materials of different activities. Knowledge of reaction mechanisms including self-organization processes on catalytic surfaces can be exploited through joining sets of microreactors coupled by diffusive transport of adsorbed molecules. Moreover, the role of promoters, a central topic in catalysis, is now accessible to detailed investigations.

The study of model catalytic and electrochemical systems also contributes to the general understanding of nonlinear dynamics. Major issues are the control of spatiotemporal chaos and the study and the control of the catalytic and electrochemical reaction dynamics. The suppression of chemical turbulence in the catalytic oxidation of carbon monoxide on a platinum single-crystal surface by global delayed feedback via the carbon monoxide partial pressure is an impressive example (*7*). Another is the synchronization and cluster formation in large sets of chaotic electrochemical oscillators consisting of nickel electrodes in sulfuric acid via global coupling through external resistors (*8*).

The analysis, simulation, and control of complex nonlinear processes benefit many areas of science and engineering. For example, interest in the dynamics of electrochemical systems has been revived because of progress in the analysis and simulation of spatiotemporal patterns, such as the inhomogeneous distribution of reaction currents (*9*). Spatiotemporal patterns form on electrode surfaces during electrochemical reactions because of a subtle interplay between electrode kinetics, the conductivity and composition of the electrolyte, the geometry of the electrochemical cell, and the external electric circuit. The nature of the patterns is determined by long-range coupling between reacting sites through the electric field. This coupling can be easily fine tuned, opening a whole new window toward the understanding of pattern formation. Recently, the adjustment of long-range inhibition by migration currents has led to the appearance of structures on the surface of an electrode that could be identified as Turing patterns (*10*). Such findings may even help to understand structure formation in biological systems with gradients in electrical potential.

A rich harvest of basic knowledge will continue to result from studies of the self-organization of surface reactions, as exemplified by the report by Sachs *et al.* (*1*). I expect to see practical applications within the first decade of the 21st century in many areas, including electrocatalysis in fuel cells, corrosion control, electrochemical machining of metals, and—most importantly—industrial and environmental catalysis.

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PERSPECTIVES: BIOMEDICINE

Clotting Factors Build Blood Vessels

Peter Carmeliet

For blood vessels to deliver oxygen to distant tissues, they must remain intact. When the vessel is injured, bleeding is stopped by clotting (coagulation) factors that form a thrombus (clot) of fibrin threads that trap platelet aggregates. The protease thrombin is essential for fibrin formation and platelet activation. Platelets become activated when thrombin binds to several protease-activated G protein-coupled receptors (PARs) expressed on their surface (*1*). One member of the PAR family, PAR1, is not expressed by mouse platelets—yet, mouse embryos lacking this receptor die of fatal bleeding. The question is why? Emerging evidence implicates the clotting system in the building and stabilization of new blood vessels (angiogenesis) during embryonic development. On page 1666 of this issue, Griffin *et al.* report that expression of PAR1 by endothelial cells rescues the fatal vessel fragility and bleeding of mouse embryos engineered to lack PAR1 (*2*). These findings emphasize the importance of thrombin and its receptors in angiogenesis, not only in the embryo but also in the adult, where angiogenesis has been implicated in numerous disorders including cancer and ischemic heart disease (*3*).

The primary task of the clotting system is to form blood clots composed of fibrin and platelet aggregates. Tissue factor, the initiator of blood coagulation, usually remains separate from blood and circulating clotting factors. It is expressed by smooth muscle cells in and surrounding blood vessels, and at low levels by blood cells or activated endothelial cells that line blood vessels. At sites of vascular injury, plasma coagulation factor VIIa (FVIIa) contacts extravascular tissue factor, thereby triggering the coagulation cascade (see the figure). Tissue factor is a cofactor for activation of factor X (FX) by FVIIa. Activated factor X (FXa) with the assistance of cofactor Va (FVa) then converts prothrombin to active thrombin, which converts circulating fibrinogen to fibrin. Not surprisingly, therefore, the absence of any of these clotting factors (TF, FVIIa, FVa, FXa, prothrombin, fibrinogen) predisposes animals to severe, often life-threatening, bleeding disorders (*4*, *5*).

