SEMINARS IN
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Coagulation Disorders in Uremia

Daniela Molino, Domenico De Lucia, and Natale Gaspare De Santo

In patients with end-stage renal disease on maintenance hemodialysis, coagulation abnormalities such as hypercoagulability and thrombosis are common. Thrombotic complications in uremic patients are frequent and include those occurring at the vascular access and in the coronary, cerebral, and retinal arteries. Data do not entirely clarify the mechanisms involved and further investigations are necessary. Here we show a summary of coagulation and fibrinolytic disorders in uremia and the novelties foreseen in molecular biology.
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KEYWORDS uremia, coagulation, fibrinolysis, endothelial cell, factor V Leiden, antiphospholipid antibodies

In patients with end-stage renal disease on maintenance hemodialysis, abnormalities of coagulation occur and may cause blood hypercoagulability and thrombosis of the vascular access.[1] Thrombotic complications in uremia are found in dialysis blood access, subclavian veins, coronary arteries, cerebral vessels, and retinal veins, and in priapism.

Vascular access dysfunction represents the most frequent cause of hospitalization for dialysis patients, and it is responsible for major health care costs.[2] The prevention of vascular access thrombosis ultimately depends on early detection of risk factors.

The study of the coagulation cascade is very important in uremia and includes the evaluation of intrinsic and extrinsic factors, natural anticoagulants, fibrinolysis, antiphospholipid antibodies, and other markers of endothelial cell damage and inflammation. In recent years, studies on coagulation in patients on maintenance hemodialysis have investigated (1) abnormalities of coagulation; (2) the relationship between circulating levels of coagulation factors, fibrinolysis, and thrombophilia; (3) the role of endothelial cell damage in blood hypercoagulability; and (4) the role of antiphospholipid antibodies in thrombosis of vascular access.

Departments of Nephrology, General Pathology, and Clinical Pathology, the Laboratory of Hemostasis and Thrombosis, and the PhD Program in Nephrological Sciences, Second University of Naples, Naples, Italy.
Supported by Ministero Istruzione, Università e Ricerca.
Address reprint requests to Natale Gaspare De Santo, MD, 1st Chair of Nephrology, Second University of Naples, 5 Via Pansini, Pad 17, 80131 Naples, Italy. E-mail: nataleg.desanto@unina2.it

Physiology of Hemostasis

The coagulation system is a special protein system designed to maintain blood in a fluid state under physiologic conditions but primed to react to vascular injury in a manner to stem blood loss by sealing the defect in the vessel wall. Hemostasis should be viewed as an integrated system in which vascular wall, platelets, the coagulation system, and fibrinolysis interact to protect the integrity of the vascular system by reacting to tissue damage, and to pathogenic noxae of various nature, physical (rays), chemical (toxins), and biologic (bacterial and viral agents).[3]

The hemostatic system generally is distinguished in 2 phases: primary hemostasis (vasoplatelet), and secondary hemostasis (coagulative phase). During primary hemostasis there is a reaction to tissue damage with vasoconstriction, which dampens blood loss. When this phenomenon takes place, platelets are claimed at the lesion site through molecular mechanisms, which bind platelets to the endothelium and to the underlying collagen.[3] This first process leads to the activation of platelets, which aggregate and adhere to the damaged vascular surface through a multimeric protein known as von Willebrand factor, and with the contribution of activated monocytes and mastocytes. Secondary hemostasis takes place through a chain of biochemical reactions leading to clot formation, the final product of coagulation, composed of fibrin, platelets, and red cells trapped in the fibrin net (Fig 1). The activation of the coagulation cascade and fibrin clot formation is modulated finely and limited in time and space by an inhibitory system. Proteins assigned to this type of biochemical control are represented by natural anticoagulants. The most studied proteins are antithrombin III and the protein C system. The last phase of hemostasis is

