

Applications of Coagulation Testing and Methodology in the Trauma Patient.

Ali Gabali*, Tarek Jazearly, Radhakrishnan Ramchandren, and Martin H Bluth.

Wayne State University, Department of Pathology, 3990 John R St, Detroit, MI 48201.

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***For Correspondence**Wayne State University,
Department of Pathology,
3990 John R St, Detroit, MI
48201.

Tel: 313-745-1869

Fax: 313-993-4325

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partial thromboplastin time and
aPTT**ABSTRACT**

Trauma-induced coagulopathy and subsequent patient outcome depends on prompt recognition and monitoring of the coagulopathy and rapid restoration of hemostasis. Understanding of the application of common tests employed in the coagulation laboratory tests with respect to trauma patients allows an informed decision on how to proceed with patient treatment. This article focuses on the application and understanding of the basic coagulation laboratory tests in trauma patients.

INTRODUCTION

Post-traumatic coagulopathy with its challenging hemostatic presentation may impose an acute life threatening situation in the trauma patient. Besides that, acute blood loss in trauma imposes a life-threatening bleeding problem. To this end studies show that approximately 30% of trauma patients will have some sort of bleeding diathesis that requires medical intervention either early on during initial presentation or later during the patient's hospital course [1]. The most important risk factors for blood diathesis in trauma patients are hypothermia and acidosis [2]. Both of the risk factors cause dramatic reduction in the enzymatic activities of clotting factors. In addition, patients receiving massive transfusion will have hemodilution or even depletion of coagulation proteins. The impairment in the enzymatic activities and the marked reduction in coagulation proteins put trauma patients at greater risk for developing disseminated intravascular coagulation (DIC) and its life-threatening consequences of microvascular thrombosis and fibrinolysis. However, knowing that most trauma patients require an urgent surgical intervention, a rapid and careful investigation into the diathesis problems should be performed in all trauma patients. This article will review laboratory testing that could be helpful in recognizing coagulopathies associated with trauma.

Historic view

In the late 19th century, Wooldridge's experiments on animals showed that injecting tissue extracts into animals caused thrombosis followed by a tendency for hemorrhage [3]. This condition is known now as disseminated intravascular coagulation (DIC). Since then the awareness of the medical community of coagulopathies encountered in patients with excessive hemorrhage has been steadily increasing, although there was some disagreement as to its cause. Reports go back to the early 20th century describing decreased fibrinogen and other coagulation proteins in such patients. One of the most significant early papers describing coagulopathy in patients with excessive hemorrhage was published in 1960 by Drs. Penick and McLendon [4]. They reported several cases of coagulopathy associated with excessive bleeding. The growing utilization of routine coagulation testing in trauma patients or those with excessive hemorrhage has shown that coagulopathies are frequently common and they are major contributors to death.

The most common panel used to evaluate patients with trauma includes platelet count, prothrombin time-international normalized ratio (PT-INR), activated partial thromboplastin time (aPTT) and fibrinogen level.

Specific Laboratory Testing

Platelet Count

Platelets are essential for primary hemostasis. In most laboratories, a normal platelet count is $150-450 \times 10^3/\mu\text{l}$. A platelet count is ordered as a part of complete blood count (CBC) test. In trauma patients, platelets quantitative abnormality (thrombocytopenia) is seen more frequently than qualitative abnormality. Packed red blood cells (PRBC) are platelet depleted and contain approximately 35 ml plasma; therefore the major cause of thrombocytopenia in trauma patients is hemodilution due to massive blood transfusion and receiving intravenous electrolyte solution as pre-hospital therapy. But knowing that 30-40% of platelets are normally stored in the spleen^[5], in real practice, platelet hemodilution due to massive transfusion happens at a slower rate than that of the coagulation proteins. Therefore, a marked sharp drop in platelet counts to levels below $50 \times 10^3/\mu\text{l}$ is uncommonly seen in trauma patients who received less than 20 units of PRBC.

Trauma patients are also susceptible to hypothermia and acidosis that both have a rapid detrimental effect on platelet adhesion and aggregation, likely contributing to the coagulopathy seen in such patients^[6,7,8]. There is still no reliable method to test for impaired platelet function in trauma patients and studies showed that the use of PFA-100 to test for platelet dysfunction in trauma patients is unreliable^[9]. However, some clinical laboratories continue to receive requests for PFA-100 platelet function testing even though sufficient data are lacking to support its routine use in trauma patients. Trauma also happens to individuals with underlying platelet defects and more work up is needed in such individuals to test for defects in platelet aggregation and secretion.

Prothrombin Time-International Normalized Ratio (PT-INR)

Almost 80 years ago, Armand Quick et al and Paul Owren have reported separately the first two methods used to measure prothrombin time^[10, 11]. Since 1954, both methods were increasingly used, when warfarin was approved for prevention of thrombosis and treatment of thromboembolic disorders. PT measures the integrity of extrinsic (factor VII) and common (Factors X, V, II and I) coagulation pathways and is very sensitive to vitamin-K dependent clotting factor (factors X, IX, VII and II). It is a clot based assay that measures time required for clot formation in seconds after adding thromboplastin and CaCl_2 to platelet-poor plasma. Different techniques are used to measure clot formation but the ones most used are the photo-optical and electromechanical methods. The photo-optical technique measures the amount of reduction in transmitted light associated with clot formation as soluble fibrinogen is converted to insoluble fibrin. However, this technique is not suitable for a hemolyzed sample or a sample with high levels of interfering substances such as bilirubin and lipid. Many laboratories use electromechanical method for such samples. The PT is highly dependent on the type of thromboplastin used in these studies and it shows some degree of variation from one laboratory to another even for the same sample^[12]. In 1980, the INR was introduced to standardize PT results and to eliminate the variability seen with reporting PT values^[13]. A prolonged PT-INR with a normal aPTT is seen in factor VII deficiency and warfarin therapy.