Thrombin further enhances blood clot formation by activating circulating platelets, which then aggregate (see the figure). This clotting enzyme promotes activation of human platelets by cleaving the amino-terminal extracellular domain of PAR1 or PAR4, cleavage of either being sufficient to trigger platelet aggregation (*1*, *6*). PAR3 and PAR4 are expressed by mouse platelets but, curiously, PAR3 is an accessory cofactor for PAR4 activation at low thrombin concentrations (*7*). In contrast to the residual thrombin response in PAR3-deficient platelets, PAR4-deficient platelets lose all thrombin signaling, and PAR4-deficient mice, although viable, suffer prolonged bleeding. Remarkably, however, despite severe platelet defects, PAR4-deficient embryos develop normally, indicating that platelets are not essential for blood clotting (hemostasis) in the embryo. Moreover, genetic studies suggest that fibrinogen is also not required for embryonic hemostasis. As expected, loss of PAR2, which is not expressed by platelets, does not affect hemostasis (*6*). Surprisingly, as Griffin *et al.* report, loss of PAR1 (which is not expressed by mouse platelets) leads to fatal bleeding defects in a fraction of early mouse embryos (*2*). Yet,

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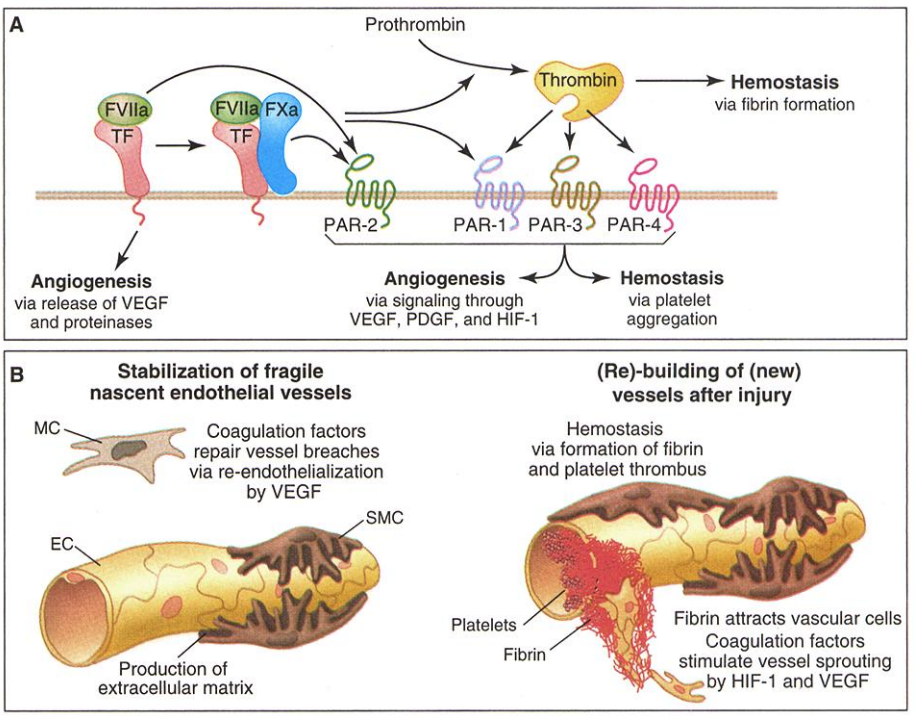
PAR1-deficient platelets aggregate normally in response to thrombin.

Why then do 50% of PAR1-deficient mouse embryos die of bleeding? As Griffin *et al.* demonstrate, the answer may lie in the involvement of coagulation factors in the building and stabilization of new blood vessels (see the figure). When blood vessels grow, they initially consist only of fragile endothelial cells. Stabilization of these naked endothelial vessels depends on the recruitment of smooth muscle precursor cells, which, upon differentiation, produce an extracellular matrix that strengthens and matures newly forming vessels. Vascular endothelial growth factor (VEGF) initiates endothelial growth, whereas platelet-derived growth factor (PDGF) and angiopoietin are essential for vessel wall stabilization (*3*).

