

## Case Reports

# Severe Thrombotic and Bleeding Complications in a Baby with Heterozygous Factor V Leiden and Acquired von Willebrand Disease on ECMO

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**Abstract:** We aim to present the case of a 5-week-old girl with severe respiratory failure placed on veno-venous extracorporeal membrane oxygenation (ECMO) that was then switched to veno-arterial ECMO. She required up to 60 units/kg/hr of heparin to keep her heparin level within the target range at .3-.7 units/mL. During the ECMO course, substantial thrombus formation was observed within the venous site of the ECMO cannula, which led to two circuit changes on ECMO day 9 and day 20. On ECMO day 15, she was noticed to have purpuric lesions on her chest and her right hand with no obvious arterial or venous clot detected by Doppler ultrasound. She was also noted to have remarkable hemolysis as the plasma free hemoglobin levels were

substantially elevated up to 700 mg/dL. She was noted to have continuous oozing from the catheter insertion sites despite adequate underlying coagulation status. Her subsequent platelet function analysis, the thromboelastography, and thromboelastography platelet mapping suggested substantial platelet dysfunction. Her von Willebrand panel revealed absence of high molecular weight multimers. Further coagulation workup was prompted which revealed heterozygosity for factor V Leiden. The patient developed severe pulmonary hemorrhages and ECMO was discontinued on day 40. **Keywords:** extracorporeal membrane oxygenation, coagulation, anticoagulation, thrombosis, acquired von Willebrand disease, factor V Leiden. *JECT. 2011;43:64-69*

## OVERVIEW

Extracorporeal membrane oxygenation (ECMO) is based on temporary use of extracorporeal circulation for respiratory and/or cardiac support. Although it is a life saving tool, activation of the clotting system after exposure to the foreign surfaces may result in overt thrombosis, whereas the need for anticoagulation may lead to hemorrhage. Thrombosis and bleeding are the main cause of mortality and morbidity for patients on ECMO and the management of anticoagulation is challenging since

many of these patients may have an underlying disease at the time of ECMO initiation. Although various tests are available to monitor the coagulation status, there is no single parameter that can reliably predict the emergence of either thrombotic or hemorrhagic complications. Given this, there is wide variation between institutions in terms of anticoagulation regimens and the tests of coagulation used to monitor these patients (1).

In addition to the challenges by ECMO, unexpected complications may arise if the patient has an underlying risk factor for thrombosis. We present what we believe to be the first case found to have heterozygous factor V Leiden after developing severe thrombotic complications and limb ischemia while on ECMO. We discuss multiple coagulation parameters tested in the course of this patient and their implications.

The ECMO circuits at Texas Children's Hospital use the Jostra Quadrox<sup>D</sup> oxygenator (Maquet Cardiopulmonary

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AG, Hirrlingen, Germany), a Jostra Rotaflow® centrifugal pump (Maquet Cardiopulmonary AG, Hirrlingen, Germany), and the tubing packs are coated with Terumo X® coating, an amphiphilic, biopassive polymer coating (Terumo Cardiovascular Systems, Ann Arbor, MI). The ECMO coagulation panel at the Texas Children's Hospital consisted of the following measured parameters including prothrombin time (PT), activated partial thromboplastin time (PTT), PTT with heparinase, fibrinogen, heparin level measurement, D-dimer, and functional antithrombin (Table 1). The panel was ordered every 6 hours and 5.4 mL volume was drawn. The specimen was collected by normal methods.

The platelet count was measured separately at the same time. A plasma free hemoglobin level measurement was performed once a day using plasma hemoglobin orthotoluidine method. The plasma exchange was done with 1.0 plasma volume using fresh frozen plasma. Gambro COBE Spectra (Gambro, Inc., Lakewood, CO) was used. Prothrombin fragment 1.2 and thrombin-antithrombin complex were performed by Esoterix Laboratories (Denver, CO) using enzyme-linked immunosorbent assay method. Thromboelastogram (TEG) was performed from 3.2% citrated whole blood using kaolin as the activator. The platelet factor 4 (PF4) was performed by Esoterix Laboratories (Denver, CO) using enzyme-linked immunosorbent assay method. TEG platelet mapping was performed according to the company instructions. Both protein C and S were measured by clot-based assay using STA STACLOT PROTEIN C® and STA STACLOT PROTEIN S® (Diagnostica Stago, Inc., Parsippany, NJ), respectively. Von Willebrand factor (VWF) multimer analysis was performed by Blood Center of Wisconsin, Milwaukee using gel electrophoresis method. This study was approved by the Internal Review Board at Baylor College of Medicine.

