Current Transplantation Reports (2018) 5:251–263
https://doi.org/10.1007/s40472-018-0203-z

ANESTHESIA AND CRITICAL CARE IN TRANSPLANTATION (D AXELROD AND M KAUFMAN, SECTION EDITORS)

Current Hematological Concepts and Viscoelastic-Based Transfusion Practices During Liver Transplantation

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Published online: 3 August 2018

Abstract

Purpose When the cascade model of coagulation was postulated in 1964, it convincingly explained the conventional tests of coagulation and their therapeutic applications for existing anticoagulants. But the conventional tests only tend to measure the procoagulant factors and not the anticoagulant factors present in the blood, as a result, the coagulation concept was updated to cell-based model in 2001. Despite these facts, the conventional tests are still used perioperatively in liver transplantation for blood product management, at the risk of causing over-transfusion and deleterious prothrombotic effects. This article reviews the current understanding of coagulation and suggests an improved method to manage intraoperative blood product replacement.

Recent Findings We set out to develop a diagnostic and dosing protocol based on viscoelastic tests, which more accurately reflect the dynamic interplay between pro and anticoagulants in the end-stage liver disease patient. This approach reduces the overtransfusion and resulting harm from excessive coagulation without increasing the risk of intraoperative bleeding.

Conclusion While we were successful in formulating a dosing regimen based on available literature and our own institutional practices for treating deficiencies of clotting factors and fibrinogen, more research is needed to arrive at a dosing regimen for platelets based on functional deficiency.

Keywords Liver transplant · Current concepts · Hematology · Viscoelastic tests · ROTEM-based protocol

Abbreviations

ADAMTS	A Disintegrin And Metalloproteinase with a ThromboSpondin Type 1 motif
ADP	adenosine diphosphate
APTEM	aprotinin-added ROTEM
aPTT	activated partial thromboplastin time
AT	antithrombin
CFT	clot formation time
CT	clotting time
ESLD	end-stage liver disease
EXTEM	extrinsic pathway component of ROTEM
Gp	glycoprotein
HEPTEM	heparinase-added ROTEM
INTEM	intrinsic pathway component of ROTEM
Leu	leucine
LT	liver transplant
MCF	maximum clot formation
ML	maximum lysis
PAF	platelet-activating factor
PAI	plasminogen activator inhibitor
PAR	protease-activated receptors
PI	plasmin inhibitor
PT	prothrombin time
RGD	arginine-glycine-D aspartate
ROTEM	rotational thromboelastometry
TAFI	thrombin activatable fibrinolysis inhibitor
TEG	thromboelastogram
TF	tissue factor
TFPI	tissue factor pathway inhibitor
TM	thrombomodulin
tPa	tissue plasminogen activator

This article is part of the Topical Collection on *Anesthesia and Critical Care in Transplantation*

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uPa	urine plasminogen activator
val	valine
vWF	von Willebrand factor

Introduction

The hematological profile of an end-stage liver disease (ESLD) patient is very different from a healthy person as the pro- and anticoagulants synthesized in liver decrease. The decrease in factor levels is compensated by increases in factors synthesized outside the liver. The conventional tests of coagulation are limited in informing a practitioner about balance between achievement of hemostasis and avoidance of thrombosis. It is therefore imperative for the transplant anesthesiologist to have an in-depth understanding of the thrombopathy and coagulopathy of the ESLD patient, the ability to rapidly interpret the results of viscoelastic tests, and a protocol to decide on the appropriate use of a blood product at the relevant time. In this review article, we will discuss the current understanding of cell based concept of coagulation and the role of fibrinogen, rebalanced coagulation in patients with ESLD, and use of viscoelastic monitoring to develop a protocol based blood component therapy.

Basic Science for Cell-Based Concept of Hemostasis

Blood clot formation starts with the activation of platelets and their interaction with the blood vessel endothelium by the process of primary hemostasis, resulting in the formation of platelet plug. This plug is further strengthened with the addition of fibrin during the process of secondary hemostasis, which starts with the activation of clotting factors and results in the formation of thrombin.

Primary Hemostasis

Primary hemostasis occurs immediately after a vascular injury, when subendothelial collagen gets exposed to blood (Fig. 1). von Willebrand factor (vWF) quickly attaches to the collagen, followed by platelet binding through vWF receptors (glycoprotein Gp 1b-v-1x). This adhesion is further stabilized by direct interaction of platelets with subendothelial collagen through collagen receptors (Gp Ia/IIa and Gp VI). Subsequent platelet activation leads to release of the contents inside the platelet (coagulation factors, calcium, ADP, thromboxane A2, PAF) and aggregation with adjacent platelets through Gp IIb/IIIa receptors, mediated by vWF or fibrinogen [1, 2].

Secondary Hemostasis

The traditional funnel-shaped coagulation cascade model (extrinsic pathway starting with factor VII and intrinsic pathway starting with factor XII, finally culminating in the activation of thrombin) has given way to cell-based model of hemostasis with the following three phases: initiation, amplification, and propagation (Fig. 2) [3, 4].

Initiation The vascular injury that initiates the primary hemostasis, thereby activating platelets, also releases FVII, which on contact with the tissue factor (TF) commences the process of secondary hemostasis. This leads to activation of FX and IX [5, 6]. FX activates FV, and small amounts of thrombin is formed. Thrombin in turn activates FXI on-activated platelet surface, making FXII not necessary for hemostasis. Tissue factor pathway inhibitor (TFPI) inhibits FVIIa/TF before it could produce enough FXa for complete hemostasis [7–9].

Amplification During amplification, thrombin binds to platelets, which through its protease-activated receptors (PAR receptors) amplifies platelet activation and releases FV and through its non-PAR receptors, activates FVIII by cleaving vwf/FVIII complex [10–13].

Propagation Starts with FIXa formed during initiation phase, diffusing from TF bearing cells to platelet surface. Plasma FXI also binds to activated platelets, facilitating its activation by thrombin. This FXIa provides additional FIXa directly on platelet surface [9]. Tenase (VIIIa/IXa) and prothrombinase (Xa/Va) are assembled on platelet surface leading to massive amounts of thrombin being generated to convert fibrinogen into fibrin [3].

