Anticoagulation and Thrombolytic Therapy*
Practical Considerations
Craig M. Kessler, M.D.

Although thrombolytic agents have been available for over 10 years and have demonstrated safety and efficacy in an increasing number of clinical conditions involving thrombotic phenomena, their general acceptance as first-line therapeutic agents in medical management has been slow. Much of the reluctance to use these drugs is based on their associated incidence of hemorrhagic complications, which is several-fold greater than with use of conventional anticoagulants. With the introduction of second-generation thrombolytic agents, made possible through successes in recombinant DNA technology and chemical modifications of previously available compounds, increased fibrin specificity has been achieved and has been translated into increased clinical efficacy and safety. These results will likely improve as innovative regimens using dose modifications or multiple agents with combined synergy are developed. An appreciation of basic coagulation and the pharmacology of anticoagulants and thrombolytic agents is necessary to maximize the benefits of currently available medications, to develop new treatment strategies, and to minimize potential hemorrhagic complications.

The therapeutic armamentarium for the medical management of diseases with thromboembolic manifestations has rapidly expanded, primarily owing to the recent successful clinical applications of innovative pharmacologic agents produced through recombinant DNA technology. An understanding of the coagulation process and an appreciation of the clinical pharmacology of potentially useful antithrombotic and thrombolytic medications are imperative for the formulation of the optimal strategy for the treatment of these disorders.

**Platelets**

The processes of platelet adherence and aggregation are initiated when the continuity of the vessel wall is disrupted with exposure of subendothelial matrix structures, such as collagen (Fig 1).1,2 Recently, amorphous electron-dense substance (AEDS) has been reported to serve as an indispensable contact-mediating prerequisite for adhesion of platelets around damaged endothelial cells.3 The release of AEDS from damaged endothelium is enhanced by intrinsic coagulation pathway activity and can be inhibited by antplatelet-aggregating agents, anticoagulants, and thrombolytic agents.4 Platelet adhesion- vessel wall interactions are mediated by the large molecular weight multimers of von Willebrand factor protein (vWF),5 which is synthesized by megakaryocytes and endothelial cells.6 vWF is present in the subendothelial matrix7 and in the platelet α granule8 and circulates in the plasma as the noncovalently complexed carrier of factor VIII coagulant protein.9 At high shear rates, vWF “bridges” platelets to sites of vascular damage by binding directly to platelet membrane glycoproteins Ib and IIb-IIIa and to subendothelial components.10,11 vWF also may mediate the development of atherosclerosis in vivo; its absence in the plasma, platelets and vessel walls of pigs with von Willebrand’s disease has been associated with a resistance to dietary induced atherosclerosis.12,13 Theoretically, atherogenesis requires platelet deposition at sites of vascular injury or plaque formation with the subsequent release of platelet contents, which in turn function as agonists to initiate and potentiate further platelet aggregation (ADP and serotonin from the dense granules and thromboxane A2 [TXA2] generated from arachidonic acid metabolism) and to stimulate vascular smooth muscle and fibroblast proliferation (platelet-derived growth factor from the α granule). These processes would not be expected to proceed efficiently without vWF; however, the physiologic relevance of vWF for these processes remains to be established in humans.

Platelet aggregation is propagated by the recruitment of circulating platelets to form a platelet plug to minimize blood loss (Fig 1). Local vasoconstriction mediated by the release of serotonin and TXA2 also reinforces the functional integrity of the platelet plug, as does the exposure of platelet membrane derived phospholipids (platelet factor 3), which ultimately function as a template for catalyzing the generation of activated factor X (Fig 1) and localizing the formation of thrombin.

Thrombin, adenosine diphosphate (ADP), epinephrine, and collagen are agonists of platelet aggregation and are employed in the in vitro evaluation of platelet function by platelet aggregometry. These agents activate platelets by releasing membrane-bound phospholipases, which in turn initiate arachidonic acid metabolism to generate unstable vasoactive prostaglandins (Fig 2). In the platelet, thromboxane A2 is produced by cyclooxygenase metabolism of arachidonic acid and exerts potent aggregatory and vasoconstrictor properties. The same pathway exists in the endothelial cell except that the enzyme thromboxane synthetase is present in relatively insignificant quantities compared with prostacyclin synthetase, which mediates the formation of prostacyclin (PGL2), a very potent vasodilator and antiplatelet aggregatory substance. Cyclooxygenase activity can be inhibited irreversibly and almost completely by low doses of acetylsalicylic acid (aspirin)14,15 (≤2 mg/kg)16,17 and reversibly by nonsteroidal anti-inflammatory drugs (NSAIDs) and sulfinpyrazone (Fig 2).18 The aspirin effect lasts for the life span of the platelet and in the absence of de novo TXA2 production and cyclooxygenase regeneration, a qualitative