## DESCRIPTION

The patient was a 5-week-old female, a previously healthy child born at term who had received all required vaccinations. The patient was transferred from an outside hospital with a 4-day history of rhinorrhea, cough, and

fever and was found to be respiratory syncytial virus (RSV) positive. She was intubated for rapidly progressive respiratory distress and quickly escalated to high frequency oscillatory ventilation. Inhaled nitric oxide was attempted for continued desaturations. Of note, her right leg appeared swollen on presentation. An attempt had been made to place a central venous line in her right groin at the previous hospital. Doppler ultrasound of the deep venous system demonstrated an incompletely occluding thrombus within the right common iliac vein. On hospital day 7, due to continued desaturations and hypotension, the patient was placed on veno-venous ECMO with a 13 F Avalon Elite® BiCaval dual lumen catheter (Avalon Laboratories, Rancho Dominguez, CA) via cannulation of the right internal jugular vein. The ECMO coagulation panel was performed every 4 hours during the ECMO course. Unfractionated heparin was started at 25 units/kg/hr and was titrated every hour with a goal of activated clotting time values between 180–200 seconds. The heparin level goal was .3–.7 units/mL. The goals for fibrinogen, PT, and PTT were >200 mg/dL, <17 seconds, and 70–93 seconds, respectively and PTT hepzyme <40 seconds. If any of these parameters were outside of the goal, the patient was transfused with appropriate blood components such as plasma or cryoprecipitate. The minimum target level for platelet count was 100,000/mm<sup>3</sup>. The goals for the antithrombin level were above 60–80%. If the level was below the target range, antithrombin concentrate was given (Table 1). Soon after initiation of ECMO, the patient needed progressively higher doses of heparin to keep the heparin levels within target limits with the dose escalating to 60 units/kg/hr. The antithrombin level was kept above 80% during this period and the fibrinogen level remained within normal ranges of 220–440 mg/dL.

On ECMO day 5, the patient suffered a brief cardiopulmonary arrest due to a malfunction of the circuit. On ECMO day 7, she had a second brief cardiopulmonary arrest secondary to worsening right heart failure and supra-systemic right ventricular pressures. On the same day, she was then converted to veno-arterial ECMO via cannulation of her right internal carotid artery with a 10 F cannula. On ECMO day 9, substantial thrombus formation was observed within the venous side of the ECMO

**Table 1.** Extracorporeal membrane oxygenation coagulation panel.

Tests	Products	Normal Range	Desired Range for ECMO	Purpose
PT	STA Neoplastine CI+®	12.2–15.5 seconds	<17 seconds	To assess the underlying coagulable state
PTT-hepzyme	Dade Hepzyme®	26.5–35.5 seconds	<40 seconds	
Fibrinogen	STA Fibrinogen®	220–440 mg/dL	>200 mg/dL	To assess the heparin effect
PTT	STA PII A5®	26.5–35.5 seconds	70–93 seconds	
D-dimer	STA Liatest D-Di®	<.4 µg/mL FEU		
Heparin level	STA Rotachrom Hep 4 UF®		.3–.7 units/mL	The range is determined for “normal” underlying coagulable state
Antithrombin	STA Stachrom AT III®	85–130%	>60–80%	To maximize the heparin effect

circuit, so the circuit was changed. On ECMO day 15, the patient's right upper extremity was noted to have some purpura and this progressed to a cold, purple hand. A Doppler ultrasound revealed good arterial and venous flow proximal to the hand, but poor flow in the hand. Over the course of the ECMO run, the purpura progressed up the arm. Concurrently, a purpuric rash was noted on the chest and upper arm, sparing abdomen and legs (Figure 1). Despite the intensive coagulation management, multiple thrombi were noticed within the ECMO circuit, which led to another circuit change on ECMO day 20.

The patient required multiple platelet transfusions to keep the count above the desired level as the platelets were rapidly consumed. The D-dimer levels elevated rapidly (Figure 2) and remained strongly positive as the levels of the prothrombin fragment 1.2 and the thrombin-antithrombin complex were markedly elevated at  $>4800$  pmol/L (normal 87–325 pmol/L) and 241.0 ng/mL (normal  $<5.1$  ng/mL) respectively; correlating with the clinical findings of substantial clot formation. The heparin-induced thrombocytopenia was ruled out by heparin antibody assay.

