The Unique Hemostatic Dysfunction in Acute Promyelocytic Leukemia

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The association between the clinical bleeding manifestations in acute promyelocytic leukemia (APL) and the abnormal hemostatic function has long been recognized.1,2 Early investigators realized that the leukemic cells were responsible for the presenting coagulation defects.3 Studies in the following years have shown that the hemostatic function in this disorder is perturbed in ways unique to APL. Molecular studies revealed a chromosomal transformation t(15;17) encoding the formation of a promyelocytic leukemia gene-retinoic acid receptor alpha [PML-RARa] fusion protein, leading to differentiation arrest of the promyelocyte. Treatment with agents such as all-trans-retinoic acid (ATRA) or arsenic trioxide (ATO) results in differentiation of the APL leukemic cells and will restore the hemostatic function. Despite the dramatic reduction in mortality seen since the introduction of differentiation therapy with ATRA or with ATO, a large number of deaths still occur before complete remission is achieved. The early deaths are mostly attributable to the presenting coagulopathy. The prevention and management of this hemostatic abnormality have thus far been unsuccessful and remain a challenge to bring about a higher cure rate for this disease.

The hemostatic abnormalities seen in acute promyelocytic leukemia (APL) are unique and account for much of the morbidity and mortality of this disorder. Almost all patients present at diagnosis with laboratory findings of intravascular coagulation along with increased fibrinolysis. This unusual combination is correlated to the clinical manifestations with high risk of both bleeding and thrombosis. Recent studies have revealed that the leukemic promyelocytes in APL express increased amounts of tissue factor as well as elements of the fibrinolytic system, including tissue plasminogen activator, annexin A2, and plasminogen activator inhibitor type 1. These changes are responsive to differentiation therapy with all-trans-retinoic acid (ATRA) or with arsenic trioxide (ATO). Despite a dramatic reduction in mortality seen since the introduction of differentiation therapy with ATRA or with ATO, a large number of deaths still occur before complete remission is achieved. The early deaths are mostly attributable to the presenting coagulopathy. The prevention and management of this hemostatic abnormality have thus far been unsuccessful and remain a challenge to bring about a higher cure rate for this disease.

Although there is a better understanding of the pathogenesis of the hemostatic dysfunction, there are as yet no effective and specific tools in controlling the bleeding and thrombotic complications. There have been many recent reviews on this topic5–9 and the present article will be confined to report the highlights of these advances.

Clinical Manifestations

Most patients have varying degrees of bleeding manifestation at the time of presentation. These are, in the order of frequency, intracranial hemorrhage (ICH), pulmonary intralveolar hemorrhage, gastrointestinal bleeding, and generalized ecchymoses.4,10 Before the advent of differentiating therapy, bleeding was the cause of as much as 14% of the mortality; with ATRA treatment, bleeding still accounts for about half that rate.12 The most common form of bleeding is ICH (65–80%), located intracerbrally, and usually fatal. Patients prone to bleeding are older (> 60 years), have higher...
white blood count (> 10 × 10⁹/L), high blast count (> 30 × 10⁹/L), impaired renal function, and increased fibrinolysis with fibrinogen < 100 mg/dL.⁶,⁷,¹⁰,¹³ Severe thrombocytopenia is also a risk factor, though not verified in one study.¹³

In addition to bleeding, thromboembolism is also a complication in approximately 10% of patients.¹⁴,¹⁵ Additional risk for thrombosis is seen when the leukemic cells express CD2, CD5, FMS-like tyrosine kinase 3-internal tandem duplications (FLT3-ITD), and the bcr3 isoform.¹⁶ Also, the microgranular form of APL has been associated with portal vein thrombosis.¹⁷ The incidence of thrombosis was found to be 8.8% after treatment with the AIDA regimen (ATRA + idarubicin). Most patients manifested with acute myocardial infarction or deep vein thrombosis.