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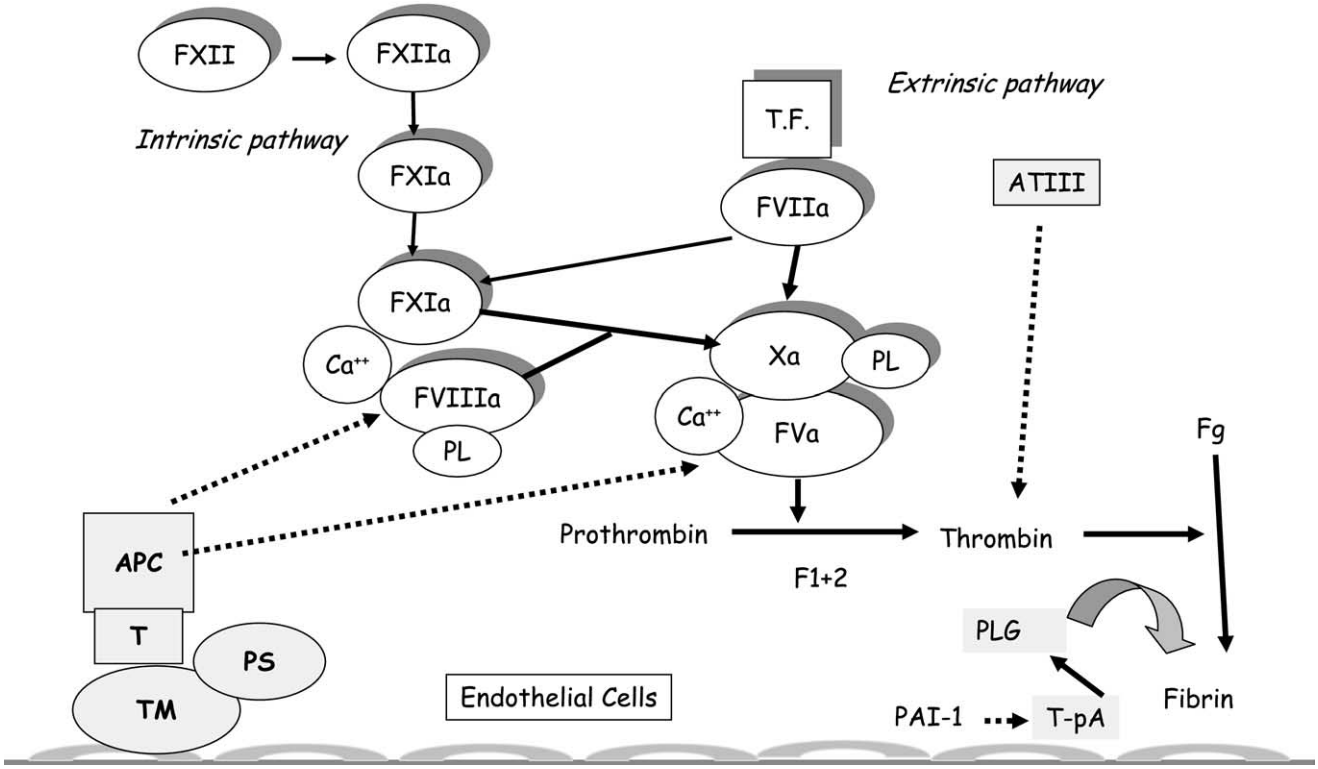

Figure 1 Physiology of the coagulation system.

fibrinolysis, a process leading to enzymatic degradation of fibrin through a proteolytic enzyme, plasmin, generated by the activation of its inactive plasmatic precursor (Fig 2). This molecule is activated as a result of the proteolysis induced by plasminogen tissue activator (t-PA) and of urokinase (u-PA, activator of renal origin). Fibrinolysis, a sophisticated inhibitory control mechanism, is exerted prevalently by the inhibitor of the plasminogen activator of type 1 (PAI-1), which normally is synthesized by endothelial cells, and to a lesser degree by activated platelets.

Role of Endothelium

Endothelium is heterogeneous, both metabolically and structurally.[4] Normal endothelium maintains blood fluidity by producing inhibitors of blood coagulation and platelet aggre-

