subunits, which increase the zymogen's plasma half-life but must be dissociated for full enzyme activity. Platelet factor XIII, by contrast, is a dimer that contains only the two α subunits. Both forms of the zymogen require thrombin cleavage and fibrin as a cofactor, but plasma factor XIII activation proceeds at a considerably slower rate owing to the need for dissociation of the β subunits. Thrombin-activated factor XIIIa binds to fibrin and cross-links the fibrin units, thereby rendering them less permeable and more resistant to lysis. Furthermore, factor XIIIa cross-links the major plasmin inhibitor, α_2-antiplasmin, directly to fibrin, positioning it for neutralization of any invading plasmin.

Fibrinolysis

The fibrinolytic system operates to prevent fibrin from occluding healthy vessels. During clot formation, Xa and thrombin stimulate healthy ECs to release tissue-type plasminogen activator (t-PA) and urokinase-type plasminogen activator (u-PA), both of which are capable of cleaving plasminogen into plasmin. The vast excess of plasminogen in the plasma dictates that under normal circumstances, the concentration of these enzymes is rate-limiting for plasmin formation. The kinetic efficiency of t-PA is improved by at least an order of magnitude by the presence of fibrin. This helps to keep t-PA most active in the microenvironment of the clot. By contrast, u-PA appears to require binding to activated platelets for its ability to liberate plasmin.

Acting to contain fibrinolysis are plasma mediators that either inactivate formed plasmin (such as α_2-antiplasmin and possibly α_2-macroglobulin) or block plasmin formation. Foremost among the latter is PAI-1. PAI-1 is present in several-fold molar excess in the plasma and is also released by activated platelets, thereby protecting clots from premature lysis. Plasma levels of PAI-1 can be highly variable, in part because of its circadian pattern of secretion, but also because of polymorphisms of the PAI-1 gene. The 4G promoter region polymorphism of PAI-1 is associated with higher PAI-1 levels and a higher risk for thromboembolic disease (see Chapter 52).

Another mediator that limits fibrinolysis in the vicinity of the clot is thrombin activator fibrinolysis inhibitor (TAFI). TAFI is synthesized in an inactive form by the liver and circulates in the plasma, possibly in a complex with plasminogen. TAFI cleaves specific fibrin lysine residues that would otherwise promote binding of fibrinolytic enzymes (e.g., plasmin). TAFI requires either plasmin or thrombin for activation; however, thrombin activation of TAFI requires extraordinarily large amounts of *free* thrombin. By contrast, EC-associated thrombomodulin increases thrombin-induced TAFI activation 1250-fold, making this an essential cofactor and one that is predominantly available only at the interface between the blood and the vessel wall.

In addition to the EC surface, macrophages are also critical to fibrinolysis. Macrophages degrade fibrin clot through lysosomal proteolysis by a plasmin-independent mechanism. The macrophage binds to fibrin and fibrinogen through its surface integrin receptor, CD11b/CD18; this binding is followed by internalization of the complex into the lysosome, where fibrin and fibrinogen are degraded.

Tissue repair and regeneration are the physiologic end points of clotting, and they eventually lead to dissolution of the fibrin-based clot. Besides t-PA and urokinase, the intrinsic pathway activators kallikrein, XIIa, and XIa also generate active plasmin from plasminogen. Plasminogen binding to cell surface receptors promotes its own activation to plasmin by placing it in proximity to t-PA and fibrin clot and protects plasmin from inactivation by circulating (not clot-bound) α_2-antiplasmin (see Fig. 50-4). Plasmin eventually dissolves the fibrin matrix to produce soluble fibrin peptides and D-dimer; it also activates metalloproteinases that further degrade damaged tissue. Fibroblasts and leukocytes migrate into the wound, the latter mediated by selectin binding, and these inflammatory cells act in concert with growth factors secreted by leukocytes and activated platelets (e.g., transforming growth factor-β) to enhance vascular repair and tissue regeneration.

Laboratory Testing of Coagulation

For purposes of laboratory testing, the coagulation cascade is artificially divided into extrinsic and intrinsic pathways, measured as PT and PTT, respectively. These converge to form a common pathway that leads to thrombin and fibrin generation (see Fig. 50-2A). In the laboratory, the *extrinsic* pathway (PT) is assessed by measuring the interaction of circulating VIIa with exogenously added TF (also known as *thromboplastin*). The PT is highly sensitive to deficiencies of factors II, VII, V, and X, all of which are associated with significant bleeding. Because II, VII, and X are also vitamin K dependent factors, with VII having the shortest circulating half-life, the PT is currently the best test for monitoring warfarin (Coumadin) therapy. The PT is unaffected by intrinsic pathway deficiencies of XII, XI, IX, or VIII. The degree of prolongation of the PT by warfarin depends on the strength of the particular thromboplastin (based on its international sensitivity index [ISI]) and the specific coagulation instrument used for the assay. The international normalized ratio (INR) takes these factors into account to standardize among laboratories for variations in prolongation of the PT induced by warfarin. The INR is calculated for each patient as follows: $(\text{patient PT}/\text{mean control PT})^{\text{ISI}}$. Therapeutic INRs with warfarin allow for global application of anticoagulant recommendations; these vary according to the specific disease indication and are covered in Chapter 51. In contrast, the PT is relatively insensitive to therapeutic anticoagulation with unfractionated heparin except in very high doses.

The aPTT measurement is based on in vitro contact activation (e.g., plasma stimulation with a negatively charged compound such as kaolin). The aPTT is sensitive to deficiencies of contact factors—prekallikrein (PK), high-molecular-weight kininogen (HMWK), and factor XII—and to deficiencies of coagulation factors of the intrinsic pathway (XI, IX, and VIII) and the common pathway (V, X, and prothrombin). Deficiencies of PK, HMWK, and XII prolong the aPTT but do not result in clinical bleeding, implying that these factors are irrelevant to physiologic hemostasis. By contrast, severe deficiencies of XI, and especially IX and VIII, cause significant bleeding. The aPTT is also highly sensitive to unfractionated heparin and is used to monitor therapeutic heparin anticoagulation. Compared with the INR for anticoagulation with warfarin, the range for therapeutic aPTT levels with unfractionated heparin is much wider and not as easily standardized. Therapeutic unfractionated heparin levels can be measured by sensitive assays of anti-Xa activity; therapeutic levels of