**Coagulation Profile**

Laboratory findings at the time of presentation show signs of activation of coagulation with elevated fibrinopeptide A, prothrombin fragment 1 + 2, and thrombin–antithrombin complexes.¹⁸–²⁰ There are also signs of consumption of coagulation factors as evidenced by an increase in fibrin degradation products and D-dimer with a decrease in the level of fibrinogen. Thrombocytopenia is usually present as a result of impaired platelet production in the bone marrow and because of peripheral consumption. The leukemic promyelocytes contribute much to the pathogenesis of the coagulopathy (► Fig. 1). Both the human APL cell line NB4 and the leukemic promyelocytes in APL patients have been shown to increase expression of two procoagulants, tissue factor (TF), and cancer procoagulant (CP).²¹–²³ TF is activated by phospholipids, forms a complex with factor VII which activates factor X, and is the major procoagulant that initiates the coagulation process. CP can activate factor X directly but studies of its action are sparse. The activation of TF is enhanced by apoptosis, during which the phospholipids in the cell membrane is exteriorized.²⁴ As a result, apoptotic cells are more thrombogenic than the corresponding normal cells.²⁵ Thus, the state of hypercoagulability is increased during periods of increased apoptosis of promyelocytes, such as in active leukemic proliferation and during treatment with ATO or chemotherapy. The increased expression of TF in the promyelocytes is corrected with differentiation therapy with ATRA or ATO.¹²,²⁶,²⁷

**Changes in Fibrinolytic System**

Fibrinolytic activity is increased as shown by elevated plasma levels of tissue plasminogen activator (tPA), urokinase-type plasminogen activator (uPA), and plasmin and decreased plasminogen activator inhibitor type 1 (PAI-1) and α₂-antiplasmin.⁵,²⁰,²⁸–³³ The abnormal promyelocytes express tPA and uPA. In addition, annexin A2, a receptor for plasminogen and tPA,³⁴,³⁵ and PAI-1 are also highly expressed. At the same time, there is reduced activity of another inhibitor of fibrinolysis, thrombin activatable fibrinolytic inhibitor (TAFI), by as much as 60%.²⁷,³⁶ With these changes in activators and inhibitors of fibrinolytic system, the resultant fibrinolytic activity has not been clearly delineated. It may vary with the different stages of the disease and may also be different in different tissues. Of note, in the microvascular endothelial

![Fig. 1](image.png)  
**Fig. 1** The abnormal hemostatic function in acute promyelocytic leukemia. ATO, arsenic trioxide; ATRA, all-trans-retinoic acid; DIC, disseminated intravascular coagulation; PAI-1, plasminogen activator inhibitor type 1; TNF, tissue necrosis factor; tPA, tissue type plasminogen activator; uPA, urokinase type plasminogen activator (modified from Kwaan and Rego³⁸).
cells in human and rodent brain, annexin A2 is constitutively highly expressed.\textsuperscript{37} In vitro plasmin generation on addition of tPA to microvascular endothelial cells was found to be much higher in those derived from brain than those derived from the rest of the body.\textsuperscript{37} These characteristics may explain the higher risk of intracerebral hemorrhage when compared with bleeding elsewhere in the body.

The changes in both the coagulation and fibrinolytic profiles can also be found in microparticles in the plasma of APL patients. Increased antigen levels of TF, tPA, PAI-1, and annexin A2 were found in microparticles derived from APL myeloid cells at the time of diagnosis.\textsuperscript{38,39} These elevated levels trend to normal with remission of the disease.

Thus, it can be seen that these altered hemostatic functions result in a hemostatic picture unique to APL. The changes in the coagulation profile may appear at first sight as disseminated intravascular coagulation (DIC). However, on further analysis, this is incorrect as there are notable differences between DIC and the picture seen in APL. The increased fibrinolytic activity in severe DIC is a secondary response to intravascular coagulation, whereas in APL, there are additional factors contributing to the excessive fibrinolysis as discussed above.