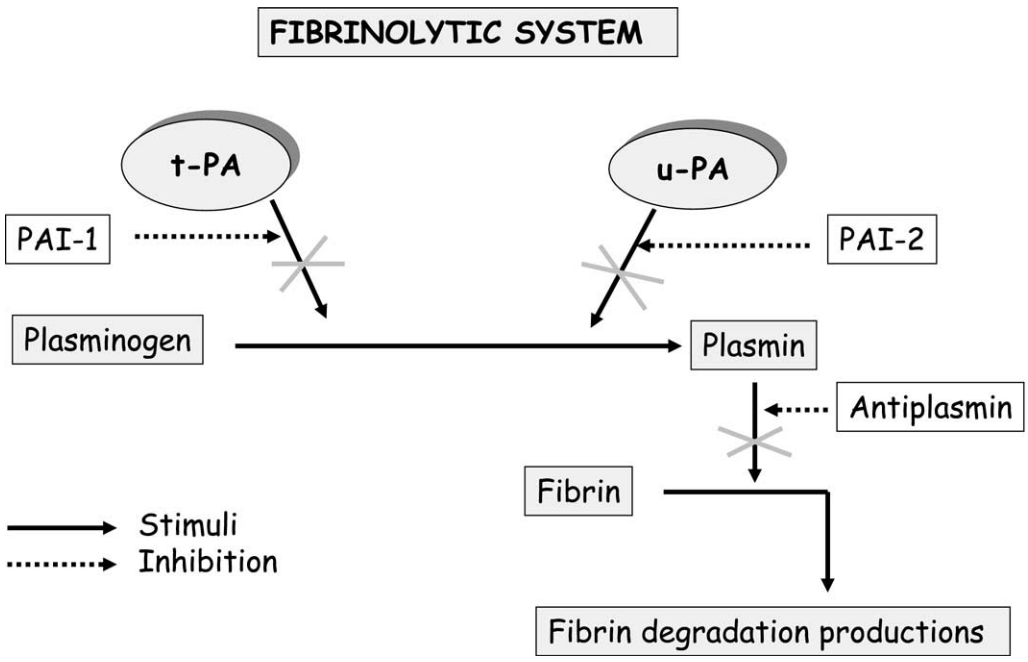

Figure 2 Physiology of the fibrinolytic system.

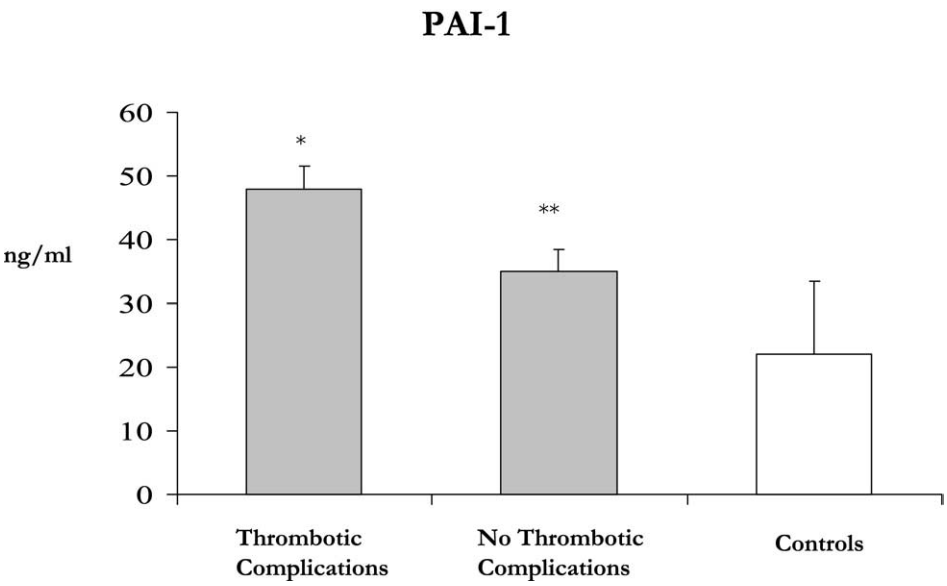

Figure 3 PAI-1 levels in healthy patients and in patients with thrombotic complications and without thrombotic complications.

gation, by modulating vascular tone and permeability, and by providing a protective envelope separating hemostatic blood components from reactive subendothelial structures and extracellular matrix. Therefore, endothelial cells synthesize and secrete basement membrane and extracellular matrix, which contain adhesion proteins, collagen, fibronectin, laminin, vitronectin, and von Willebrand factor. Endothelium inhibits blood coagulation by synthesizing and secreting thrombomodulin and heparan sulfate. In addition, the endothelium modulates fibrinolysis by synthesizing and secreting t-PA, urokinase plasminogen activator, and PAI-I, and it inhibits platelet aggregation by releasing prostaglandin I_2 (PGI_2) and nitric oxide. It also regulates vessel wall tone by synthesizing endothelins (type 1), which induce vasoconstriction, and by synthesizing PGI_2 and nitric oxide, which produce vasodilation. Endothelial cells lose their thrombogenic protective properties when stimulated by enzymes such as thrombin; hypoxia; fluid shear stress; oxidants; cytokines such as interleukin-1, tumor necrosis factor, and γ-interferon; synthetic hormones such as desmopressin acetate; and endotoxin.