Activation of angiogenic signaling pathways is an integral part of the cellular initiation of coagulation. Although normally absent from quiescent endothelium, tissue factor is expressed in angiogenic endothelium (for example, in tumor vessels). Tissue factor and FVIIa could stimulate angiogenesis by down-regulating the angiogenic inhibitor thrombospondin-1 and by up-regulating expression of VEGF, fibroblast growth factor–5 (FGF-5), proteinases (collagenases) and their receptors (u-PAR), as well as a number of other targets (*8*). Tissue factor is expressed constitutively in smooth muscle cells and together with FVIIa could—by amplifying the activity of PDGF—also assist in recruiting smooth muscle cells during vessel stabilization. FXa, a downstream product of the tissue factor/FVIIa pathway, also stimulates growth of smooth muscle cells. Apart from generating a provisional fibrin scaffold that attracts migrating vascular cells, thrombin could stimulate angiogenesis by activating PAR1, PAR2, and possibly PAR3 on endothelial cells. Thrombin increases the expression of VEGF receptors and nitric oxide (a downstream mediator of VEGF) by endothelial cells, promotes production and extravasation of matrix proteins, loosens endothelial cells from the extracellular matrix by up-regulating and activating metalloproteinases, and induces endothelial cell proliferation and migration (*1*, *6*). Thrombin may also recruit smooth muscle cells by up-regulating endothelial expression of PDGF (*1*, *6*). Once the naked endothelial vessels are covered, thrombin, by activating PAR1, PAR3, and PAR4, could stimulate smooth muscle cell differentiation and growth and regulate vessel tone. By activating the hypoxia-inducible transcription factor HIF-1α, thrombin triggers expression of a master transcription factor, which up-regulates production of numerous angiogenic molecules (*3*, *9*). In the adult, thrombin can further amplify angiogenesis by releasing VEGF, angiopoietin-1, and other angiogenic molecules from platelets. Thus, when coagulation is initiated, a cascade of angiogenic signals is also generated. Together, coagulation and angiogenesis could cooperate in stabilizing new blood vessels, repairing injured vessels, and stimulating the sprouting of new vessels (see the figure).



(A) Initiation of coagulation by tissue factor/FVIIa sequentially generates FXa, thrombin, and fibrin—all essential for clot formation. Activation of PARs by thrombin stimulates platelet aggregation. Coagulation factors also induce angiogenic signaling in vascular cells. Arrows indicate interactions between receptors and their ligands. **(B)** Possible coagulation factor involvement in angiogenesis and vessel stabilization. (Left) Fragile newly forming endothelial (EC) vessels are stabilized by recruitment of mesenchymal precursor cells (MC), which differentiate into contractile smooth muscle cells (SMC) and produce extracellular matrix (ECM). (Right) Upon vascular injury, coagulation stops bleeding (hemostasis) through formation of a fibrin and platelet thrombus. Coincidently, coagulation factors stimulate the repair of injured vessels and may contribute to the building of new vessels by activating HIF-1 and releasing VEGF. Fibrin provides a provisional matrix for migrating vascular cells during the formation of new blood vessels.

How do all of these molecules, generated during the initiation of coagulation, trigger angiogenesis? Emerging evidence suggests that tissue factor and FVIIa participate in angiogenic signaling through the involvement of tissue factor's intracellular tail (*10*). However, vascular development is normal in embryos expressing a truncated version of tissue factor without the intracellular domain (*11*). It is probable that proteolytic activity of FVIIa, independent of thrombin formation, is also involved in angiogenic signaling. Indeed, recent evidence indicates that PAR2, although activated by trypsin and a mast cell–derived tryptase outside the vessel wall, is likely to be activated by FVIIa in the vessel wall. However, activation in the vessel wall only happens when sufficient tissue factor and FXa are present—as is precisely the case for angiogenic endothelial cells (*12*). The tissue factor/FVIIa/FXa complex also efficiently activates PAR1 (*12*). Thus, even though thrombin is likely to be the primary activator of PAR1, upstream coagulation factors may also activate this thrombin receptor. Cross talk between distinct PARs on vascular cells may significantly expand the complexity and diversity of angiogenic signals. For example, PAR2 can be transactivated by cleaved PAR1, implying that, even though PAR2 is not capable of being activated by thrombin, it nonetheless may contribute to the thrombin response (*6*). Individual PARs have distinct activities—for example, only PAR2 is up-regulated by inflammatory mediators in endothelial cells, whereas activation of PAR1 selectively leads to smooth muscle contraction.