The abnormal hemostatic function is reversed following treatment with ATRA\textsuperscript{20,27,31,40} and ATO.\textsuperscript{9,26,40} Although clinical bleeding from the coagulopathy generally abates by 5 to 7 days of treatment, the coagulation and fibrinolytic profiles return to normal after 14 days or longer.\textsuperscript{12}

**Clinical Implications**

As the bleeding and thrombotic complications account for much of the morbidity and mortality of this disease, many attempts have been made to correct the abnormal hemostatic functions. Unfortunately, these have not in general been met with success. Heparin anticoagulation, for example, has no effect on the coagulopathy.\textsuperscript{41}

Inhibitors of fibrinolysis, such as aprotinin, epsilon-aminocaproic acid (Amicar; Xenodyne Pharmaceuticals, Inc., Newport, KY), or tranexamic acid (Cyclokapron; Pfizer Corporation, Hong Kong, China), have also not been shown to prevent bleeding as demonstrated in the “GIMEMA” clinical trial involving 268 patients,\textsuperscript{41} and in the “LPA 99” trial.\textsuperscript{10,42} Conversely, these antifibrinolytic agents may pose a higher risk for thrombosis in APL patients, as shown in several case reports.\textsuperscript{43–45} Other hemostatic agents such as recombinant factor VIIa (Novoseven; Novo-Nordisk, Inc., Copenhagen, Denmark) have been shown to be successful in a few anecdotal reports.\textsuperscript{46–48} Like antifibrinolytic agents, however, this agent should be used with caution as it also carries a risk of thrombosis. Another antithrombotic agent, recombinant thrombomodulin (rTM) was recently reported to be of benefit in patients with severe coagulopathy and life-threatening bleeding.\textsuperscript{49} TM inhibits thrombin, forming a TM-thrombin complex. This complex, in the presence of plasmin, activates TAFI,\textsuperscript{36} an inhibitor of fibrinolysis. TM has been shown to be beneficial in patients with DIC associated with various etiologies.\textsuperscript{50,51} Thus in APL, TM will inhibit the coagulopathy and at the same time upregulate the reduced TAFI level, leading to reduced fibrinolysis. Verification of the successful use of rTM will add a new approach to the management of the coagulopathy and the excessive fibrinolysis.

The current guidelines for prevention and treatment of bleeding complications in APL recommend aggressive supportive measures with platelets, cryoprecipitate, and fibrinogen with a target of keeping the platelet count more than 30 to 50 $\times$ 10^9/L and the fibrinogen level above 1 to 1.5 g/L (100 mg-150 mg/dL).\textsuperscript{13,52}

They also strongly recommend the initiation of ATRA or ATO therapy as soon as the clinical diagnosis is made without waiting for molecular confirmation of t(15;17), as even a short delay in the initiation of ATRA therapy can adversely affect outcomes.\textsuperscript{53}

**Conclusion**

Recognition of the factors responsible for the altered hemostatic functions in APL has helped us to understand the pathogenesis of the bleeding and thrombotic complications in this life-threatening disease. Although the use of differentiating therapy with ATRA or ATO has resulted in a dramatic improved outlook for patients with APL, including molecular remission rates in excess of 90%, the persistence of early deaths from bleeding remains the main obstacle to the therapeutic objective of a cure of this form of leukemia. The reasons for the inability of the present therapeutic agents to correct the abnormal hemostatic factors are still unclear and need to be addressed. This author suggests a novel approach through our knowledge of the epigenetic control of tPA in endothelial cells. Inhibitors of histone deacetylase (HDAC) induce tPA expression in endothelial cells while class I HDAC repressed tPA expression.\textsuperscript{54} It is also known that aberrant HDAC modulation of the PML-RARa transcription is present in APL. It is thus possible that the epigenetic control of TF and PAI-1 expression is also affected in a similar manner resulting in their high expression. If so, interference of such control may be another means to earlier control of the coagulopathy and the fibrinolysis than that presently shown by differentiating therapy.

**References**


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