Hemostasis in Uremia

Factors of Coagulation and Fibrinolysis

Thrombotic complications are frequent and include those occurring at the vascular access and in the coronary, cerebral, and retinic arteries.[5-7] Data on the topic, although extensive, do not entirely clarify the mechanisms involved. This lack of knowledge is of particular importance because vascular access thrombosis may be considered the Achilles' heel of modern hemodialysis.

Hypofibrinolysis may be present in hemodialysis patients, as shown by high plasma levels of PAI-1 (Fig 3), a factor capable of blocking plasminogen tissue activator and therefore the chain of events leading to the formation of plasmin, a potent enzyme catalyzing clot dissolution.[8,9] In addition, high plasma levels of factor VII and VIII[9,10] may be found (Fig 4), whereas the levels of activated factor XII are constantly lower. Finally, the presence of low levels of protein C, antithrombin III, and plasminogen have been described, although not consistently.

Vaziri et al[1] have studied various coagulation, inhibitory, and fibrinolytic proteins before and after hemodialysis in a group of patients with end-stage renal disease at risk for ischemic cardiovascular complications and vascular thrombosis. The study showed that despite ultrafiltration, plasma factor IX activity, von Willebrand factor, and fibrinogen concentrations decreased after hemodialysis with little or slight changes in the remaining coagulation parameters. They therefore suggested that these abnormalities might be involved in the pathogenesis of cardiovascular complications and vascular thrombosis in uremic patients.

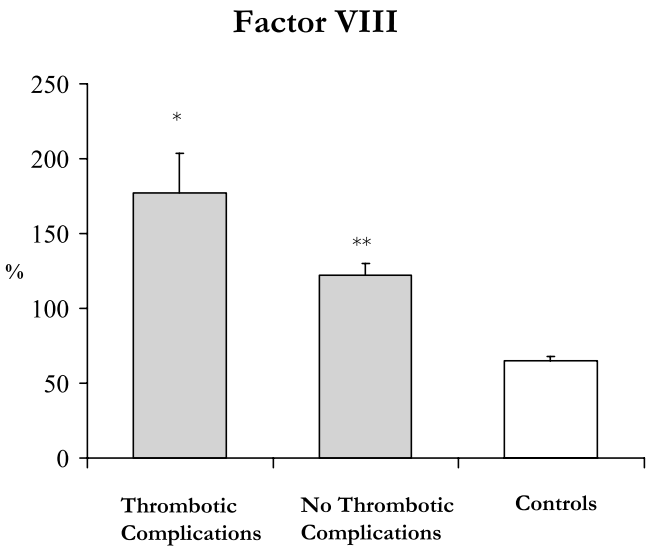

Figure 4 Factor VIII in healthy patients and in patients with thrombotic complications and without thrombotic complications.

Endothelium

The endothelial dysfunction is the common precursor and denominator of cardiovascular events including the development of atherosclerosis. It is known that patients with end-stage renal disease have endothelial cell damage,[11] which is a cause of coagulation abnormalities and of the thrombophilic state in uremic patients. According to some investigators, renal dysfunction is associated with markers of endothelial damage and inflammatory activity.[12] Plasma homocysteine may be an intermediate factor in the relationship between endothelial dysfunction and renal function. Homocysteine fast plasma levels produce a direct interaction with the anticoagulant pathway and affect the fibrinolytic system and endothelial cell functions. These interactions produce an inhibition of the thrombomodulin-dependent activated protein C system, which leads to persistent thrombin activation and formation of fibrin but also interferes with t-PA endothelial release into the vasculature, which predisposes to hypofibrinolysis. Finally, high homocysteine plasma levels may interfere with subendothelial cell proliferation via metalloproteinase-inducible genes though the activation on the metalloproteinase P-9 (MPP-9) subtype metalloproteinase. Biologically plausible mechanisms of vascular damage have been suggested, including effects on endothelium, platelet function, coagulation factors, and lipoprotein oxidation.