Clotting Factors

The following section contains a brief description of the structure and function of the key clotting factors involved in the coagulation pathway: thrombin, fibrinogen, and factor XIII.

Thrombin Thrombin is necessary for the activation of fibrinogen to form fibrin. Thrombin also has other essential functions like activating platelets, certain clotting factors like discussed in the previous section, and facilitating thrombomodulin-based anticoagulant effects. Thrombin is a serine protease generated from zymogen prothrombin through proteolytic cleavage by FXa. It has two polypeptide chains (A and B), which are covalently bound by disulfide bonds (Fig. 3) [14–16]. The thrombin active site contains serine-histidine-D aspartate residues and proteolyses substrates. Exosite I mediates binding of fibrinogen, fibrin, thrombomodulin (TM), and PAR-1,4. The fibrin bound to exosite I activates FXIII [17]. However, when TM, which

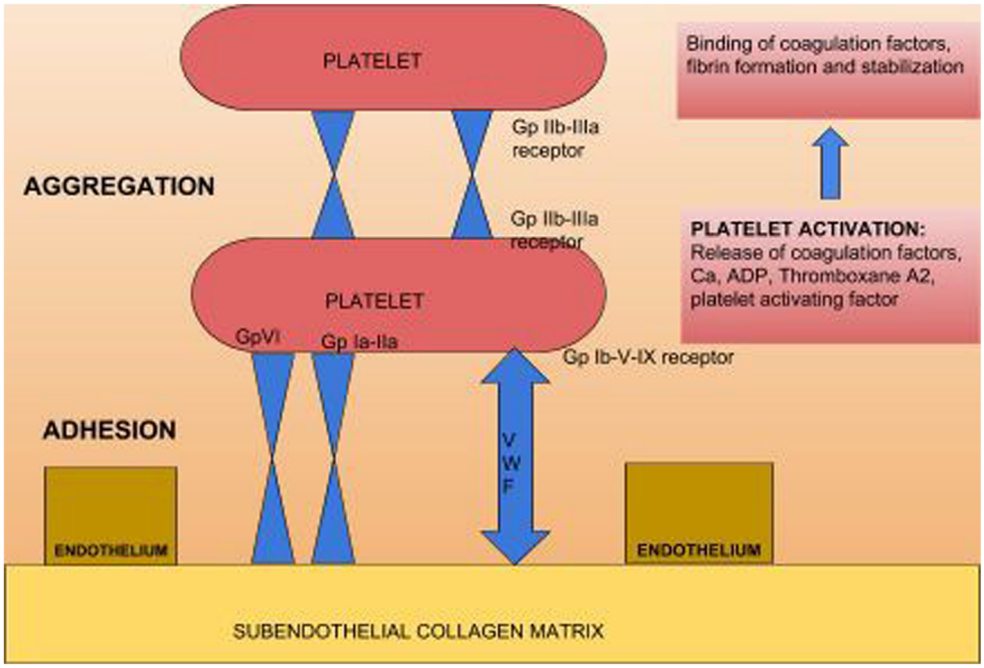

Fig. 1 Schematic diagram for primary hemostasis

has more affinity to the same site is present, it terminates the activation and also removes thrombin from clot. Exosite II mediates interaction with heparan sulfate (endothelial surface), glycoprotein 1b α (platelet surface), specific monoclonal antibodies, certain snake venoms, and protein T. Cofactor competition occurs between heparan sulfate and glycoprotein 1bα, and the higher concentration substrate predominates. The active site and exosite 1 are allosterically regulated by binding of sodium. Sodium binding switches thrombin from slow to fast form, which has higher specificity for procoagulant substrates like fibrinogen and PAR-1 and also greater reactivity to antithrombin. The slow form has higher specificity for protein C activation [18–20].

Fibrinogen Fibrinogen is a plasma glycoprotein synthesized in the liver. It is composed of three pairs of polypeptides-2Aα, 2Bβ, and 2γ chains. (Fig. 4) Thrombin cleaves the N-terminal ends of both Aα and Bβ polypeptides to remove fibrinopeptides A and B. Cleaving thrombin exposes "knobs" on E domain, which can interact with the "holes" always present on the D domain. Cross-linking happens by interaction of one E domain with four D domains of different fibrin molecules, thereby polymerizing to form staggered oligomers that build into protofibrils, then to three-dimensional network of entangled fibers and then fibrin clot. The platelets attach to the arginine-glycine-D aspartate site (RGD) on α subunit and c-terminal peptide on the γ subunit to facilitate aggregation at