PRBC contains approximately 35ml of plasma and is devoid of the two most labile clotting factors, namely factors V and VIII. Therefore, trauma patients receiving massive transfusion of PRBC develop hemodilution of clotting proteins. An isolated prolonged PT-INR in the trauma patient is usually not associated with significant bleeding unless the PT-INR is markedly prolonged, more than 1.8 times the control value, and there is also an associated prolonged aPTT.

Activated Partial Thromboplastin Time (aPTT)

In 1953, Langdell et al described PTT for the first time as a test for diagnosing hemophilia^[14]. Partial PTT measures the integrity of intrinsic (factors XII, XI, IX and VIII) and common coagulation pathway factors (X, V, II and I). Like the PT, PTT is also a clot based assay that measures the time required for clot formation in seconds after adding partial thromboplastin (the phospholipid part of thromboplastin) and CaCl_2 to platelet-poor plasma. The prefix "activated" is used because an activating agent such as silica or kaolin is used in the reaction to produce activated partial thromboplastin time (aPTT). In daily practice, the aPTT is utilized to monitor heparin and coagulation factor replacement therapy and as an initial screening test to detect inhibitors of the intrinsic pathway. The aPTT is measured by the same methods that measure the PT-INR.

A slight prolongation in the aPTT and PT starts to appear after administering 10 to 12 units of PRBC. The aPTT is more susceptible to hemodilution than the PT. In fact, studies have found that a prolonged aPTT is the most common coagulation abnormality in general practice and that the most common cause of isolated prolonged aPTT is lupus anticoagulant^[15]. Therefore, in the trauma patient an isolated prolonged aPTT is not significant predictor of bleeding tendency unless the aPTT is markedly prolonged, more than 1.8 times the control value, and there is also an associated prolonged PT-INR.

Fibrinogen Level

The final result in intrinsic and extrinsic pathways is to activate thrombin that converts fibrinogen, the most abundant clotting factor, to the insoluble fibrin that makes the building blocks for clot formation. Many techniques are used to measure fibrinogen levels. The gold standard method, the washed clot method, is time consuming and not used by many clinical laboratories. Instead, many laboratories are measuring fibrinogen using the von Clauss technique, first described in 1957, and the Clotting Rate Assay^[16]. The former requires more technical skills than the latter. The von Clauss technique is a clot-based assay that measures the time required for clot formation in seconds after adding thrombin to different dilutions of plasma. The Clotting Rate Assay measures the rate of increase in plasma turbidity when the soluble fibrinogen is converted to the insoluble fibrin by thrombin^[16].

Fibrinogen is an acute phase reactant that may increase early on in trauma. However, fibrinogen levels are decreased by the same factors that decrease other clotting proteins in trauma patients including hemodilution and development of DIC^[17]. The normal level of fibrinogen is 200-400mg/dl. A fibrinogen level less than 80mg/dl is associated with a prolonged PT-INR and aPTT. A level below 50mg/dl is usually associated with bleeding^[17].

Thromboelastography (TEG)

TEG is a method that was developed in 1948 and now is gaining an increasing popularity in evaluating patients with trauma as an important point-of-care test. The test uses less than 0.35 ml of whole blood sample placed in an oscillating cuvette with a pin that records different stages of clot formation and lysis including platelet function (maximal amplitude, MA), coagulation factor activity (r time), speed of clot formation (k time), fibrin formation (alpha angle) and finally the activity of fibrinolytic enzymes (blood lysis index)^[18, 19]. TEG is described by many as a method that provides rapid point-of-care testing. The whole process takes less than 40 minutes, and the instrument provides calculations for all the parameters. The turnaround time for basic coagulation testing in trauma patients (platelet count, PT, aPTT and fibrinogen) in most of clinical laboratories offering this test is about 60 minutes. Therefore, TEG may guide prompt hemostatic monitoring and therapy in trauma patients. Although variations of viscoelastic testing exist including rotational thromboelastometry – ROTEM™ and thromboelastography - TEG™, the basic principles are similar in that in the former the cuvette is fixed and the pin oscillates and in the latter its reversed. Although there have been well established studies regarding the use of viscoelastic testing implementation in the trauma setting^[20], there are caveats associated with this such as time from blood draw to test, operator variability, time to test result^[21], effective implementation of viscoelastic testing methodologies in the appropriate clinical setting may facilitate efficient utilization / reduction of blood products^[22].

CONCLUSION

Trauma patients are prone to coagulopathy that may be associated with unfavorable outcomes. Prompt diagnosis of trauma induced coagulopathy allows fast restoration of hemostasis and avoidance of many life-threatening complications. To this end the current and emerging testing panels offered by the clinical laboratory serves to provide real time value towards effective patient management.

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