Segarra et al[8] suggested that increased levels of circulating PAI-1 could indicate a chronic endothelial activation and could be an additional tool to identify dialysis patients at risk for atheromatous cardiovascular disease. Malyszko et al[13] showed that in renal failure, particularly in patients maintained on chronic ambulatory peritoneal dialyses, there is evidence of endothelial cell injury and a higher degree of hypercoagulation, than in healthy patients. In this case, it may lead to fibrin deposition in the vascular wall, thrombus formation, and development and progression of atherosclerosis with its vascular complications. Data have been collected showing that high plasma concentrations of fibrinogen, D-dimer, thrombin-antithrombin III complex, coagulation factor VII, von Willebrand's factor, thrombomodulin, and PAI-1 are indicative of a thrombophilic state and endothelial cell injury.

We recently showed that in uremia, in addition to endothelial cell damage,[14-16] other pathogenetic mechanisms may cause hypercoagulability. We hypothesized that the alterations of secondary hemostasis and fibrinolysis are caused by endothelial damage and by the additive effects of some classes of antiphospholipid antibodies. These changes, for unknown reasons, are more severe in some patients, who consequently are more prone to thrombotic complications.[17]

Antiphospholipid Antibodies in Uremia

Recent studies have shown a high prevalence of antiphospholipid antibodies (lupus anticoagulant and anticardiolipin antibodies) among patients on hemodialytic treatment.[18,19] Quereda et al[20] were the first to report the presence of lupus anticoagulant in hemodialysis patients. Gronhagen-Riska et al[21] reported on the presence of the immunoglobulin G (IgG)-anticardiolipin antibody. Anticardiolipin antibodies represent a group of antibodies from the family of antiphospholipid antibodies and are autoantibodies with specificity for negatively charged phospholipid molecules. Antiphospholipid antibodies stabilize antigen binding to the anionic phospholipid surface by forming bivalent complexes and competing with coagulation factors for the anionic phospholipid surface. Antiphospholipid antibodies have been implicated in a variety of diseases but the exact meaning in uremia is not understood clearly.[22] For some scientists the presence of anticardiolipin antibodies represents a risk factor for vascular access thrombosis. Prakash et al[23] investigated the relationship between dialysis access thrombosis (arteriovenous grafts and arteriovenous fistulas) and the presence of increased concentrations of IgG anticardiolipin antibodies and concluded that a great prevalence of increased IgG-anticardiolipin antibodies (ACA) plasma titer is found in patients with recurrent arteriovenous graft thrombosis. For other investigators the presence of these antibodies simply is caused by aspirin use and access stenosis, thus their role in the pathogenesis of thrombosis is less clear. Adler et al[24] showed that IgM anticardiolipin antibodies are associated with stenosis of vascular access in hemodialysis patients but do not predict thrombosis. Therefore, further investigations are required to determine whether the association between aspirin use and IgM-ACA, or of IgM-ACA and access stenosis, is implicated in the pathogenetic mechanism underlying access stenosis. Palomo et al[25] found additional subtypes of ACA in patients on chronic hemodialysis with vascular access thrombosis. However, no association between the presence of antiphospholipid antibodies and arteriovenous fistula thrombosis could be established. Therefore, the presence of these antibodies in uremic patients should be clarified to determine whether these antibodies can predict thrombotic events, or represent a simple epiphenomenon of dialytic treatment.[26,27]

We report a novel subpopulation of antiphospholipid antibodies (anti–protein C and anti–protein S) in hemodialysis patients. Knowledge in this field to date has come from patients with systemic lupus erythematosus, or patients without systemic lupus erythematosus but with venous thromboembolism.[28-31] A significantly greater prevalence of anti–protein C antibodies and anti–protein S antibodies was seen in hemodialyzed patients with thrombosis of vascular access.

To understand the possible role for anti–protein C and anti–protein S antibodies in the hemostatic derangement in uremia, one should start with the fact that anti–protein C antibodies induce hypercoagulability by inhibiting the protein C system, and by blocking the enzymatic inactivation of activated factor VIII, resulting in an increased plasma level of activated factor VIII (Fig 5). This, in turn, stimulates the formation of thrombin, which after saturating all thrombomodulin binding sites, is diverted to the formation of fibrin. The thrombin excess favors the procoagulation pathway rather than the anticoagulation effect of binding with thrombomodulin.