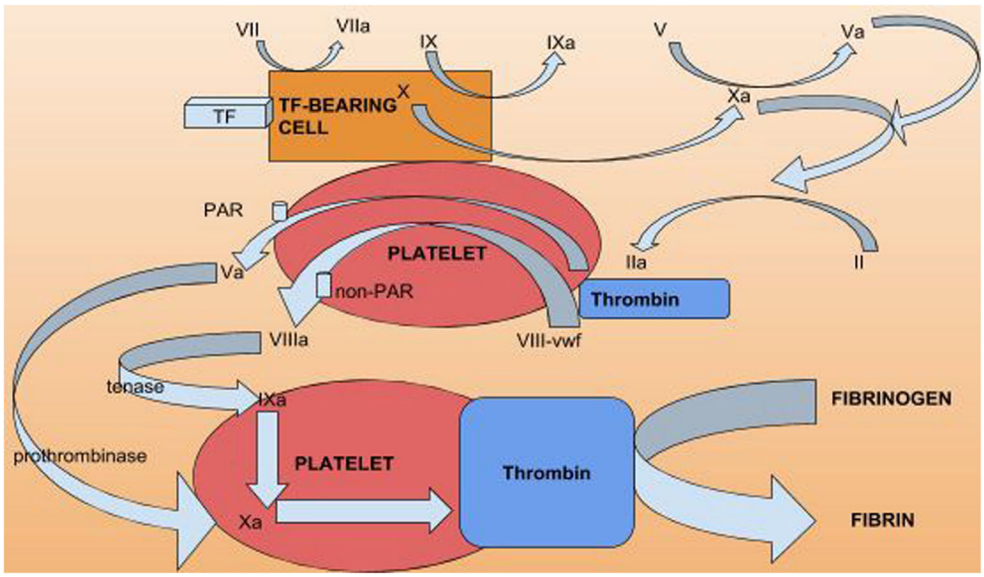

Fig. 2 Schematic diagram for secondary hemostasis

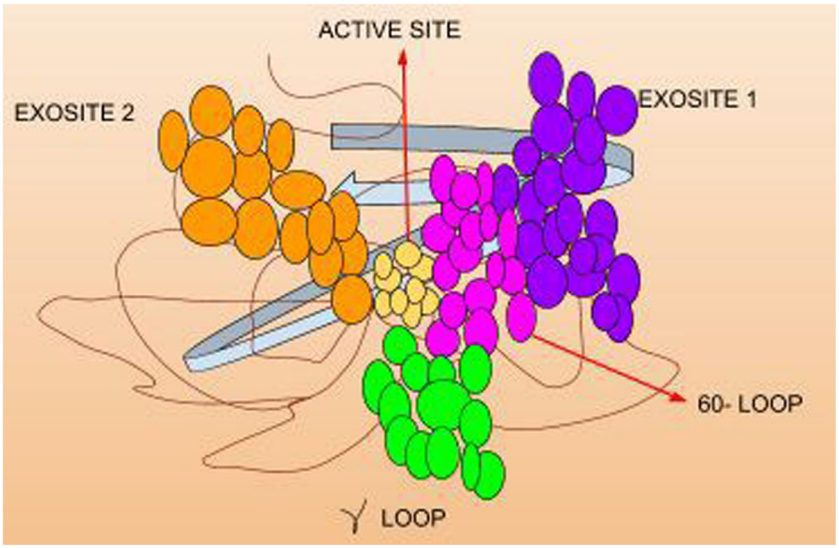

Fig. 3 Structure of thrombin

Gp IIb-IIIa receptors on the platelet surface. Thus, we see that fibrinogen is also involved in the primary hemostasis as much as platelets being involved in secondary hemostasis [21].

Factor XIII Factor XIII (FXIII) is necessary for the stabilization of blood clot. FXIII is a transglutaminase that circulates in blood plasma as a tetramer composed of two A- (possessing catalytic site) and two B- (that act as carrier proteins for the A-subunits) subunits (A2B2) (Fig. 5). It is also present in platelets as a dimer composed of two A units. FacXIII is activated by thrombin and serine proteases like endogenous platelet acid protease and calpain by getting cleaved at A subunits. Fibrin polymer acts as a cofactor when it reaches a certain level (2% of total fibrinogen) to activate factor XIII. It also decreases the calcium requirement for plasma factor XIII activation. Factor XIII cross-links glutamine residues on one fibrin molecule to the lysine residues on another by forming strong isopeptide bonds. This occurs between C-terminal ends (Aα protuberances) of the Aα polypeptides as well as C-terminal ends of γ chains. These cross-links help strengthen the fibrin clot, making it resistant to any damage. FXIII also cross-links α_2 antiplasmin to α chain of fibrin, thereby preventing fibrinolysis by plasmin. FXIII val 34 leu mutation results in fibrin becoming unstable, causing increased lysis and seems to have a protective effect against venous thrombosis, which is primarily a fibrin-rich clot as opposed to arterial thrombosis, which is a platelet-rich clot. However, it is also associated with excessive bleeding tendencies [22].

