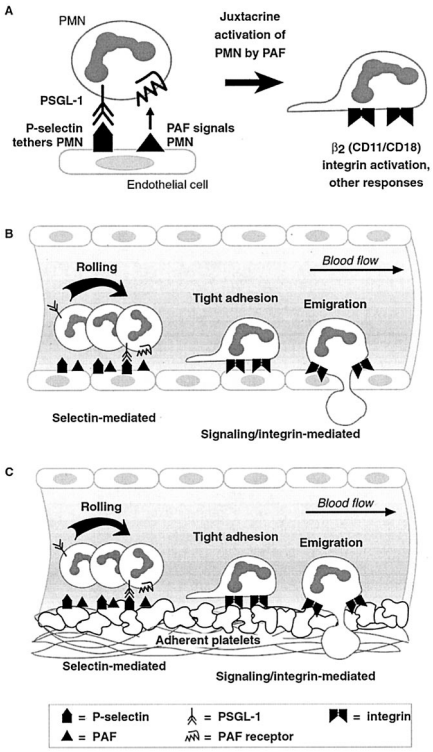
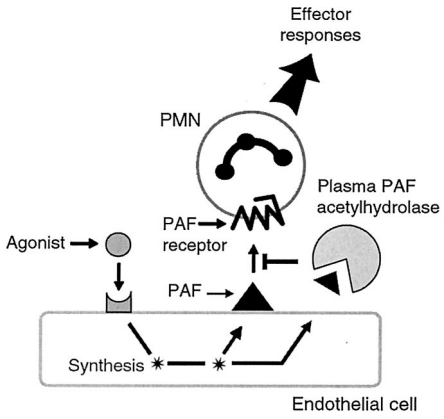


Figure 2. Platelet-activating factor (*PAF*) mediates juxtacrine signaling at the surfaces of inflamed endothelial cells and adherent, activated platelets. *A*, PAF mediates juxtacrine signaling of neutrophils *in vitro* when it is rapidly synthesized and translocated to the plasma membranes of human endothelial cells stimulated with thrombin. Neutrophil responses that are triggered by the PAF juxtacrine signal include activation of β_2 integrins, which contribute to tight adhesion of the leukocytes to the endothelial surface together with tethering initially provided by P selectin. Increased intracellular calcium, shape change and polarization, and priming for enhanced granular secretion and oxygen radical generation are additional effector responses that are induced via the PAF receptor. *B*, the P selectin–PAF juxtacrine system provides a molecular basis for rolling, tight adhesion, and emigration of neutrophils *in vivo* (45). Bottom, P selectin and PAF are displayed on the surfaces of adherent, activated platelets and mediate juxtacrine signaling, which triggers activation of β_2 integrins on the leukocytes in a fashion analogous to that occurring at the surfaces of thrombin-stimulated endothelial cells (*A* and *B*). This system also provides a molecular basis for rolling, tight adhesion, and localization of neutrophils. Juxtacrine signaling of neutrophils by PAF displayed by activated platelets is one of the mechanisms of cross talk between the thrombotic and inflammatory cascades that is mediated by the PAF signaling system. *PMN*, polymorphonuclear leukocyte; *PSGL-1*, P selectin glycoprotein ligand-1. Adapted with permission from Dixon et al (52).

adhesion of PMNs (Fig. 2) (45, 54, 55). Thus, considerable evidence indicates that PAF is strategically located and signals target cells at key sites in the molecular topography of inflammation and thrombosis by virtue of its synthesis by endothelial cells and platelets acting in response to relevant stimuli.