---

*From the Division of Hematology-Oncology, George Washington University Medical Center, Washington, D.C.
platelet defect will persist. Endothelial sources of cyclooxygenase appear to be less sensitive to aspirin, and even the complete inhibition of cyclooxygenase activity by large quantities of aspirin can be overcome eventually by regeneration of the enzyme. However, animal studies suggest that large amounts of aspirin (150 mg/kg) may produce a temporary hypercoagulable state, since the absence of the antiplatelet aggregatory effects of PGI₂ promotes the accumulation of platelet aggregates on damaged vessel walls. This phenomenon may be exacerbated by the relative inability of aspirin to inhibit thrombin-induced platelet aggregation. These observations have important implications for clinical trials that assess aspirin regimens in the primary or secondary prevention of coronary or cerebrovascular disease. The inability to detect biologically important benefits in these studies may be due to the high and frequent doses of aspirin. On the other hand, the safety margin of aspirin for clinical use as an antithrombotic agent has recently become a critical issue in the Physicians Health Study, in which the use of small doses of buffered aspirin (325 mg every other day) dramatically reduced the incidence of nonfatal and fatal MI but was associated with an increase (albeit not statistically significant) in disabling intracerebral hemorrhagic events. Lower doses of aspirin could adequately prevent TXA₂ generation, minimize the platelet contribution to atherogenesis and coronary and cerebrovascular thromboembolic processes, and reduce the incidence of clinically important bleeding.

Dipyridamole is a relatively weak inhibitor of platelet aggregation and adhesion, and when administered in combination with aspirin or warfarin, prolongs the shortened platelet survival associated with prosthetic heart valves and arterial thrombotic disease. Its mechanism of action remains unclear, although its inhibition of platelet phosphodiesterase in vitro increases cAMP levels, an event associated with reduced platelet aggregation and adhesion. Dipyridamole and prostacyclin possess many of the same pharmacologic actions and may be synergistic in vivo.

In the future, platelet aggregation and adhesion may be
THE COAGULATION CASCADE

pharmacologically modified by specific inhibitors of thromboxane and prostacyclin synthetase, by monoclonal antibodies directed against platelet membrane glycoproteins IIb-IIIa and Ib, by agents that block TXA₄ receptor sites, and by synthetic analogs of prostacyclin.

The platelet function of aggregation can be monitored in vitro by platelet aggregometry following the addition of various agonists (ADP, epinephrine, and collagen) to platelet-rich plasma. The resultant tracing patterns and changes in light transmission represent platelet activation, release, and aggregation. The tracings produced by the addition of ristocetin reflect platelet agglutination mediated by vWF rather than aggregation, although high concentrations of ristocetin can induce aggregation. Aspirin, nonsteroidal anti-inflammatory agents, dipyrindamide, β-blockers, and prostacyclin all can inhibit in vitro platelet aggregation induced by conventional agonists. Thrombin- and ristocetin-induced platelet aggregation is not consistently blocked by aspirin.

In vitro testing of platelet function is accomplished with the template bleeding time, a standardized procedure that depends on platelet number, aggregation function, platelet interaction with subendothelial components (adhesion), and the presence of normal subendothelial matrix collagen. Prolonged bleeding times are usually observed following the use of aspirin, NSAIDs (except sulfinpyrazone), and prostacyclin, but not with dipyrindamide, despite its hypothesized effects on platelet adhesion and aggregation.

In vitro platelet activation can be monitored specifically and sensitively by measuring urinary or plasma levels of released α granule constituents (β-thromboglobulin and platelet factor 4) and generated TXB₂, the stable metabolite of TXA₄. Elevated levels of prostacyclin metabolites are produced in disease states affecting endothelial cell integrity and viability⁶⁹ and in patients with evidence of platelet activation in vitro.⁶⁹

THE INTRINSIC AND EXTRINSIC PATHWAYS

Replacement of the initial hemostatic platelet plug requires the participation of the fluid phase of blood coagulation via the intrinsic and extrinsic pathways to form thrombin in the final common pathway and eventually generate fibrin (Fig 3). Normally, this process is localized to regions of damaged endothelium and is initiated by the activated platelets within the platelet plug by exposing their phospholipid surfaces to serve as templates on which the coagulant proteins can concentrate, amplify, and modulate their sequential proteolytic reactions. These complex proteolytic sequences compose the coagulation cascade, which traditionally has been separated into the intrinsic and extrinsic pathways. Although in vitro data and clinical observations indicate that some of the coagulant proteins may not participate exclusively within the pathway to which they have been designated (ie, factor XIIa can activate factor VII; factor VIIa can activate factor IX), the laboratory assays used to screen coagulation pathway activity remain essential in localizing deficiencies and formulating therapeutic decisions.

The intrinsic pathway is triggered when factor XII is activated by the high negative surface charges of exposed subendothelial collagen in vitro and ellagic acid or celite in vitro. The activation of factors XII and XI is accelerated by high molecular weight kininogen and prekallikrein.

The intrinsic and extrinsic pathways merge at the point of factor Xa activation, which can be achieved either by the proteolytic effects of a factor IX, factor VIII, Ca²⁺, and phospholipid complex, or by the action of a multimolecular complex of factor VII, Ca²⁺, and tissue thromboplastin derived from damaged extravascular ("extrinsic") cells. Thrombin, a pivotal protease for the promotion and modulation of coagulation (Fig 4), is generated by the common pathway and cleaves fibrinopeptides A and B from the α and
β chains of fibrinogen (factor I) to produce fibrin monomers (Fig 5). These monomers rapidly form an unstable polymerized complex, which is crosslinked in the presence of Ca²⁺ and factor XIIIa to form the stable tensile clot.