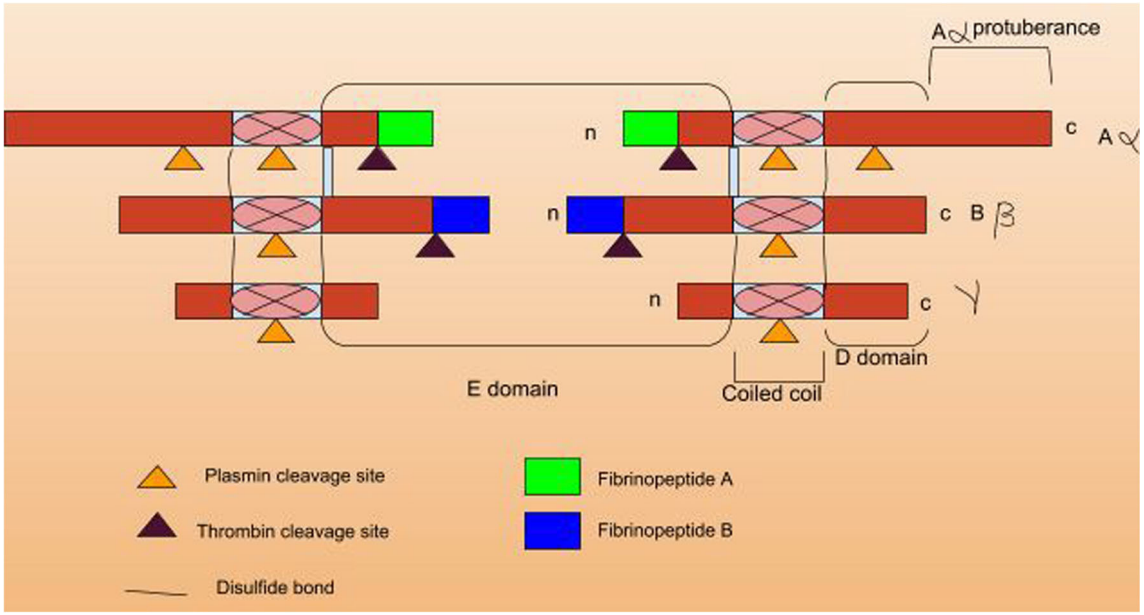

Fig. 4 Structure of fibrinogen

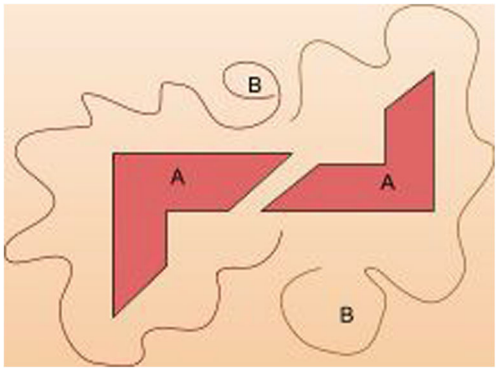

Fig. 5 Structure of factor 13

Antithrombotic Mechanisms

The blood clot performs its intended role of stopping the bleeding from the damaged endothelial site. Once the bleeding is stopped, the blood clot needs to be broken down, so that the normal perfusion of the organ distal to the vascular injury is restored. The blood clot is broken down into fragments by the fibrinolytic system, protein C/S system, and antithrombin III. In addition, intact endothelial cells play an import role preventing clot formation and propagation.

Fibrinolytic System Fibrinolysis is the last stage of the coagulation process, whereby the fibrin clot undergoes lysis to fibrin degradation products [23]. It is brought about by profibrinolytic agents like tissue plasminogen activator (tPa)/urine plasminogen activator (uPa) and Fac XIIa (Fig. 6). This process is counterbalanced by antifibrinolytic factors, which include plasminogen activator inhibitor (PAI), Plasmin inhibitor (PI) or α_2 antiplasmin and thrombin activatable fibrinolysis inhibitor (TAFI).

Protein C and S System The Protein C and S is the main system to terminate activated clotting process [24]. Thrombin, in the presence of endothelial receptor thrombomodulin and endothelial protein C receptor, activates protein C. Protein C, in the presence of cofactor protein S, cleaves FVa and FVIIIa, thereby inactivating them.

Antithrombin III Antithrombin III (AT III) is required to localize coagulation process to the site of injury [25]. It inactivates circulating thrombin by forming enzymatically inactive thrombin-antithrombin complexes.

Thrombomodulin (TM) is present in intact endothelial cells and binding of thrombin to TM changes its functions from being a procoagulant to anticoagulant. The thrombin-TM complex also activates TAFI.

At a rate of this activation is 1000-fold greater than thrombin alone TAFI stabilizes fibrin clot by proteolytic removal of terminal lysine residues from fibrin, where the fibrinolytic proteins bind.

TAFI also has anti-inflammatory function as it inactivates complement cVa.

Endothelial Cells Under normal conditions, endothelial cells possess anticoagulant activity due to several factors [26]. First, the presence of thrombomodulin on the cell surface. Second, ADPase activity prevents platelet aggregation. Finally, endothelial cells indirectly enhance the activity of antithrombin III and TFPI in plasma, which inactivates coagulation factors. Under conditions of endothelial injury or inflammation, endothelial cells become more procoagulant by decreasing TM expression and enhancing expression of TF (which normally is expressed by extravascular cells) and surface adhesion molecules.

Rebalancing of Coagulation Factors in Cirrhosis

Cirrhosis causes procoagulation and anticoagulation factors synthesized in the liver to decrease and subsequently, there is a compensatory increase in factors synthesized outside the liver (Table 1) [1, 27]. The liver does not synthesize FVIII and von Willebrand factor (vWF), which are produced in vascular endothelial cells. vWF level increases in patients with cirrhosis as a compensatory mechanism for decreased hepatic synthesis. This leads to increased FVIII binding and protecting it from cleavage by plasma proteases. The increased FVIII leads to increased production of thrombin, which in turn activates FVIII on platelet surface through non-PAR receptors.

Another factor, A disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13 (ADAMTS13), which is normally synthesized in the stellate cells of liver, is involved in cleaving the vWF. In severe liver disease, ADAMTS13 levels decrease, increasing the level of vWF, and resulting in the formation of platelet microthrombi occluding the sinusoidal microcirculation. These microthombi are believed to contribute to the progression of liver injury. Low levels of ADAMTS13 and high levels of vWF are also seen to be associated with decreased functional liver capacity, hepatic encephalopathy, hepatorenal syndrome, and intractable ascites. Abnormal ratios of ADAMTS13 and vWF are associated with reduced long term survival in patients with cirrhosis [1].

Platelet count decreases in patients with portal hypertension due to sequestration of platelets in the spleen as well as decreased production of thrombopoietin by the liver. The quantitative decrease is compensated by increase in platelet adhesion and aggregation. There is also platelet dysfunction due to increased nitric oxide and prostacyclin levels.

Fibrinogen levels increase in mild to moderate cirrhosis. In severe disease, levels decrease with accompanying dysfibrinogenemia due to increased sialic acid residues, which impairs fibrin polymerization and clot stabilization.