The Contribution of Molecular Biology

Resistance to activated protein C recently was identified as a new thrombophilic defect caused by a single point mutation

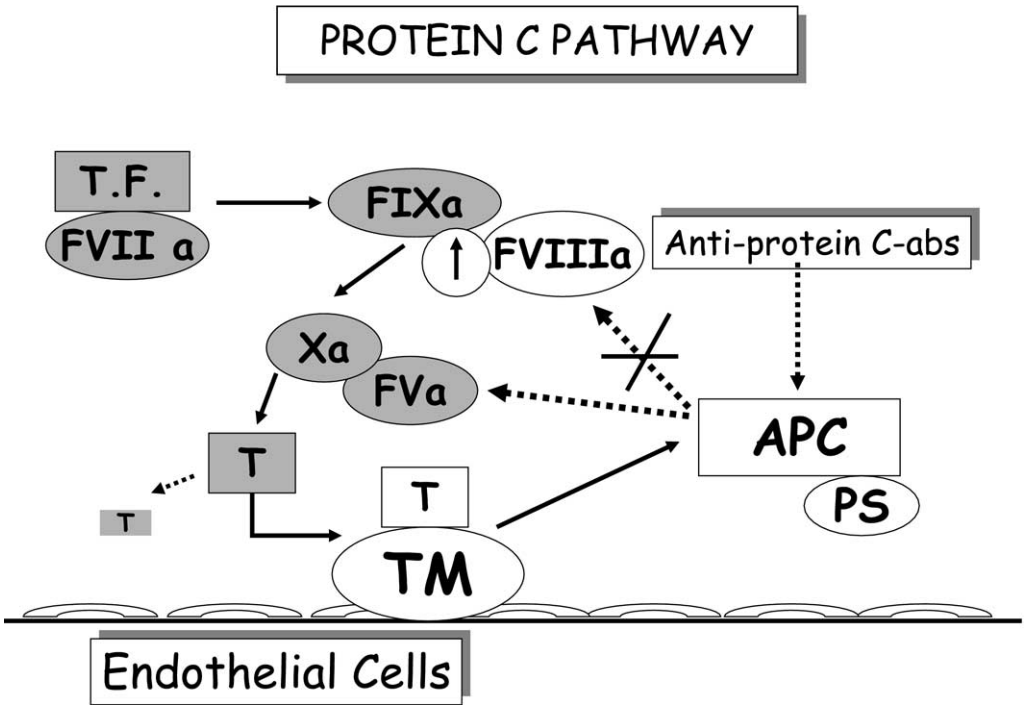

Figure 5 Mechanism of action of anti–protein C antibodies.

in the factor V gene. However, little is known about relevance of resistance to activated protein C for vascular access thrombosis in hemodialysis patients. Nevertheless, Fodinger et al[32] recently reported that the heterozygous carrier status for factor V Leiden does not represent a risk factor for shunt thrombosis in hemodialysis patients, a finding in contrast to other studies. In fact, hemodialysis patients with recurrent severe vascular access thrombosis, with a heterozygous state for factor V Leiden gene, are at high risk. Comparable discrepancies between genotype and phenotype already have been described by Greengard et al,[33] who reported a case of 4 siblings homozygous for factor V Leiden, 2 of them suffering from severe thromboses while the other 2 remained asymptomatic.

In addition, a recent study evaluated a variety of risk factors for thrombophilia (factor V Leiden, prothrombin gene mutation, factor XIII genotype, methylenetetrahydrofolate reductase genotype, lupus anticoagulant, anticardiolipin antibody, factor VIII, homocysteine, and lipoprotein [a] concentrations) in 419 hemodialysis patients. The study suggested that thrombophilia is associated with access thrombosis in dialysis patients.[34]

Conclusions

In conclusion, patients on maintenance hemodialysis are in a prothrombotic state similar to that occurring in nephrotic syndrome, unstable angina pectoris, acute myocardial infarction, fatal cardiovascular disease, essential hypertension, and vascular access thrombosis. The reported mechanism remains unclear. Our recent findings show that patients with thrombotic complications, compared with patients without thrombotic complications, have greater blood hypercoagulability and endothelium cell damage caused by important abnormalities in the coagulation cascade. We therefore hypothesized that in uremia hypercoagulability is caused by modifications of the coagulation pathway and fibrinolysis induced by endothelial cell activation and cell damage.[17]

Further studies are necessary to identify thrombotic risk factors in patients on hemodialysis. A biochemical and molecular screening of coagulation is now mandatory at the start of dialysis to rule out risk factors for hypercoagulability. This may help in preserving vascular access. One should keep in mind that maintaining patency of vascular access is still a challenge for nephrologists.

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