A moderate or severe deficiency of any coagulation factor (except factor XII) potentially will produce clinically significant bleeding. The prothrombin time (PT) measures the combined activities of components in the extrinsic and common pathways, but is most sensitive to factor VII deficiency and least sensitive to decreases in prothrombin (factor II) and fibrinogen. The activated partial thromboplastin time (APTT) monitors the activity of the intrinsic and common pathways but is most sensitive to abnormalities in the intrinsic system. The thrombin time exclusively measures the conversion of fibrinogen to fibrin by thrombin.

Decreased fibrinogen concentrations (≤ 80 mg/dl), dysfibrinogenemias, circulating fibrinogen/fibrin degradation products, myeloma proteins, and heparin prolong the PTT by interfering with thrombin activity directly, (heparin) by blocking fibrinopeptide cleavage, or by impeding fibrin polymerization. Coagulation cascade activation can be detected by measurement of fibrinopeptides A (thrombin generation) and the prothrombin activation fragment F₁⁺₂ (factor Xa generation) (Fig 3).

Coagulation activity is modulated most effectively by naturally circulating inhibitors of serine proteases (antithrombin III) and inhibitors of factors V and VIII (protein C and its cofactor protein S). The congenital or acquired deficiency of antithrombin III, protein C, or protein S is associated with a predisposition to recurrent venous...

**Figure 4.** The pivotal role of thrombin in the propagation and modulation of coagulation.

**Figure 5.** The effect of thrombin generation and fibrinolysis on fibrinogen and fibrin.
Fibrinolysis

![Fibrinolysis Diagram](http://journal.publications.chestnet.org/pdaccess.ashx?url=/data/journals/chest/21594/)

**Figure 6.** Fibrinolysis is achieved by activators of plasminogen intrinsic to plasma including thrombin and activated factor XII (XIIa). Activators of fibrinolysis extrinsic to plasma include stimuli that culminate in release of tissue plasminogen activator (tPA) from vascular endothelium.

thrombosis\(^{39-40}\) and, rarely, to arterial thrombosis.\(^{30-31}\) Antithrombin III inhibits the serine protease enzymes of the intrinsic pathway, specifically factors IIa (thrombin), IXa, Xa, XIa, and XIIa, by forming a 1:1 stoichiometric complex. Complex formation and subsequent inhibition of coagulation factor activity are physiologically insignificant in the absence of heparin but are accelerated 1,000-fold with heparin, which induces a conformational change in the antithrombin III molecule to expose an arginine-reactive binding site. The physiologic correlates of heparin in *vitro* are presumed to be heparin-like proteoglycans synthesized and secreted by endothelial cells.\(^{36}\) These species exhibit anticoagulant activity and can activate antithrombin III adjacent to its specific endothelial cell receptor sites.\(^{38,39}\)

Protein C is a vitamin K-dependent protein that is activated by thrombin bound to its endothelial surface receptor cofactor, thrombomodulin. Activated protein C (protein C\(_a\)), in turn, complexes with its vitamin K-dependent coenzyme protein S on endothelial and platelet surfaces and inactivates the coagulation cascade at the levels of factors Va and VIIIa. Protein C\(_a\) can also stimulate fibrinolysis, presumably by inactivating the specific inhibitor of tPA (Fig 6).

**Heparin**

Heparin and warfarin are the two agents most commonly used for the treatment and prophylaxis of thrombotic diseases. They share bleeding as the most prevalently observed complication. The conventionally available unfractionated heparin is a highly anionic mucopolysaccharide extracted from porcine and bovine alimentary tract and bovine lung and has a mean mol w of 12-15,000. Its antithrombotic properties are related to its capacity to catalyze the inhibition of serine protease clotting factors by antithrombin III. The antithrombin III-heparin complex has particular affinity for factor Xa, which is the pivotal clotting protein in the common pathway of coagulation (Fig 3). Less heparin is required to neutralize factor Xa activity and thus prevent thrombin generation than is necessary to neutralize the thrombin produced once the thrombotic process is initiated. This observation forms the rationale for the prophylactic antithrombic regimens in which heparin is administered subcutaneously in very low doses.

In *vitro* investigations indicate that thrombotic events are propagated by continuous incorporation of new fibrin, even during physiologic or pharmacologic fibrinolysis.\(^{34-36}\) Heparin impedes the formation and deposition of new fibrin,\(^{27}\) and therefore, pretreatment or simultaneous administration of heparin with thrombolytic agents may greatly enhance therapeutic efficacy by preventing thrombus extension or reocclusion. In addition, heparin by itself may enhance fibrinolysis in *vitro*\(^{40}\) and in experimental animal models.\(^{41,42}\) Whether this intrinsic property of heparin is physiologically important in humans has not been established; however, spontaneous thrombolysis with reduction of the size of acute deep venous thromboses has occasionally been documented with heparin anticoagulation alone.\(^{44,46}\)

Paradoxically, heparin anticoagulation may also be associated with propagation of recently formed thrombi.\(^{46}\) This might be due to the consumption of antithrombin III and/or plasminogen by the thrombus, or it might reflect the platelet proaggregatory properties of conventional heparin preparations.\(^{46-47}\)