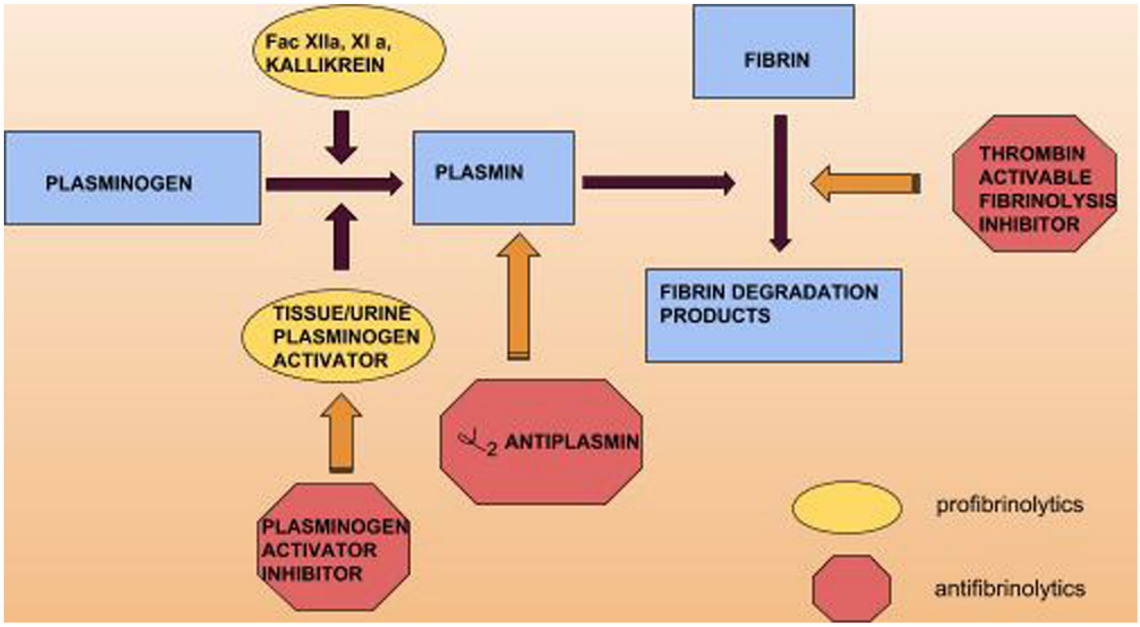

Fig. 6 Interplay of profibrinolytic and antifibrinolytic systems

The sum total of all these effects varies among individuals as some becomes coagulopathic, while others are prothrombotic. Similarly, variations in fibrinolytic system may have a beneficial impact by controlling thrombotic potential or, conversely, result in harm to do excessive bleeding. The net effect of this alteration in coagulation function should be ascertained in patients with cirrhosis before any surgical intervention. In addition, periodic assessments need to be made during the course of procedure, particularly in liver transplant, as well as in the postoperative phase, because the entire hemostatic system composition can change drastically in a matter of minutes. To assess the patient's coagulation status, blood tests are done periodically. These tests can either be conventional tests like prothrombin time (PT), international normalized ratio, and activated partial thromboplastin time (aPTT) or viscoelastic tests like thromboelastography and rotational thromboelastometry.

Viscoelastic Tests

Hemostatic interventions in liver transplant surgery should balance the need to temporize excessive bleeding with the risk of inadvertent clot formation along vascular anastomoses and elsewhere in the body. Excessive thrombotic tendency may result in the development of intracardiac thrombi during surgery. During liver transplant, the incidence of portal vein thrombosis is 10-15%, hepatic artery thrombosis is 5% and intracardiac thrombosis is 1.9% [28]. Hence, in the search for modalities that rationalize the use of less coagulation factors, viscoelastic tests (thromboelastogram—TEG, rotational thromboelastometry—ROTEM) have become more widely used during perioperative management of LT [29–32]. These tests reflect the interplay of coagulation factors, fibrinogen, and platelets in accordance with the current cell-based concept of coagulation, as opposed to the conventional tests, which measure the absolute platelet count, fibrinogen levels, and conform to the outdated cascade concept of coagulation [33•].

Table 1 Rebalancing of coagulation factors in end-stage liver disease

	Decreased (20–70%)	Increased (200%)
Procoagulant	Vit K dependent: II, VII, IX, X Vit K independent: V	vWF, f VII
Anticoagulant	Vit K dependent: Protein C, S Vit K independent: antithrombin III, ADAMTS—13	
Fibrinolytic	Plasminogen	Tissue plasminogen activator (TPA)
Antifibrinolytic	α_2 antiplasmin, thrombin activatable fibrinolysis inhibitor (TAFI)	Plasminogen activator inhibitor-1 (PAI-1)

Table 2 Specific reagents used in ROTEM

ROTEM test	Reagent
INTEM	Phospholipid, ellagic acid—similar to PTT
EXTEM	Tissue factor—similar to PT
FIBTEM	Cytochalasin-D (to inhibit platelets)—assesses fibrinogen qualitatively.
HEPTEM	Heparinase—assesses heparin effect comparing to INTEM
APTEM	Aprotinin (antifibrinolytic)—assesses fibrinolysis

Limitations of Conventional Coagulation Tests Conventional coagulation tests only measure the decrease in procoagulant factors without taking into account the decrease in anticoagulant factors as happens in liver disease [34]. They also do not reflect the progression, strength, and stability of the clot, because they are read at the time of early fibrin polymerization brought about by a very small amount of thrombin. An isolated elevation in aPTT, as happens in FXII deficiency or presence of heparin, tells little of its cause and cannot guide management algorithms. Similarly, PT elevation does not accurately predict prothrombin deficiency. Both may be prolonged in the presence lupus anticoagulant, which actually makes the patient prothrombotic in vivo. Therefore, a prolonged international normalized ratio (PT/INR) and aPTT does not necessarily translate to increased surgical bleeding. These tests were designed to monitor the administration anticoagulation therapy and were not intended for use perioperatively in a patient with liver disease. Often, algorithms centered around conventional tests for blood product management will lead to overtransfusion of blood products and increased incidence of acute lung injury, which is more common in cirrhotic patients than in general population [35]. Studies by Tapia et al. demonstrated the superiority of TEG over massive transfusion protocol by optimizing and reducing plasma and platelet transfusions in severely bleeding blunt trauma patients requiring damage control resuscitation [36•]. Westbrook et al. showed that protocol-based TEG management is superior to conventional tests during and after cardiac surgery by decreasing the blood product usage by 58% [37]. Romlin et al. demonstrated the earlier detection of coagulopathy with TEG in pediatric cardiac surgery with the possibility of initiating countermeasures [38]. In a review of 27 case reports of thromboembolic events during liver transplantation, TEG profiles were hypercoagulable in more than 70% of cases, whereas all conventional coagulation tests indicated hypocoagulability [39].

ROTEM vs TEG Both tests involve activating a sample of whole blood (Table 2) and analyzing the impedance caused by the clotting blood [33•, 40]. The analyzer used in TEG is electrical-mechanical as opposed to optical in ROTEM. The analyzed information is transmitted electronically and transformed into numerical and graphical read-outs. (Fig. 7, Tables 3 and 4).