Throughout the ECMO course, the patient was noted to have remarkable hemolysis as the plasma free hemoglobin levels were substantially elevated up to 700 mg/dL. The patient required a total of five plasma exchange procedures to clear the plasma free hemoglobin from circulation, which is known to be strongly thrombogenic (Figure 3) (2).

The patient also experienced significant bleeding from the catheter insertion sites throughout the ECMO course. The TEG revealed very weak clot strength despite ade-

quate platelet counts above  $100,000/\text{mm}^3$  and adequate fibrinogen level above 200 mg/dL, suggestive of a functional platelet defect.

Further laboratory testing was performed to evaluate platelet function. TEG platelet mapping revealed substantially decreased aggregation of platelets with arachidonic acid and adenosine diphosphate with a platelet count of  $131,000/\text{mm}^3$ . A platelet function assay (PFA) showed prolongation of the closure time with collagen/epinephrine at  $>240$  seconds (normal ranges: 84–183 seconds) and with collagen/adenosine diphosphate at  $>251$  seconds (normal ranges: 69–126 seconds). Overall these were suggestive of a severe platelet function defect and/or defect in platelet-VWF interaction. To detect a possible acquired von Willebrand disease (aVWD), which also could be contributing to the continuous bleeding symptoms of the patient, a von Willebrand panel was performed which showed elevated factor VIII level at 346%, elevated VWF antigen at 302%, high-normal ristocetin cofactor activity was at 142%, and the antigen/activity ratio was decreased at .5. The multimer analysis was performed which revealed absence of high molecular weight VWF multimers, highly suggestive of aVWD (Figure 4).

Despite all the efforts done for coagulation management, the patient continued to have remarkable clot formation in the circuit and multiple systemic microthrombi, as evidenced by the worsening limb necrosis and purpuric skin lesions. Further coagulation workup revealed normal protein C and protein S levels for age at 76% and 49%, respectively. Prothrombin gene mutation was negative but factor V Leiden analysis revealed heterozygosity for factor V Leiden.

The patient developed severe pulmonary hemorrhages with no improvement in the lung parenchyma and ECMO was discontinued on day 40. The autopsy revealed end stage lung disease with extensive hemorrhage, bronchiectasis, and intimal hypertrophy of pulmonary arteries. The right hand digits and part of palm were gangrenous. Kidneys showed changes consistent with hemoglobinuria nephropathy and ongoing acute tubular necrosis.



**Figure 1.** Necrotic changes of right hand and purpuric lesions on the chest.

## COMMENTS

ECMO is a potential life saving tool in pediatric patients with cardiopulmonary failure. The nonbiologic surface of an extracorporeal circuit induces a massive inflammatory response leading to activation and thus, consumption of both procoagulant and anticoagulant components (3).

The principal causes of mortality and morbidity are bleeding and thrombosis. To prevent clot formation within the circuit, unfractionated heparin is widely used in most institutions, including our own. Advances in ECMO management have helped to reduce complications recently,