PAF MAY BE A CRITICAL MEDIATOR IN SHOCK, SEPSIS, AND TRAUMA

The synthesis of PAF is highly regulated. In addition, several other mechanisms have evolved to control its biological activities. These include spatial regulation of signaling (juxtacrine activation of leukocytes and other target cells by membrane-associated PAF), cell-specific expression of its receptor, receptor desensitization, and degradation of PAF by plasma and cellular hydrolases (Fig. 3). Evolution of a precise set of regulatory checks and balances for the PAF signaling system strongly indicates that it has physiologic roles. A corollary is that PAF, and related molecules that are recognized by the PAF receptor, may be mediators of injury if their generation is inappropriate or unregulated or if control mechanisms that limit their biological activities are inoperative or circumvented. Evidence that PAF has injurious actions in shock and syndromes of tissue damage is consistent with this idea. Injection of PAF into experimental animals causes hypotension and other features of anaphylactic or septic shock and induces sequelae that include gastric mucosal erosions, ischemic bowel necrosis, and cerebral ischemia (56). The latter observations suggest that PAF, or PAF-like lipids, contributes to multiple organ dysfunction or failure, in addition to shock states. Furthermore, PAF activity is increased in blood or tissue samples in some of these models, and PAF receptor antagonists ameliorate tissue injury in many of these experimental syndromes (56). Activation of target leukocytes via the PAF receptor may be a mechanism of injury in experimental hemorrhagic shock (57) and in models of anaphylaxis and other shock states (56).



- Tightly regulated synthetic pathways
- Receptors on specific target cells
- Receptor desensitization
- Juxtacrine activation of target cells by membrane-associated PAF (spatial regulation of signaling)
- Degradation by cellular and plasma PAF acetylhydrolases

Figure 3. The platelet-activating factor (*PAF*) signaling system is regulated by multiple control mechanisms. The plasma PAF acetylhydrolase is a particularly important regulator of the PAF signaling system because it limits the half-life of PAF to minutes in whole human blood and also terminates signaling upstream from the PAF receptor. Plasma PAF acetylhydrolase degrades PAF at cellular surfaces and in solution. *PMN*, polymorphonuclear leukocyte.

PAF and PAF-like Lipids as Mediators of Sepsis. Many observations in experimental animals suggest that PAF or PAF-like lipids are mediators of septic shock and the sepsis syndrome (56, 58). Among these is evidence that PAF contributes to acute sequestration of neutrophils and their adhesion to endothelial cells after endotoxin (lipopolysaccharide) administration (59) and to induction of nitric oxide synthase in experimental endotoxemia (60). A provocative observation is that overexpression of the PAF receptor increases lethality in response to lipopolysaccharide administration in mice (10).

In humans, an initial observation was the presence of intravascular PAF activity in children with sepsis (61). Another early study demonstrated that platelets from patients with sepsis have reduced numbers of binding sites for PAF, indicating receptor occupancy; in addition, there was increased bioactivity characteristic of PAF in samples from septic patients when compared with controls (62). In subsequent studies, increased PAF bioactivity was also reported in plasma samples from patients with bacteremia compared with blood from control subjects (63) and in samples from patients with septic shock and trauma (64). Plasma PAF acetylhydrolase (PAF AH) activity was reported to be reduced in critically ill patients with the clinical diagnosis of sepsis (65, 66). In addition, reduced PAF AH activity was correlated with multiple organ failure in patients with critical illnesses, some of whom had sepsis (67). Recently, neutrophils from septic patients were found to have substantially increased adhesion to immobilized platelets (68) under conditions in which PAF and P selectin are co-expressed (Fig. 2) (54, 55). Together, these findings suggest that PAF may be a critical mediator in human sepsis and its complications.

PAF Receptor Antagonists in Sepsis. Preliminary data from therapeutic trials with PAF receptor antagonists supported a role for the PAF signaling system in sepsis caused by Gram-negative organisms (69, 70). Although no regimen based on blockade of the PAF receptor that is clearly efficacious in human sepsis has subsequently emerged, neither have there been persuasive observations indicating that this strategy is not rational (71). However, it is possible that termination of signals upstream in the signaling cascade may be more effective than competing with PAF and PAF-like lipids downstream at the receptor level. Most receptor antagonists developed to date bind to the receptor with equal or lesser efficiency than PAF itself (8), necessitating a concentration of the antagonist several-fold higher than the natural ligand to reduce cellular effector responses to very low levels (i.e., $<50\%$, which may not be sufficient to yield a clinical benefit) or to block them entirely. This may be difficult to achieve in a sustained fashion in sick patients. In addition, the effective concentration of PAF may be very high at cellular surfaces where it acts in a juxtacrine fashion (Figs. 2 and 3), making competition by an antagonist more difficult.