ROTEM has been found to readily differentiate dilutional coagulopathy, thrombocytopenia, hyperfibrinolysis, and the presence of heparin compared to a basic TEG and therefore facilitates rapid institution of appropriate management perioperatively during liver transplantation [33•]. The speed of result acquisition is also faster with ROTEM than basic

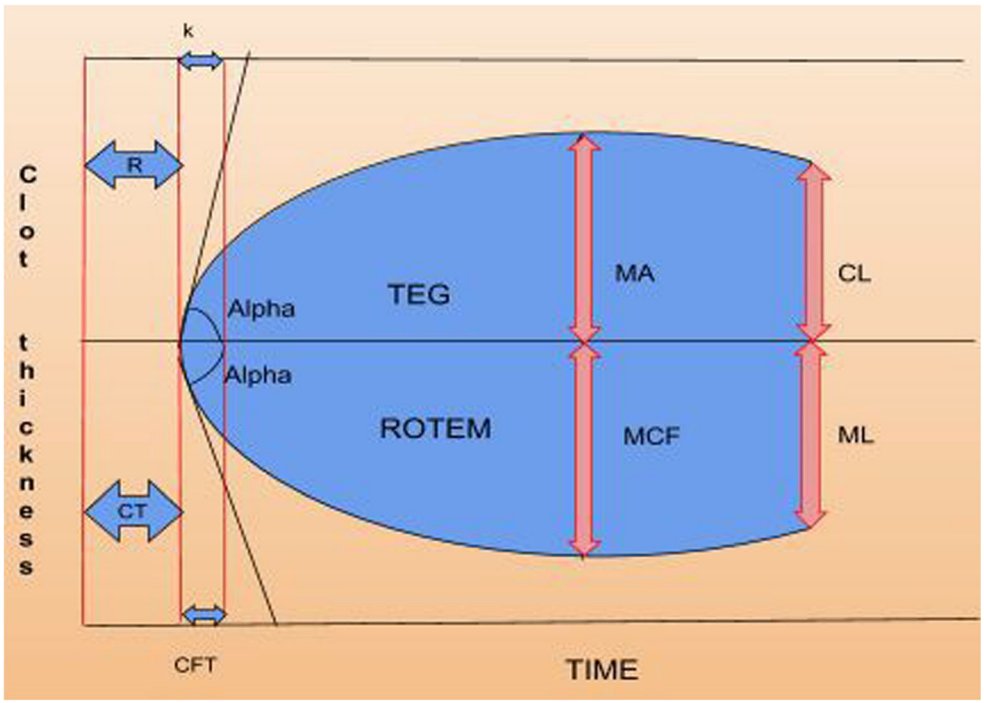

Fig. 7 Different terminologies used in TEG and ROTEM for the measured parameters

Table 3 Differences in terminologies used in TEG and ROTEM

ROTEM	TEG	Measure
Clotting time (CT)	Reaction time (*R*)	Time from start of measurement to beginning of blood clot or clot thickness of 2 mm
Clot formation time (CFT)	Kinetics time (*K*)	Time for clot thickness to increase from 2 mm to 20 mm
Alpha angle (α)	Alpha angle (α)	Angle of tangent at 2 mm amplitude
Maximum clot formation (MCF)	Maximum amplitude (MA)	Peak amplitude of clot
Maximum lysis, lysis index 30 (ML, LY 30)	Clot lysis 30, lysis 60 (CL 30, CL 60)	Percent reduction in MCF/MA after certain time

TEG, because ellagic acid used in ROTEM coagulates faster than Kaolin used in TEG [42]. We follow the ROTEM test in our institution, and our protocol is based on the same.

Interpretation and Management Based on ROTEM Results

The viscoelastic tests give a real-time analysis of blood clot dynamics. Initially, it identifies time to clot formation brought about by the coagulation factors involved in secondary hemostasis. Next, it assesses progression in the size and strength of the clot, brought about by interaction of platelets and fibrinogen and stabilized by FXIII, the speed with which the clot formation takes place and finally the degree of fibrinolysis that starts to dissolve the clot, simulating the progress of clot in vivo. [43–46] The parameter CT measures the time for the blood to start clotting and reach an amplitude of 2 mm, and this is facilitated by clotting factors (intrinsic pathway in INTEM and extrinsic pathway in EXTEM). The next step in coagulation (polymerization) is indicated by the parameter CFT, which measures the time from start of coagulation to an amplitude of 20 mm. This reflects early polymerization of fibrin and its interaction with platelets, facilitated by thrombin and calcium. Thus, CFT can be used to diagnose polymerization disorder. The next parameter, MCF is based quantitatively and qualitatively on the presence of fibrinogen and platelets and their interaction to produce a strong clot. Since MCF takes 30–40 min, A_{10}, which is the thickness of the clot in 10 min, can be used as an early indicator of MCF, as both of these parameters have been shown to have excellent correlation [47•]. MCF and A_{10} can be analyzed in both EXTEM or INTEM, but EXTEM is faster due to the shorter CT. Since these parameters depend on both fibrinogen and platelets, FIBTEM, which involves adding an antiplatelet agent to the blood sample like cytochalasin-D, indicates the solitary effect of fibrinogen on the strength of the clot. The α angle is an indicator of the speed of the clot formation, brought about by activated platelets in the presence of fibrinogen. If the angle is low in the presence of adequate amounts of fibrinogen, one may infer qualitative platelet deficiency. The last parameter, ML indicates lysis after MCF is achieved and gives an indication about the degree of in vivo fibrinolysis. To differentiate fibrinolysis from normal clot retraction, APTEM can be performed. This test involves adding the antifibrinolytic agent aprotinin to the blood sample (Table 5).

Table 4 Reference values for ROTEM [41]

	CT	CFT	α angle	A10	A20	MCF
INTEM	122–208	45–110	70–81	40–60	51–72	51–72
EXTEM	43–82	48–127	65–80	40–60	50–70	52–70
FIBTEM						8–24

The objective of following viscoelastic tests during liver transplant is to give the least amount of blood product possible to achieve reasonable clinical hemostasis. Towards this end, the recommendations suggested in our protocol (Fig. 8, Table 6) are guidelines that should be correlated in each clinical scenario. Periodic assessments can be made as more products are transfused.