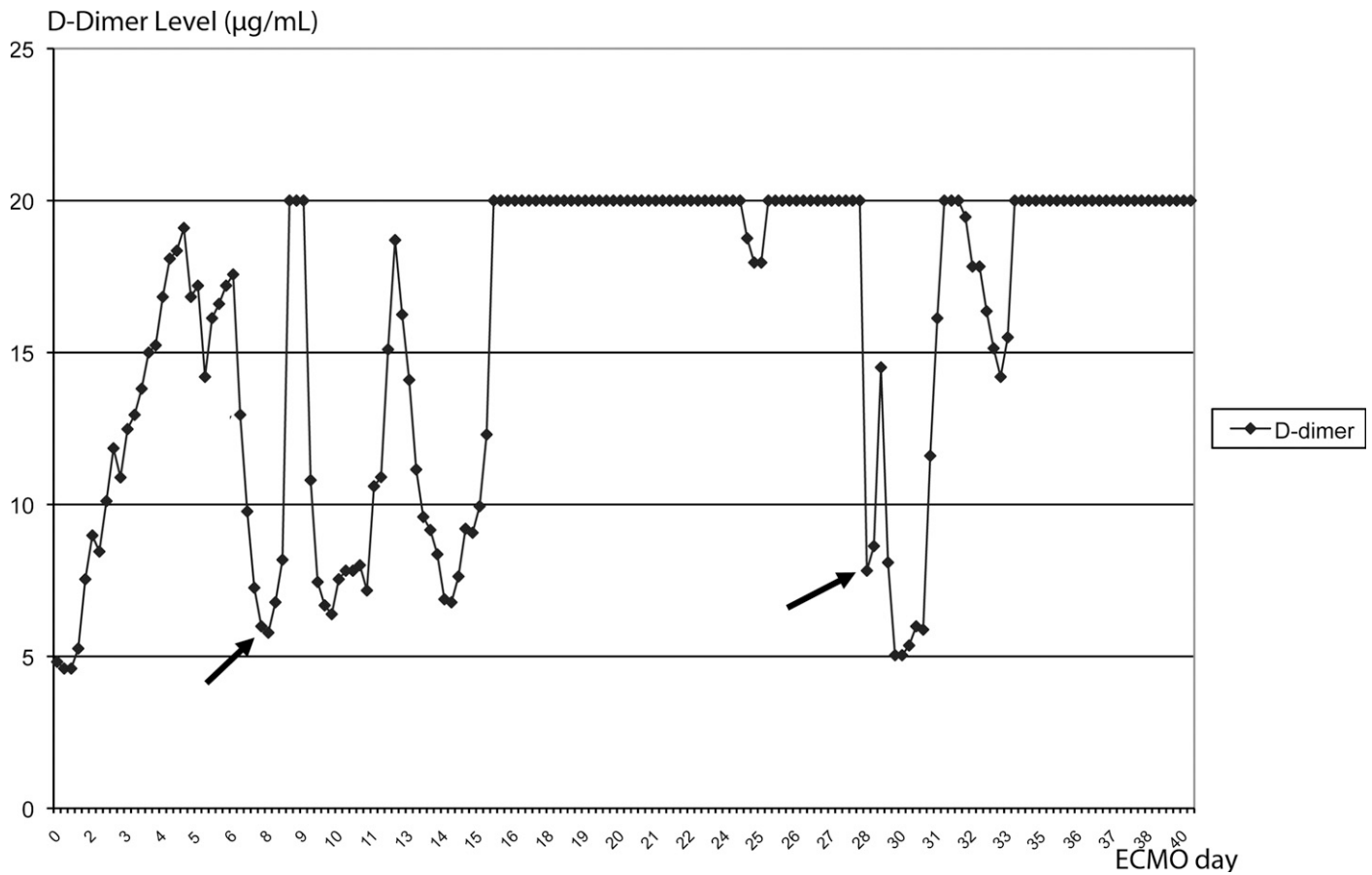


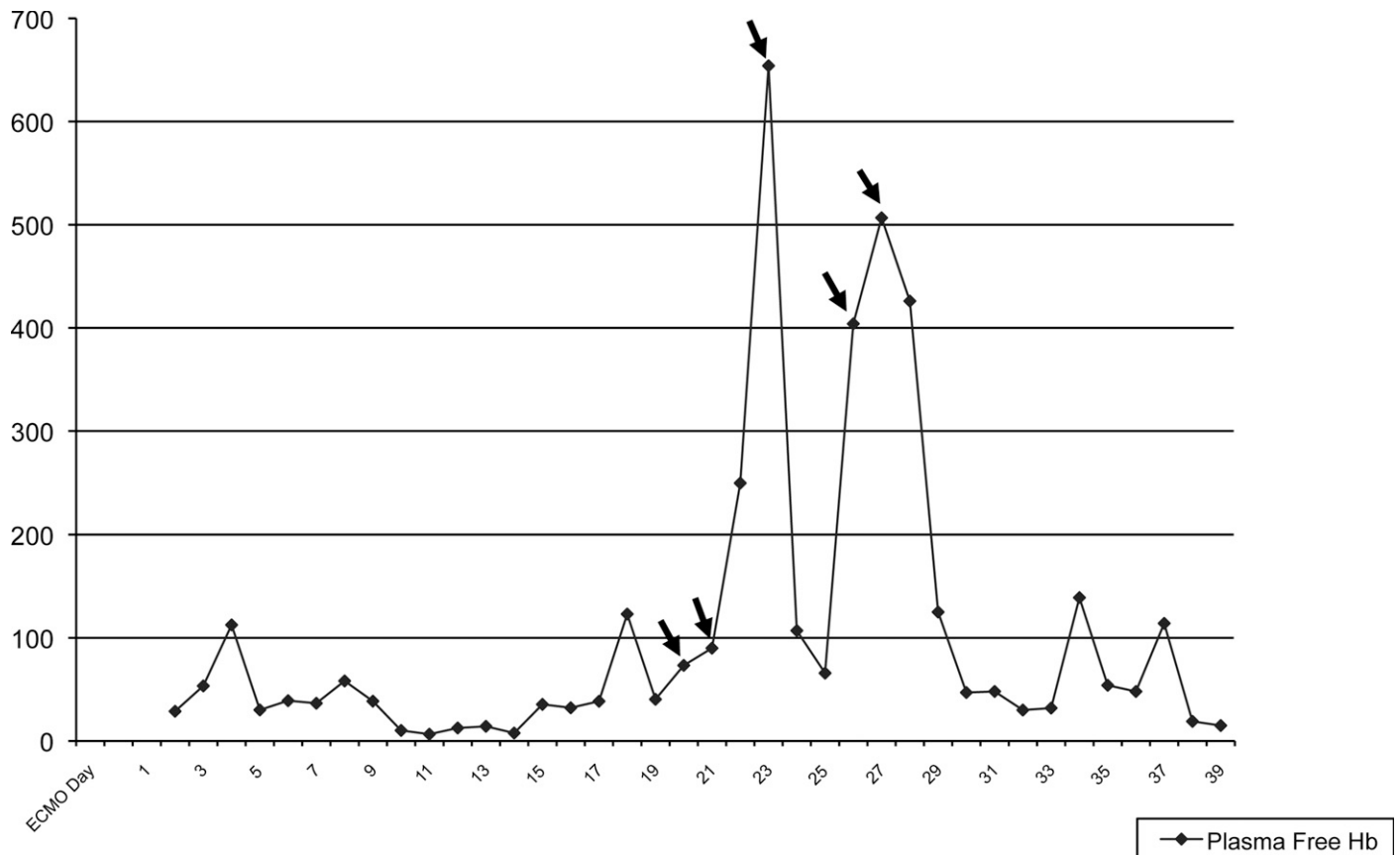
Figure 2. D-dimer level versus ECMO day. The arrows indicate ECMO circuit change.

but they still remain high primarily due to the immature coagulation system of infants and the underlying clinical condition of the patients before starting on ECMO. The vast differences in neonatal and adult anticoagulation and transfusion requirements demand tremendous clinical knowledge to provide the best care.

Here we report a patient who presented with severe RSV pneumonia and was placed on veno-venous ECMO and was then switched to veno-arterial ECMO. It was noticeable that, soon after initiation of ECMO, her heparin requirements quickly increased up to 60 units/kg/hr within a few days. The high amounts of heparin required were likely due to a disproportionately high rate of coagulation activation as well as the inhibition of heparin by the PF4 released from the activated platelets (4). PF4 is stored in the alpha granules of platelets and released upon activation. The heparin assay may also have been affected by the high level of plasma free hemoglobin caused by the hemolysis induced through the circuit and the microthrombi within the circuit (5,6). The substantial platelet activation and consumption through this process of widespread thrombus formation led to severe platelet dysfunction as was evident by prolonged closure time of PFA, weak