OXIDIZED PHOSPHOLIPIDS: INFLAMMATORY PAF-LIKE SIGNALING MOLECULES THAT ARE GENERATED IN AN UNREGULATED FASHION

Enzymatic synthesis of PAF is not the only mechanism by which cellular activation triggered through the PAF receptor is induced; this feature of the PAF signaling system is important when considering its role in pathologic states. Oxidized, or oxidatively modified, phospholipids are generated by oxidant attack on hydrogen atoms adjacent to olefinic double bonds in unsaturated fatty acid side-chains (7). Oxidation of phosphatidylcholines, which are structural phospholipids in the membranes of all cells, generates a

large series of phospholipids in which the polyunsaturated fatty acid at the *sn-2* position, which is often arachidonic acid, is fragmented to shorter chain lengths. Some of these oxidized phospholipids have sufficiently short *sn-2* residues and other structural features that allow them to be recognized by the PAF receptor (7). Other compounds, including phospholipids that bind to nuclear peroxisome proliferator-activated receptors, are also produced (7). Analysis of the structural features and biological characteristics of the subset of oxidized phospholipids with PAF-like activity has been accomplished using neutrophils, monocytes, and other primary cells that bear the PAF receptor, heterologous cells expressing the cloned PAF receptor, and other strategies (7, 72). Together, these studies demonstrate that oxidized PAF-like lipids ligate the PAF receptor and trigger responses that are similar to those induced by PAF itself (G Marathe et al., unpublished observations), although the binding affinities of the oxidatively modified compounds vary depending on specific structural features of the individual oxidized phospholipid.

There is considerable evidence that oxidized phospholipids with PAF-like activity are produced in inflammatory and vascular conditions *in vivo* (7, 46, 72). A key point is that they are generated by free radical attacks on membrane phospholipids, which are unregulated reactions (7). This contrasts with enzymatic synthesis of PAF in endothelial cells and other cells in response to receptor-mediated agonists, oxidants, and other stimuli, which are highly regulated and often of short duration (5, 6, 41). In principle, oxidized phospholipids can be generated in high local concentrations on an ongoing basis if there is a continuous source of oxygen radicals and if membrane phospholipid precursors are not depleted. Thus, oxidized phospholipids with PAF-like activity may be equally or more important compared with PAF itself in ischemia-reperfusion and other consequences of shock and resuscitation and also in sustained inflammatory events that involve generation of oxygen radicals. High, local concentrations of these PAF-like ligands may be difficult to block with receptor antagonists in clinical syndromes for reasons outlined above. A second important point is that oxidized PAF-like phospholipids can be released from injured cells in membrane microvesicles (46, 73), a mechanism that disrupts spatially regulated juxtacrine signaling and may propagate intravascular activation of leukocytes and other target cells (4, 5, 41, 45, 52).

PLASMA PAF AH: A SIGNAL TERMINATOR

PAF AHs are enzymes that recognize PAF and PAF-like oxidized lipids as substrates. They cleave the short *sn-2* residues to yield products that are no longer recognized by the PAF receptor when they are present in physiologic concentrations (74). The preference for short acyl groups at the second position of the glycerol backbone prevents these enzymes from attacking "building block" phospholipids in cell membranes. A group of structurally diverse isoenzymes, including both intracellular and extracellular proteins, have this unusual substrate selectivity and other distinctive properties (74).

Plasma or Secreted Form of PAF AH. The plasma or secreted form of PAF AH (74, 75) has been called a signal terminator (76). The primary structure of plasma PAF AH is unique and includes only a small region of homology, a GXSXG motif, with esterases and other lipases. An active site triad, which was determined by site-directed mutagenesis, is critical for its catalytic properties and is similar to that in several neutral lipases (74). The plasma enzyme limits the half-life of PAF to a few minutes in whole human blood (77, 78). Plasma PAF AH cleaves oxidatively modified PAF-like lipids with an efficiency similar to that of PAF as a substrate (74). The secreted form of PAF AH is constitutively present in plasma in tight association with low- and high-density lipoproteins (74, 78).