Because fibrin- and platelet-dependent hemostasis and angiogenesis are closely

intertwined, it has remained unclear whether bleeding in embryos lacking tissue factor, PAR1, or other coagulation factors results from defective hemostasis or blood vessel formation. Griffin *et al.* now provide some insights by demonstrating that loss of PAR1 does not prevent vessel formation per se but rather impairs the stabilization and maturation of newly forming vessels, thereby causing breaches and abnormal fragility in the vessel walls (*2*). By switching on expression of PAR1 in endothelial cells, these investigators were able to rescue PAR1-deficient mouse embryos from bleeding to death. This demonstrates that activation of PAR1 and its signaling pathway in endothelial cells is essential for vascular integrity. By stimulating changes in endothelial cell shape, migration, and growth through enhancement of VEGF-dependent signaling, PAR1 may facilitate repair of vessel wall breaches and secure vascular integrity. Although the Griffin study, at first glance, seems to indicate that PAR1 affects only endothelial cells, endothelial PAR1 may also indirectly affect the recruitment, differentiation, interaction, or stabilization of perivascular cells (pericytes) by release of PDGF or matrix proteinase. Considering that abnormal vascular fragility is also apparent in embryos lacking angiopoietin-1, its Tie2 receptor, and several other angiogenic molecules (*9*), the interaction between coagulation and these angiogenic molecules deserves further study. Whether the vascular fragility in PAR1-deficient embryos results from defective thrombin or tissue factor/FVIIa/FXa signaling remains to be determined. Indeed, tissue factor is involved in the stabilization of newly forming fragile vessels, because loss of tissue factor causes vessel fragility owing to defective recruitment of pericytes (*13*).

Considering the pleiotropic activity of thrombin on vascular cells and its ability to trigger crucial angiogenic signaling by VEGF and HIF-α, why does loss of PAR1 impair stabilization but not growth of new vessels in the embryo? Even more important, to what extent do these findings in the embryo extrapolate to the adult, where angiogenesis contributes to, and coagulation factors are up-regulated in, numerous disorders? Surviving PAR1-deficient mice exhibit normal re-endothelialization after vascular injury, but we do not know whether angiogenesis in pathological conditions is impaired in mice with inactivated PAR genes. Coagulation factors are likely to contribute to pathological angiogenesis because overexpression of tissue factor promotes tumor angiogenesis. Finally, even though coagulation factors and, in particular, PARs, have not been a direct target of pro- or anti-angiogenic drug development, they may become so in the future. Building stable vessels with PAR agonists would improve perfusion of ischemic tissues, whereas destabilizing vessels with PAR antagonists may suppress tumor growth. The observations of Griffin *et al.* should prime sufficient interest to ensure that these questions are answered in the near future.

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PERSPECTIVES: SOCIAL SCIENCE AND ECOLOGY

Networking Tips for Social Scientists and Ecologists

Sean M. McMahon, Kevin H. Miller, Jim Drake

For the past 30 years, a subdiscipline of the social sciences known as social network analysis has developed structural models to analyze human interactions (*1*). In social network analysis, discrete mathematics and statistics are combined with the emerging epistemology of complex systems to explore processes and phenomena as diverse as the diffusion of information through an organization, the adoption of innovations in society, and the spread of infectious disease in a population. Scientists working on social network analysis draw upon myriad disciplines: sociology, anthropology, psychology, geography, mathematics, statistics, and computer science. Like social network analysis, analyses of trophic structure in ecological communities and of energy flow and nutrient transfer through ecosystems incur the problem of how to conceptualize and test interactions within these complex systems. The striking similarities between social networks and biological communities suggest that there exist constraining or structuring forces common to both. Social and ecological networks also share the need to reduce the elements and interactions of the network to an order simple enough to analyze, yet complex enough to reflect reality.

Simplifying complex system analysis was just one of the many topics discussed at an NSF-sponsored workshop on network theory and biocomplexity held at the Duke University Marine Laboratory in North Carolina (*2*). Computer scientist Stephen Seidman (Colorado State University) explained the social science concept of "cohesion" (how tightly knit a group of people is) and the benefits of graph theory for delimiting nested networks in order to better understand influences within complex systems. The concept of cohesion parallels that of "guilds" (*3*) or "trophospecies" (*4*) in ecology, where organisms that eat and are eaten by similar species are treated as one group. Steven Borgatti (Boston College) and Martin Everett (University of Greenwich) discussed the concept and mathematical application of "regular equivalence," an approach to social networks that provides a formal model defining the notion of social roles. An example of regular equivalents would be two doctors at different hospitals. Although they do not see the same patients, or interact with the same suppliers, nurses, and administrators, they have similar interactions with equivalent others, and thus play the same role. Furthermore, their equivalence is decided not by their work or credentials per se, but by the relationships they have with other members of the network. Joseph Luczkovich (East Carolina University), an ecologist, demonstrated how regular equivalence, when applied to ecological communities, can help ecologists partition species into groups that play the same roles even if, unlike trophic guilds, they do not consume the same prey or share the same predators.

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