The most crucial time in a liver transplant is immediately following the opening of the portal vein and suprahepatic inferior vena cava clamps at the time of reperfusion of the allograft. Massive fluid shifts, metabolic imbalances, cardiac dysrhythmias, and coagulation abnormalities happen during this time. The following section discusses the coagulation abnormalities that happen immediately after reperfusion.

Special Circumstances After Reperfusion

Hyperfibrinolysis Tissue plasminogen activator (tPa) is released in response to hypoxia, acidosis, use of vasoactive agents, and venous stasis during the anhepatic phase, and it persists in the circulation as it is not cleared by the liver. In addition, the absence of PAI-1, which is normally produced in the liver, leads to persistent activation of tPA. This phase persists until the new liver starts to function with gradually decreasing levels of tPA and fibrinolysis. If the fibrinolysis is very severe causing profound surgical bleeding, a small dose of antifibrinolytic like ε (epsilon)—aminocaproic acid 250–500 mg) or tranexamic acid (250 mg or 2.5 mg/kg) can be administered after confirmation of fibrinolysis by APTEM [50, 51].

Thrombocytopenia The already low platelet count is further lowered after reperfusion due to sequestration of platelets in the liver graft, platelet consumption from activation and

Table 5 ROTEM-based diagnostics

Parameter	Test	Interpretation	Management
CT prolongation	INTEM	Implies deficiency of clotting factors involved in Intrinsic pathway of coagulation cascade. It usually happens after reperfusion of the liver graft, when residual heparin from the donor graft causes the CT in INTEM to be selectively prolonged.	HEPTEM can confirm the presence of heparin. If associated with significant clinical bleeding, protamine is given to neutralize heparin.
	EXTEM	Tissue factor or FVII deficiency. FVII has the shortest half-life of 4–7 h and a long anhepatic phase or a tardive neograft can lead to its depletion sooner than the other factors.	FFP or FVII transfusion is indicated.
	Both	All clotting factors deficiency/depletion	FFP
MCF decrease	INTEM/EXTEM	Implies deficiency of building blocks of the clot, namely polymerized fibrin and platelets as well as factor 13, which stabilizes the fibrin clot.	Cryoprecipitate/fibrinogen concentrate and/or platelets
MCF decrease	FIBTEM	Implies fibrinogen deficiency. It is the maximum clot firmness after adding antiplatelet agent to the blood sample.	Cryoprecipitate/fibrinogen concentrate
α angle	INTEM, EXTEM	It indicates the speed of clot formation, which is mainly a function of platelets and to a lesser extent, fibrinogen and coagulation factors.	If the α angle is decreased even after correcting fibrinogen, platelets can be administered. Since platelet transfusion is associated with higher degree of postoperative morbidity and mortality, its use should be restricted to the neohepatic phase after hepatic artery anastomosis to minimize the incidence of hepatic artery thrombosis.
CFT	INTEM, EXTEM	It implies the initial rate of fibrin polymerization and a prolonged CFT happens in disorders of fibrin polymerization, low platelets or factor 13. Fibrin polymerization depends on the presence of fibrinogen, thrombin, and calcium.	A prolonged CFT needs to be analyzed with other parameters to determine which component is deficient and treated accordingly. A prolonged CFT with normal MCF indicates polymerization disorder.
ML increase	FIBTEM	If ML > 15% and especially if it occurs within 15 min (early lysis), it indicates significant fibrinolysis.	APTEM is done to confirm fibrinolysis and rule out clot retraction. If associated with significant clinical bleeding, antifibrinolytics are administered.

hyperfibrinolysis, thrombin generation, and hemodilution [52]. Even though the platelet count is low, the functional activity is often preserved as indicated by the α angle in ROTEM. If the angle is less than 60 after correction of