Expression of Plasma PAF AH by Macrophages. Extensive biochemical and biological characterization and partial purification (74, 79) led to cloning of the enzyme (75). The discovery that the gene for plasma PAF AH is induced when human monocytes differentiate into macrophages in culture was among the critical observations of the cloning strategy (75, 80, 81). Subsequent observations confirmed that plasma PAF AH is produced by macrophages (6, 82), and studies of patients with allogenic bone marrow transplantation indicate that plasma PAF AH is largely derived from hematopoietic lineage cells (83). Thus, during inflammatory states, plasma PAF AH may be a marker for macrophage activation or expansion. Analysis of the promoter for the plasma PAF AH gene demonstrated that it contains response elements for inflammatory and myeloid-specific transcription factors and that it is transcriptionally regulated during macrophage differentiation and by mediators of inflammation (82). Hormonal stimuli, glucocorticoids, and PAF itself modulate expression of plasma PAF AH by macrophages (74, 82).

Plasma PAF AH as a Terminator of Inflammation. Many observations indicate that plasma PAF AH terminates signals by PAF and oxidized PAF–like lipids and thereby regulates inflammatory responses. These include *in vitro* experiments and *in vivo* observations in animal models and in humans (5, 6, 56, 84). Partially purified and recombinant forms of plasma PAF AH block inflammatory responses of human leukocytes to PAF (1, 55, 75, 84, 85) and inhibit responses to exogenous PAF in experimental animals (75), thus providing important proofs of principle. In addition, genetic, developmental, and acquired deficiency states of plasma PAF AH have been identified in humans and are correlated with severity or negative outcomes in inflammatory and thrombotic conditions that include asthma and cardiovascular syndromes, necrotizing enterocolitis, hemolytic uremic syndrome, and sepsis (Fig. 4) (5, 56, 84). Loss-of-function mutations leading to hereditary deficiency of plasma PAF AH activity occur, and molecular characterization of these variants has been accomplished (84, 86, 87). These mutations, together with naturally occurring polymorphisms that alter the catalytic properties of the enzyme, may contribute to the spectrum of severity in inflammatory syndromes such as asthma, atopy, and perhaps, other complex inflammatory diseases (84).

Decreased Plasma PAF AH Activity in Sepsis. Studies in different populations

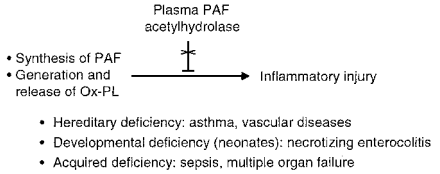


Figure 4. Plasma platelet-activating factor (*PAF*) acetylhydrolase is deficient in clinical syndromes of inflammation, thrombosis, and injury. Acquired deficiency of plasma PAF acetylhydrolase may be important in sepsis and multiple organ failure, in addition to genetic and developmental deficiency in other syndromes. *Ox-PL*, oxidatively modified phospholipid.

DOCKET ALARM

Explore Litigation Insights

Docket Alarm provides insights to develop a more informed litigation strategy and the peace of mind of knowing you're on top of things.

Real-Time Litigation Alerts

Keep your litigation team up-to-date with **real-time alerts** and advanced team management tools built for the enterprise, all while greatly reducing PACER spend.

Our comprehensive service means we can handle Federal, State, and Administrative courts across the country.

Advanced Docket Research

With over 230 million records, Docket Alarm's cloud-native docket research platform finds what other services can't. Coverage includes Federal, State, plus PTAB, TTAB, ITC and NLRB decisions, all in one place.

Identify arguments that have been successful in the past with full text, pinpoint searching. Link to case law cited within any court document via Fastcase.

Analytics At Your Fingertips

Learn what happened the last time a particular judge, opposing counsel or company faced cases similar to yours.

Advanced out-of-the-box PTAB and TTAB analytics are always at your fingertips.

API

Docket Alarm offers a powerful API (application programming interface) to developers that want to integrate case filings into their apps.

LAW FIRMS
Build custom dashboards for your attorneys and clients with live data direct from the court.

Automate many repetitive legal tasks like conflict checks, document management, and marketing.

FINANCIAL INSTITUTIONS
Litigation and bankruptcy checks for companies and debtors.

E-DISCOVERY AND LEGAL VENDORS
Sync your system to PACER to automate legal marketing.