In *vitro* platelet aggregation has also been invoked as the initiating mechanism of heparin-associated thrombocytopenia (HAT), which usually occurs within the first 3-7 days of heparin anticoagulation therapy\(^{48,49}\) and may be associated with paradoxical major venous or arterial thrombosis.\(^{50,51}\) The thrombocytopenia is variable in severity and may be immune mediated,\(^{52-54}\) although certain clinical observations appear to dispute this. For example, patients may develop severe HAT acutely without having been previously exposed to heparin; notably decreased platelet counts occasionally normalize despite continuation of heparin; some patients may receive heparin on future occasions without developing thrombocytopenia; and HAT may be heparin lot related.\(^{55}\) HAT is most commonly encountered during the administration of bovine lung-derived heparin; porcine intestinal...
mucosal heparin and calcium heparin have been implicated in 3-5% of cases. This suggests that HAT may be related to the presence of the higher molecular weight subfractions of unfractionated heparin, i.e., the subfractions which enhance platelet aggregation but which participate insignificantly in antithrombin III interactions. Porcine intestinal mucosal heparin has a greater proportion of low molecular weight subfractions than bovine source heparin; highly purified low molecular weight heparin preparations, i.e., mean molecular weight between 4,000 and 6,000, have insignificant platelet-enhancing properties and have not been associated with HAT.

The diagnosis of HAT is confirmed by *in vitro* platelet aggregation studies, in which spontaneous platelet aggregation occurs in patient platelet-rich plasma (PRP) following the addition of heparin alone or in normal PRP to which patient platelet-poor plasma or heat-treated serum is added with exogenous heparin. Treatment consists of discontinuing the unfractionated heparin and substituting a porcine source heparin, low molecular weight heparin preparations, or oral warfarin. Aspirin, prostacyclin analogs, and the defibrinatory enzyme anlodon have also been beneficial in the treatment of HAT.

The frequency of spontaneous bleeding complications associated with heparin administration ranges as high as 20-30% when strict selection criteria are not applied, and is proportional to dose and prolongation of the PTT to greater than 2½-3 times control values. Concomitant use of aspirin, thrombolytic agents, and NSAIDs, as well as advanced age, underlying anatomic lesions, invasive therapeutic or diagnostic procedures, and other coagulation defects will increase the risk of untoward hemorrhage. Life-threatening bleeds may necessitate rapid reversal of the heparin effect by administration of protamine sulfate. One milligram of protamine sulfate neutralizes approximately 100 units of heparin; the administered dose of protamine sulfate should be equivalent to one half of the previous heparin dose, infused slowly (≤5 mg/min). Excessive doses of protamine sulfate can also act as an anticoagulant. Transfusions of fresh-frozen plasma or whole blood are not always effective neutralizers, since heparin functions as a circulating inhibitor of coagulation.

Laboratory monitoring of heparin therapy is usually achieved with the PTT and less commonly with the Lee-White whole blood clotting time. The PTT is relatively insensitive to heparin effects except in high concentrations. Heparin doses are adjusted to maintain the PTT at 1.5-2.5 times the control and the Lee-White clotting time at 20-45 min. The PTT may be minimally affected or unaffected by minidose heparin regimens (5,000 units subcutaneously every 8-12 h). Conventional heparin administration significantly prolongs the thrombin time results. In determining the etiology of abnormal coagulation tests, mixing normal plasma with patient plasma will not correct the PTT in the presence of heparin. The PTT produced by heparin will correct following addition of protamine sulfate or toluidine blue, and the reptilase time will be normal. When reptilase, a snake venom that cleaves only fibrinopeptide A from fibrinogen to form a fibrin clot, is substituted for thrombin in the thrombin time, it is not inhibited by heparin but may be inhibited by circulating fibrinogen/fibrin split products, dysfibrinogens, and abnormal qualitative and quantitative immunoglobulins. The PTT and thrombin time are only partially sensitive to low molecular weight heparin. Laboratory assessment of anticoagulant adequacy during clinical administration of these preparations is best achieved with measurement of anti-factor Xa levels.

Osteoporosis and hyposalteronism are additional toxic effects of heparin administration but are rarely observed. Heparin is also associated with an increased incidence of fetal demise (20%) and prematurity (~14%) when used during pregnancy; however, in contrast to warfarin sodium (Coumadin), no teratogenicity or transplacental transport has been observed and heparin remains the anticoagulant of choice during pregnancy. In addition, because heparin is not present in breast milk, heparinized mothers may nurse. Mothers taking warfarin can nurse as well, because the levels of warfarin in breast milk are not associated with clinical problems in the infant.

Recent attention has been focused on the clinical role of heparin as a bifunctional regulator of vascular wall integrity. Both unfractionated and low molecular weight heparin preparations can abolish the smooth muscle cell hyperplasia that follows endothelial cell injury. This antiproliferative effect is independent of antithrombin III interactions and anticoagulant activity and may modulate the formation of atheromatous lesions. Further, heparin may initiate reparative processes by stimulating the growth of endothelial cells in the presence of fibroblast growth factor. The combined anticoagulant and antiproliferative effects of heparins theoretically should potentiate the benefits achieved by angioplasty procedures and thrombolytic therapy. Low molecular weight heparin preparations, some of which have been associated with significantly fewer bleeding complications than unfractionated standard heparin, may be particularly useful in this regard.