clot strength detected by the TEG, and decreased platelet aggregation detected by TEG platelet mapping.

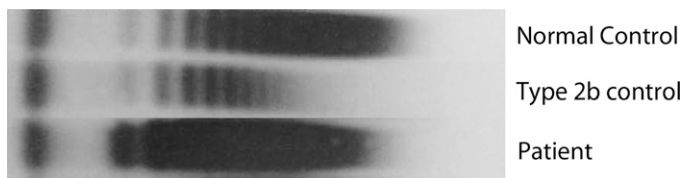
The patient experienced significant bleeding from the catheter insertion sites despite relatively adequate underlying coagulable state, platelet count, and fibrinogen level. The TEG with heparinase revealed shortened maximum amplitude despite normal platelet count and fibrinogen level. The continuous bleeding can be explained partially by the platelet dysfunction and loss of high molecular weight VWF multimers in the setting of aVWD. aVWD results from either a reduced rate of synthesis or an increased rate of clearance of the VWF. The latter may be caused by circulating antibody against VWF, by adsorption of VWF to malignant cells or activated platelets, or by proteolytic or mechanical degradation of the protein (7). The proportion of large VWF multimers is found to be decreased in aVWD. aVWD due to mechanical destruction of multimers has been described in various clinical conditions including in patients on ventricular assist device (8). We recently experienced two other patients on ECMO with ongoing bleeding complications and their testing revealed missing large molecular weight VWF multimers. We hypothesize that there is a similar mechanism of destruction of