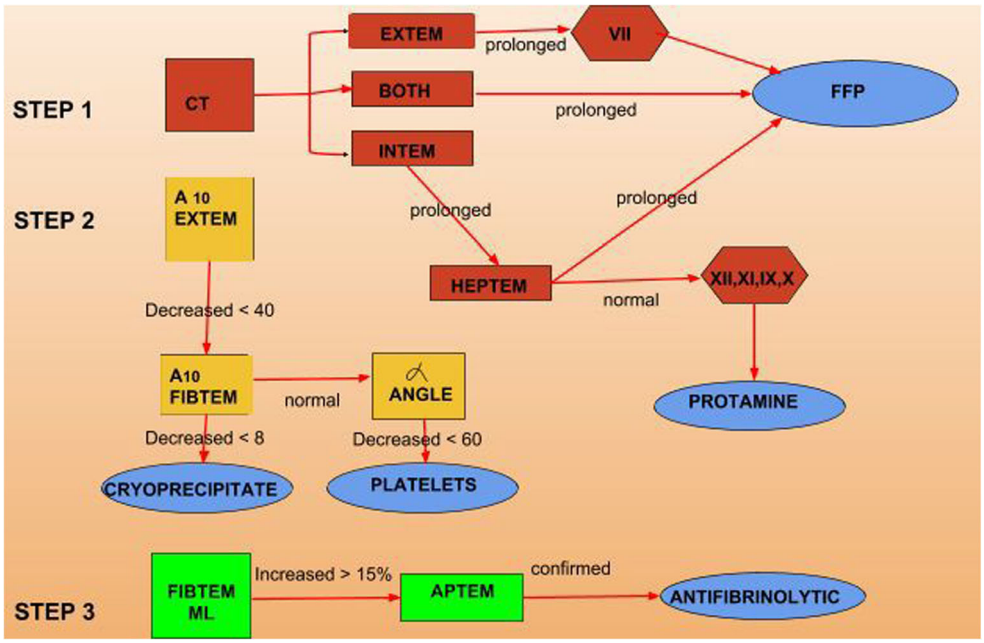

Fig. 8 Schematic ROTEM-based protocol for coagulation management

Table 6 Blood products and dosages

Product	Preparation	Contents	Dosage
Fresh frozen plasma	Prepared by collecting acellular fraction of blood and freezing at − 18*c within 6 h	Contains procoagulant factors like I, II,V, VIII, IX, X, XI, XIII, and anticoagulants like protein C, S, and antithrombin III. It also contains albumin, immunoglobulin and acute phase reactants	Only one third of the normal amounts of clotting factors is needed to maintain hemostasis. This corresponds to a PT/PTT prolongation of 1.5 times normal ($CT_{INTEM} > 300$ or $CT_{EXTEM} > 120$). The dose of FFP needed to increase clotting factors by 20–30% is 10–20 ml/kg.
Cryoprecipitate	Prepared by thawing FFP to 1–6*c, centrifuging and storing at − 18*c	Contains fibrinogen, von Willebrand factor, factor VIII, and fibronectin	1 unit of cryoprecipitate raises fibrinogen by approximately 5 mg/dl. Another formula: Number of cryounits needed to raise fibrinogen by 50–100 mg/dl = body weight (kg) ∗ 0.2. 1 unit of cryo contains 150 mg of fibrinogen and 80 IU of factor VIII. ROTEM based − fibrinogen dose (gm) = target increase in MCF fib (mm) × body weight (kg) / 140 [48]. The usual target of fibrinogen in FIBTEM is 8.
Platelets	Prepared from whole blood (500 ml yields 5.5 ∗ 10^10 platelets) or apheretic blood (500 ml yields 3 ∗ 10^11 platelets)	Platelets	1 unit of platelets increases count by 20–40 ∗ 10^9/lit. Once fibrinogen levels are corrected and α angle is still less than 60 or MCF INTEM/EXTEM < 45[44], platelets can be empirically administered and followed up in subsequent ROTEM
Fibrinogen concentrate	Prepared from pooled human plasma using the Cohn/Oncley cryoprecipitation procedure and stored as lyophilized powder at room temperature	Fibrinogen	Available as 900-1300 mg lyophilized fibrinogen concentrate powder for reconstitution with 50 ml of sterile water for injection. ROTEM based-fibrinogen dose (gm) = target increase in MCF fib (mm) × body weight (kg) / 140
Prothrombin complex concentrate	Prepared by ion-exchange chromatography from the cryoprecipitate supernatant of large plasma pools after removal of antithrombin and factor XI	F II, VII, IX, X, protein C and S	25 U/Kg, not to exceed 2500 U
FEIBA (FVIII inhibitor bypassing activity)	Prepared by lyophilizing sterile human plasma fraction	F II, activated VII, IX, X, protein C, and S. It contains equal unitages of factor VIII inhibitor bypassing activity and prothrombin complex factors	50–100 U/Kg every 6 h
Recombinant FVIIa [49]	Glycoprotein which is produced by recombinant DNA technology from baby hamster kidney cells	FVIIa	90 mcg/kg iv

fibrinogen and associated with significant clinical bleeding, platelets should be transfused after completion of the hepatic artery anastomosis to decrease the chance of arterial thrombosis.

Intracardiac Thrombi Thrombus formation in the cardiac chambers can happen at anytime during the transplant, but occur most commonly immediately after reperfusion [38]. Most thrombi are on the right side of the heart, with an incidence of 1–1.9%. The overall mortality is 68%. A left heart thrombus carries nearly a 100% mortality. Often, the right-sided thrombi are self-limiting due to natural fibrinolysis causing dislodgement and migration of small remnant thrombi to distal pulmonary circulation. If persistent and associated with hemodynamic instability, heparin (2000-U bolus) and/or tissue plasminogen activator (0.5 mg) can be administered, but this can be associated with massive hemorrhage. Once the clot lyses and hemodynamic stability is achieved, focus can be turned towards controlling bleeding cautiously. The patients who tend to have thrombotic tendencies include those with non-alcoholic steatohepatitis, autoimmune conditions like primary biliary cirrhosis/primary sclerosing cholangitis, and Budd-Chiari syndrome. Caution should be exercised in the use of prothrombotic products in these patients.

Heparin Effect Coagulopathy can be caused by release of accumulated heparin from the liver graft due to the large bolus administered to the donor [53]. This residual heparin leads to prolongation of clotting time in INTEM. It could also be caused by the non-removal of endogenous and exogenous heparinoids by the diseased liver and the newly reperfused liver. If clinically significant and confirmed by HEPTEM, a small dose of protamine can be administered to reverse the heparin effect.

Management of Thrombocytopenia in LT Platelet transfusion is an independent risk factor for decreased 1-year survival after liver transplantation [54]. Platelet count is only moderately reduced in cirrhotic patients and platelet dysfunction observed in vitro is probably less important than in vivo, because under flow conditions, the capacity of platelets for thrombin generation is not altered if thrombomodulin levels are corrected [55, 56]. Increased vWF release (10 times normal) seems to compensate for any decrease in platelet function [57]. Also, studies done with platelet function analyzer (PFA-100) have shown that increasing the hematocrit seemed to improve platelet adhesion/aggregation [58, 59]. Until more literature is available, it would be reasonable to develop a platelet dosage protocol based on analyzing the platelet function instead of the absolute count, with the understanding that even analyzing the platelet function in vitro may overestimate platelet function deficiency. [60, 61]

Conclusion

Transfusion practice during liver transplant should not be based on empirical methodology grounded in outdated principles, but rather on scientifically derived calculations from current evidence-based concepts of coagulation. Our article reviewed the contemporary understanding of the cell-based model of coagulation and explains the hematological differences in a patient with ESLD. We demonstrate the superiority of viscoelastic tests over conventional tests to diagnose and calculate the dosages of various blood products like cryoprecipitate and fresh frozen plasma, based on recent literature on the subject and validated by our own institutional practices.

Compliance with Ethical Standards

Conflict of Interest Arun Uthayashankar and Michael Kaufman declare no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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