**Oral Anticoagulation**

Oral anticoagulation is accomplished by using congeners of dicumarol, which inhibit coagulation through their actions on vitamin K. Factors II, VII, IX, X, and protein C and protein S require vitamin K, reduced to its hydroquinone form to mediate the intrahepatic carboxylation of γ-glutamic acid residues on their inertzymogens (Fig 7). This postribosomal modification provides binding sites for divalent calcium ions, which, in turn, promote binding of the coagulant zymogens to phospholipid surfaces for subsequent activation. Coumadin, the most commonly used oral anticoagulant in the United States, competitively inhibits vitamin K epoxide reductase activity in the vitamin K cycle, and thus prevents the regeneration of the active reduced hydroquinone form of vitamin K, from its inactive quinone form, vitamin K, epoxide. Therefore, the anticoagulant effect associated with warfarin sodium is produced by altering the structure of biologically inactive precursor coagulant proteins rather than by depressing their hepatic synthesis. These so-called PIVKA (protein induced by vitamin K antagonists) are antigicnically similar to their activated forms.

Warfarin sodium is rapidly and almost completely absorbed from the alimentary tract and is almost entirely bound to albumin in plasma (~99%). Because the unbound
Figure 7. The mechanism of action of warfarin. Warfarin inhibits the reduction of the oxidized storage form of vitamin K. Vitamin K$_2$H$_2$ functions as a coenzyme in the carboxylation of factors II, VII, IX, X, and protein C and protein S. Carboxylation of the glutamic acid residues of the vitamin K-dependent factors must occur for effective coagulation to occur on platelet and endothelial phospholipid templates. (Reprinted with permission from Stead RB. Clinical pharmacology. In: Goldhaber SZ, ed. Pulmonary embolism and deep venous thrombosis. Philadelphia: WB Saunders Co, 1985:107)

drug is pharmacologically active, interactions with simultaneously administered medications which displace albumin-bound warfarin, ie, phenylbutazone, will potentiate the anticoagulant effect, prolong the PT, and increase the risk of hemorrhage. Potentiation of warfarin effects can also be produced by medications that interfere with its metabolism, (ie, cimetidine, phenytoin); its plasma clearance (ie, disulfiram, trimethoprim-sulfamethoxazole); and its excretion. The concomitant use of drugs that affect platelet function or other areas of hemostasis, the presence of renal failure or hepatic dysfunction, and hyperthyroidism also increase warfarin's effects. Second-generation cephalosporin antibiotics containing N-methylthiotetrazole side rings, such as cefamandole and moxalactam, prolong the PT and warfarin effects by interfering with the vitamin K-dependent $\gamma$-carboxylation of coagulation factors.

Inhibition of warfarin effects is seen in association with drugs which induce the hepatic microsomal enzymes for warfarin metabolism, eg, phenobarbital and cisapride. Other medications enhance warfarin excretion by stimulating bile flow, eg, rifampin; or decrease its GI absorption, eg, cholestyramine. Administration of vitamin K, or ingestion of large amounts of vitamin K-containing foods, hereditary warfarin resistance, and hypothyroidism also interfere with warfarin anticoagulation. Whenever oral anticoagulation is difficult to control, potentiating or antagonizing medications or pathologic states should be searched for.

The PT monitors extrinsic pathway activity and warfarin anticoagulation and is sensitive to factors VII, X, and II. Because factor VII has a rapid half-life, early PT prolongations produced by oral loading doses of warfarin reflect predominantly depressed factor VII levels, but antithrombotic effects may be minimal. Only after depressions of the other clotting factors occur (ie, over 3-5 days) will the prolongation of the PT into the therapeutic range represent an effective antithrombotic state. The PT is less sensitive to warfarin and increases after the residual circulating factor IX has been depleted (ie, 18-24 hours). Because therapeutic decisions are based on PT results, it should be appreciated that the assay is affected by blood specimen handling and the choice of thromboplastin reagents used in the assay. Delays in assaying specimens may spuriously decrease PT results because of in vitro factor VII activation by factor XII activated by cold and/or by glass contact. This, in turn, may lead to unnecessary increases in warfarin dosing and increase the risks of bleeding.

The thromboplastin reagents used in the PT assay are derived from either human or rabbit brain. The rabbit brain thromboplastin, popularized in the United States, is less sensitive to changes in vitamin K-dependent factor activity than the human brain thromboplastin standardized in the United Kingdom. Therefore, the results of these two reagents are not equivalent, and warfarin dosing must be modified according to the assay technique.

Traditionally, warfarin therapy is initiated with a loading dose, often overlapping with heparin for several days, with adjustment of the warfarin dose until the PT reaches a therapeutic range of 1.4-2 times control values. If heparin therapy is discontinued at this point, a paradoxical hypercoagulable state may ensue. The prolonged PT may reflect depressed activities of factor VII and protein C, which has a half-life similar to that of factor VII, despite normal levels of factors II and X. Critical protein C reductions may precipitate warfarin skin necrosis, an irreversible process that rapidly progresses within the first 5-9 days of warfarin administration and often necessitates surgical amputation of the affected gangrenous area to prevent systemic sepsis. Therefore, it may be prudent to maintain concomitant heparinization for at least 5 days to allow for the depletion of the other vitamin K-dependent factors with longer biologic half-life lives.