**Figure 3.** Plasma free hemoglobin level versus ECMO day. The arrows indicate therapeutic plasma exchange.

multimers and emergence of aVWD in patients on ECMO as well. The high shear force caused by the platelet microthrombi and the ECMO circuit itself is likely to be responsible for the destructive process although this mechanism has not yet been widely studied in ECMO patients. If this phenomenon is shown in ECMO patients, possible treatment options for bleeding complications may include desmopressin and factor VIII/VWF concentrates.

Another interesting finding we observed in Figure 4 is that a darker staining of the lower molecular weight multimer portion will be appreciated compared to the controls. This could be explained by a compensatory increase in the production of VWF antigen as was observed in the patient. Whether this compensatory increase in the antigen levels can also compensate the relative decrease in the activity is yet to be understood. Further studies are



**Figure 4.** Von Willebrand factor multimer gel electrophoresis.

required to elaborate the exact mechanism of aVWD in ECMO patients and its impact in the clinical picture of the patient.

Despite high amounts of heparin anticoagulation, our patient experienced widespread clot formation as was evident by multiple visible clots within the ECMO cannula, limb necrosis, and purpuric skin rash which led to further testing including some hypercoagulable parameters which revealed heterozygosity for factor V Leiden. Factor V Leiden is a point mutation at amino acid 506 of coagulation factor V, which is one of the three cleavage sites for activated protein C. Factor V Leiden mutation, protein C, S, and antithrombin III deficiencies, and the presence of antiphospholipid antibodies were present in up to 30% of children with both venous and arterial thrombotic events (9). Heterozygous factor V Leiden per se is not a strong risk factor for thrombosis in otherwise healthy infants and it possesses a 5- to 10-fold increased risk for thrombosis whereas homozygous genotype is associated with a 50- to 100-fold increase (10,11). In those children who develop thrombotic complications, more than one risk factor for thrombosis is usually encountered (12). Due to the activation of clotting cascade and platelets, ECMO circuit possesses a significant risk of clotting for the patients. If there

is an underlying hypercoagulable state, the risk is even more as in our patient. Yet, it is difficult to predict the overall thrombotic risk in these patients as they have multiple risk factors simultaneously.

One point that was remarkable for this patient was the rapid increment in the D-dimer levels. In one of our previous studies regarding 21 patients on ECMO, we plotted the D-dimer versus time to see the trend in D-dimer. According to that data, the D-dimer becomes positive soon after ECMO initiation and doubles in 4 days and quadruples in 10 days (13). In our patient we presented here, the D-dimer level doubled the day after ECMO initiation and quadrupled in 5 days, suggestive of rapid activation of the coagulation cascade. One other possibility of a rapid rise in the D-dimer level is increased fibrinolysis as in a septic state. In our patient, the TEG reading did not show any evidence for increased fibrinolysis. One should carefully assess the trend in D-dimers as this information might prompt the clinician to suspect an underlying risk factor for thrombosis and search for them. In our institution, we do not perform hypercoagulable tests for ECMO patients on a routine basis, but it may be considered early during the course for patients with unexpected thrombotic complications. One can also consider TEG as a potential tool to detect an existing hypercoagulable state, but in our patient there was no evidence of a hypercoagulable state, but the persistently low maximum amplitude was suggestive of a hypocoagulable state.

The question of how to manage those patients is of concern. Depending on the risk factor detected, there could be a possible management plan offered. On the other hand, any intervention to overcome the hypercoagulable risk might lead to significant bleeding complications as these patients already have high risk of bleeding due to consumptive coagulopathy.

## CONCLUSION

The coagulation management for babies and infants on ECMO is challenging due to their immature coagulation system, especially if there is an underlying coagulopathy. One should be suspicious about inherited coagulation disorders if a patient on ECMO has unexpected thrombotic

complications. Hypercoagulability workup may be considered before or soon after starting ECMO for patients who develop unexpected thrombotic complications.

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