The major complication of warfarin therapy, as with heparin, is bleeding. Hemorrhage is encountered with up to a 27% frequency in warfarin users. The risk of bleeding is dose-related and directly proportional to elevations of the PT. Drug interactions and underlying pathologic disorders or lesions also are important variables. In one study, the frequency of bleeding complications was significantly reduced if warfarin dosing was based on PT results obtained using human thromboplastin compared with rabbit thromboplastin assays.
plasma. Alternatively, parenteral administration of vitamin K, specifically reverses warfarin effects within hours, but even small doses (5-10 mg IV) of vitamin K, will complicate reestablishment of oral anticoagulation. Oral vitamin K preparations have no practical role in reversing warfarin effects. The use of prothrombin complex concentrates is associated with a high risk of hepatitis and considerable tendency toward thrombogenicity, so they should be reserved for only extreme clinical situations. It should be emphasized that any occult or gross hemorrhage associated with anticoagulation should prompt an evaluation for underlying lesions.

Other less common complications of warfarin therapy include the "purple toe" syndrome associated with long-term use, alopecia, dermatitis, urticaria, and erythema. "Purple toe" syndrome consists of the rare onset of painful blue discoloration and tenderness of the toes, which occurs predominantly in males after 3-8 weeks of oral anticoagulation therapy. The discoloration persists indefinitely, and there is no progression of the lesions. The etiology is unknown. Warfarin used during pregnancy crosses the placenta and is highly teratogenic. A characteristic embryopathy, with skeletal and facial abnormalities, results that may be related to inhibited γ-carboxylation in the fetus of a vitamin K-dependent bone protein, osteocalcin. Warfarin is secreted into the breast milk of anticoagulated mothers, but this does not cause clinical problems for nursing infants.

The Fibrinolytic Pathway

The coagulation process is counterbalanced by the fibrinolytic pathway, which degrades fibrin clots and limits clot propagation (Fig 6). Plasmin is the pivotal fibrinolytic protein and acts on the fibrin clot to generate fibrin degradation products, including the fibrin-specific D-dimer fragment. Plasmin lacks substrate specificity, however, and can also proteolyze fibrinogen, factors V and VIII, vWF, and components of the complement cascade. Plasmin is rapidly and specifically inhibited by a2-antiplasmin; an acquired or inherited deficiency of a2-antiplasmin is associated with significant bleeding problems. Antithrombin III, C1-esterase inhibitor, a2-macroglobulin, and a1-antitrypsin are less important plasma inhibitors of plasmin and probably function as secondary neutralizing agents following the consumption of a2-antiplasmin.

Plasminogen is the zymogen precursor for plasmin and circulates in the G1u (terminal glutamic acid)-1 form. Small concentrations of plasmin can degrade G1u-1-plasminogen into the Lys (lysine)-77 form, which has increased fibrin-binding selectivity and increased susceptibility to plasminogen activators for conversion to plasmin. Plasminogen binds to fibrin via the lysine binding sites on its "kringle" structures (finger-like projections on the plasminogen heavy chain).

Plasminogen activators can be physiologic or pharmacologic. Activated factor XIIa activates plasminogen in vitro; however, its physiologic importance in vivo is unclear. Interestingly, the propositus for severe factor XII deficiency (Hageman) experienced recurrent pulmonary emboli; numerous thrombotic episodes have since been described in others with the same deficiency.

Tissue-type plasminogen activator (tPA), which is synthesized, stored, and then released from damaged or stimulated endothelial cells, also activates plasminogen. Modulation of tPA activity in plasma is achieved by its interaction with specific inhibitors (PAI-1) released into the circulation by platelets and endothelial cells. Increased PAI-1 activity leading to decreased fibrinolytic potential has been observed in association with several hypercoagulable states, including premature MI in young patients. In turn, it appears that PAI-1 activity may be modulated by the thrombin-activated protein C-protein S complex, which inactivates this major inhibitor of tPA.

There are currently five commercially available or experimental types of pharmacologic agents that can convert plasminogen to plasmin to initiate fibrinolysis. They differ in their pharmacokinetics, their pharmacologic mechanisms, and their fibrin specificity. These properties determine the degree of systemic lytic effects observed during laboratory monitoring, the dosing schedule and ease in administration, their ability to be administered repeatedly within months of the original therapy, and perhaps the frequency of rethrombosis associated with their use. Ultimately, these factors influence cost, safety, and efficacy.

Streptokinase has no direct effect on plasminogen. Rather, it must first complex with residual circulating, non-fibrin-bound plasminogen to form an activator complex. If the plasminogen consumption by activator complex formation and plasmin generation is complete, inadequate amounts of residual plasminogen will be available to sustain the thrombolytic state, and paradoxical hypercoagulability may ensue. The lack of fibrin specificity and the half-life characteristics of streptokinase contribute to its extreme, sustained lytic effects, consisting of marked hypofibrinogenemia and high levels of fibrin degradation products. Streptokinase is purified from streptococcal bacterial filtrates and thus may be associated with severe febrile reactions and other allergic phenomena. These necessitate pretreatment with hydrocortisone, diphenhydramine, and acetaminophen. Its antigenicity prevents retreatment for at least 6 months until antistreptococcal antibodies dissipate. Typically, in the treatment of deep venous thrombosis (DVT) and pulmonary embolus (PE), streptokinase is administered as an IV bolus dose of 250,000 units followed by a maintenance dose of 100,000 units/h for 24-72 h for DVT and for 12-24 h for PE. For coronary artery reperfusion, 1.5 million units of streptokinase is usually administered over 1 h. Its cost-effectiveness ratio is very favorable.

The chemical acylation of the catalytic center of purified plasminogen streptokinase activator complexes has resulted in APSAC, a thrombolytic agent that is theoretically more fibrin specific than streptokinase, since physiologic decylation leading to plasmin generation should occur over a time long enough for the inert complex to bind to the thrombus. Plasmin generation within the thrombus should decrease its susceptibility to inhibition by a2-antiplasmin, and efficacy of thrombolysis should be enhanced. In practice, however, APSAC produces a marked systemic lytic state. APSAC possesses antigenic properties similar to conventional streptokinase.

Urokinase is present physiologically in normal humans and can be purified for pharmacologic purposes from human urine or from the supernatants of transformed renal cell cultures. Recently, urokinase has been produced through recombinant DNA technology. Urokinase directly cleaves...
and activates plasminogen to generate plasmin and does not produce as marked a degree of hypofibrinogenemia or plasminogen consumption as does streptokinase or APSAC. Allergic reactions rarely occur; urokinase is nonantigenic and can be readministered whenever appropriate. Urokinase is usually administered as a 2,000 units/lb IV bolus dose over 10 min (half-life of urokinase is approximately 10-15 min), followed by a maintenance infusion of 2,000 units/lb/h for 12-24 h for treatment of PE and up to 48-72 h for DVT. A 3 million-unit dose (with the initial 1.5 million units given as a bolus) has been used successfully for coronary artery thrombolysis. Finally, urokinase is approximately 5-10 times more expensive than streptokinase.

Single-chain urokinase plasminogen activator (SCUPA), or prourokinase, is the single-chain precursor of conventional urokinase, which has a two-chain structure. SCUPA has recently been produced through recombinant DNA technology. Clinical evaluations are in their early stages, however, in vitro, SCUPA has demonstrated a very high degree of fibrin specificity, related to its high affinity for fibrin-bound plasminogen. In vitro, only mild to moderate systemic lytic effects are observed. Its rapid half-life (~7 min) necessitates continuous infusions to maintain a fibrinolytic state, and the simultaneous administration of heparin may enhance reperfusion and minimize the tendency for rethrombosis.

Originally isolated from human melanoma cell lines, rtPA has recently become the best characterized and most widely administered fibrinolytic agent produced through recombinant DNA technology. rtPA represents the first major advance in thrombolytic therapy since the introduction of streptokinase and urokinase into clinical medicine. Clinical studies with rtPA have revealed a high degree of safety and efficacy. rtPA contains "kringle" structures similar to plasminogen that allows its juxtaposition with plasminogen on the fibrin clot surface with efficient localization of plasminogen generation and subsequent fibrinolysis. In practice, pharmacologic doses of rtPA produce systemic lytic effects, but the magnitude of hypofibrinogenemia, α₂-antiplasmin consumption, and level of circulating fibrin split products are significantly lower than observed with most of the other thrombolytic agents. Thrombolytic efficacy may be slightly greater with two-chain rtPA, which possesses less fibrin-binding specificity than the one-chain variety of rtPA; however, the commercially available preparation rtPA (Activase, Genentech) is composed mostly of the one-chain type activator and the clinical differences between the two forms are insignificant after appropriate dose adjustments are made. Results of clinical pulmonary embolism trials in which reperfusion of pulmonary vessels is documented at the completion of rtPA administration via repeated pulmonary angiography and 24 h later by repeated perfusion lung scan indicate that the lytic effects of rtPA persist long after rtPA is cleared from plasma. The plasma half-life of rtPA is short (~8 min). A single 2-h infusion of 100 mg of rtPA has been very effective in the treatment of pulmonary emboli. Infusions (0.05 mg/kg/h) over 24 h are being studied for the lysis of DVT, and 3-h infusions (60-20-20 mg) are used for coronary artery thrombosis. rtPA is not antigenic or associated with allergic reactions. The cost of a course of rtPA therapy is comparable to that of urokinase.

Heparinization should be instituted following discontinuation of any thrombolytic agent to prevent rethrombosis at the site of vascular injury and to impede thrombus propagation. Typically, systemic heparin anticoagulation is initiated when the thrombin time decreases to 1 1/2-2 times the control value. A continuous IV dose of 1,000 U/h without a preceding bolus dose is preferred and can subsequently be adjusted to maintain the PTT at 1 1/2-2 times control values. Oral anticoagulation can be started within 24 h with at least a 5-day overlap before discontinuation of heparin.

Laboratory monitoring during the administration of thrombolytic therapy is intended to document the production of a lytic state. In this regard, the thrombin time and the fibrinogen concentration are the most readily available, the least expensive, and the least labor intensive. Thrombin time prolongations may be exaggerated with concomitant heparinization. The PT and PTT are less sensitive to lytic changes and may be spuriously prolonged by marked hypofibrinogenemia (<80 mg/dl) and by plasmin proteolysis of factors VIII and V. Measurement of fibrin degradation products is time consuming, and baseline values are often elevated by the underlying thrombotic process. Commercially popular latex agglutination assays measure fibrin fragments D and E but not the earlier-formed split products. In addition, prolongations in the thrombin time may reflect the generation of fibrin split products. Analysis of α₂-antiplasmin levels and detection of generated D-dimers, BB1-42 (Fig 5), and plasminogen consumption is usually reserved for research purposes. The euglobulin lysis time is not readily available in many laboratories, although it is very sensitive to lytic effects in plasma. It may be difficult to perform and interpret when severe hypofibrinogenemia is present.

Bleeding is the best known and most common complication of thrombolytic therapy and has limited the acceptance of this therapeutic approach. Goldhaber et al calculated that streptokinase therapy produced serious bleeding almost 3 times more frequently than heparin in the treatment of DVT. Numerous other studies indicate that life-threatening hemorrhage occurs in far fewer than 5% of patients. Heparin, on the other hand, was associated with serious bleeding in a significant 14% of patients in the UPET study. Intracranial bleeds have an overall incidence of 1.0-1.5%. Careful patient selection and minimal patient manipulation with invasive diagnostic procedures, venipunctures, and arterial sticks can minimize this risk. Careful and prolonged local compression of vascular puncture sites, which are the most common sources of significant bleeding problems, should provide adequate protection.

Bleeding is not correlated with the degree of hypofibrinogenemia or the level of fibrin split products generated, except in one recent study; neither is the degree of hypofibrinogenemia clearly correlated with the success of achieving significant thrombolysis. However, the incidence of hemorrhage is correlated to the duration of lytic therapy and to the concomitant use of antiplatelet aggregation agents or heparin. Therefore, it is reasonable to believe that bolus administration of lytic agents, which telescopes the total dose of drug into a short period, will achieve similar success rates of thrombolysis compared with prolonged infusions, which may be terminated prematurely because of bleeding
problems. An additional promising approach is the use of synergistic combinations of thrombolytic agents with different mechanisms of fibrin specificity. The use of rtPA and SCUPA in a significantly reduced dose combination in patients with coronary artery occlusion produced rapid and effective reperfusion, without evidence of systemic fibrinolytic activation or clinically important bleeding.

Since there appears to be little correlation among the degree of hypofibrinogenemia, level of circulating fibrin split products, and the incidence of bleeding, other hemostatic alterations produced by the systemic lytic state may be important. Plasmin can degrade platelet membrane glycoproteins, subendothelial matrix components, and other coagulation and adhesive proteins (von Willebrand factor, thrombospondin, fibronectin). Any of these events can induce independent coagulopathies.

Thrombolytic therapy is absolutely contraindicated in patients with recent (within 10-14 days) surgery, invasive procedures, trauma, and obstetric delivery. A history of hemorrhagic CVAs and primary or metastatic intracerebral neoplasm also renders thrombolytic therapy unadvisable.

If severe bleeding occurs and requires immediate reversal of the lytic state, the lytic drug therapy should be stopped and infusions of fresh-frozen plasma or cryoprecipitate should be initiated. This approach restores the depleted fibrinogen and clotting factors degraded by plasmin and supplies an exogenous source of α2-antiplasmin to inhibit plasmin activity. If bleeding persists, the administration of epsilon aminocaproic acid (Amicar) or tranexamic acid (Cyclokapron), both as 3-5-g boluses IV over 30 min, followed by 1.0 g/h IV, will prevent further plasmin generation and plasmin lysis by functioning as competitive inhibitors.

The effective use of thrombolytic therapy provides the teleologic advantage of rapid vascular reperfusion. Anticoagulation intends to maintain vascular patency. The benefit-risk ratio of these drugs can be enhanced by careful patient selection, appropriate agent selection, and careful management, all of which require an appreciation of the coagulation and fibrinolytic mechanisms and a working knowledge of the pharmacologic properties of the available thrombolytic agents and anticoagulants.

REFERENCES

1 Spert TH, Erickson RB. The vascular wall in the pathogenesis of thrombosis. Thromb Haemost 1966; 21(suppl):87-86
9 Weiss HJ, Turitto VT, Baumgartner HR. Effect of shear rate on platelet interaction with subendothelium in citrated and native blood. J Lab Clin Med 1978; 93:750-64
14 Vane JR. Inhibition of prostaglandin synthesis as a mechanism of action of aspirin-like drugs. Nature 1971; 231:232-34
256S

Tissue Plasminogen Activator in Cardiopulmonary Disease

83 Collen D, Stump DC, Van der Werf F. Coronary thrombolysis in patients with acute myocardial infarction by intravenous infusion of synergic thrombolytic agents. Am Heart J 1987; 112:1063-91
84 Collen D, Van der Werf F. Coronary thrombolysis with low dose synergistic combination of recombinant tissue-type plasminogen activator (rt-PA) and recombinant single-chain urokinase-type plasminogen activator (rscu-PA). Am J Cardiol 1987; 60:431-36

Downloaded From: http://journal.publications.chestnet.org/pdfaccess.ashx?url=/data/journals/chest/21594/ on 06/06/2017