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Authors

Matthay, Zachary
Hellmann, Zane
Nunez-Garcia, Brenda
et al.

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Post-Injury Platelet Aggregation and Venous Thromboembolism

Zachary A. Matthay, MD[1], **Zane J. Hellmann, MD**[2], **Brenda Nunez-Garcia, BS**[1], **Alexander T. Fields, PhD**[1], **Joseph Cuschieri, MD**[1], **Matthew D. Neal, MD**[3], **Jeffrey S. Berger, MD**[4], **Elliot Luttrell-Williams, BA**[4], **M. Margaret Knudson, MD**[1], **Mitchell J. Cohen, MD**[5], **Rachael A. Callcut, MD MSPH**[6], **Lucy Z. Kornblith, MD**[1]

[1]Department of Surgery, Zuckerberg San Francisco General Hospital/University of California San Francisco, San Francisco, CA

[2]Department of Surgery, Yale University, Hartford, CT

[3]Department of Surgery, University of Pittsburg, Pittsburg, PA

[4]Department of Medicine, New York University Grossman School of Medicine, New York, NY

[5]Department of Surgery, University of Colorado, Aurora, CO

[6]Department of Surgery, University of California Davis, Sacramento, CA

Abstract

Introduction: Posttraumatic venous thromboembolism (VTE) remains prevalent in severely injured patients despite chemoprophylaxis. Importantly, although platelets are central to thrombosis, they are not routinely targeted in prevention of posttraumatic VTE. Further, platelets from injured patients show *ex-vivo* evidence of increased activation yet impaired aggregation, consistent with functional exhaustion. However, the relationship of this platelet functional phenotype with development of posttraumatic VTE is unknown. We hypothesized that following injury impaired *ex-vivo* platelet aggregation (*PA)* is associated with the development of posttraumatic VTE.

Methods: We performed a secondary analysis of 133 severely injured patients from a prospective observational study investigating coagulation and inflammation (2011-2019). *PA* in response to stimulation with adenosine diphosphate (ADP), collagen, and thrombin was measured at presentation (pre-resuscitation) and 24h (post-resuscitation). Viscoelastic clot strength and lysis were measured in parallel by thromboelastography. Multivariable regression examined relationships between *PA* at presentation, 24h, and the delta between presentation and 24h with development of VTE.

Results: The 133 patients were severely injured (median injury severity score 25) and 14% developed VTE (all >48 hours after admission). At presentation, platelet count and *PA* were not

significantly different between those with and without incident VTE. However, at 24h, those who subsequently developed VTE had significantly lower platelet counts (126 x10^9/L vs 164 x10^9/L, p=0.01), and lower PA in response to ADP (p<0.05), collagen (p<0.05), and thrombin (p=0.06). Importantly, the magnitude of decrease in *PA* (Delta) from presentation to 24h was independently associated with development of VTE (adjusted odds ratios per 10 aggregation unit decrease: Delta-ADP 1.31, p=0.03; Delta-collagen 1.36, p=0.01, Delta-thrombin 1.41, p<0.01).

Conclusion: Severely injured patients with decreasing *ex-vivo* measures of platelet aggregation despite resuscitation have an increased risk of developing VTE. This may have implications for predicting development of VTE and for studying platelet targeted chemoprophylaxis regimens.

Level of evidence: level III, prognostic

Keywords

Platelets; platelet aggregation; platelet function tests; blood coagulation disorders; trauma; venous thromboembolism; deep vein thrombosis; pulmonary embolism

Background:

Posttraumatic venous thromboembolism (VTE) remains an important cause of morbidity and mortality despite use of chemoprophylaxis (1, 2). Deep vein thrombosis (DVT) can be detected by screening ultrasound in over a quarter of critically injured patients, and an estimated 12% of trauma deaths are attributable to fatal pulmonary embolism (PE) (2, 3). Clinical risk factors for the development of posttraumatic VTE have been well described (1, 3, 4), but our understanding of the specific changes in coagulation biology associated with posttraumatic VTE is incomplete, and as such routine standard-of-care in posttraumatic VTE prevention remains primarily limited to heparin-based chemoprophylaxis.

Severe tissue injury and hemorrhagic shock are known drivers of trauma-induced coagulopathy (TIC), encompassing a range of coagulation derangements in the platelet, endothelial, and fibrinolytic systems associated with both early hypocoagulability and subsequent hypercoagulability, thrombo-inflammatory complications, and mortality (5-9). *Ex-vivo* evidence of increased platelet activation yet impaired platelet aggregation is a principal finding in TIC, independent of platelet count (10-12). This is commonly identified after injury, but to what degree this reflects pathologically impaired *in-vivo* platelet function remains unknown (5, 9-12). It is hypothesized that this seemingly discordant biology of increased platelet activation, but impaired aggregation may represent a functionally exhausted platelet phenotype, in which the circulating platelets sampled were activated at sites of tissue injury, have degranulated, and are therefore unable to respond further to stimulation in *ex-vivo* assays (13, 14). *Ex-vivo* impairments in platelet aggregation are known to be independently associated with microvascular thrombo-inflammatory complications and mortality after severe injury (10, 11). However, whether these impairments in platelet aggregation are also associated with macrovascular thrombotic complications, including the development of posttraumatic VTE is not well understood (15). Therefore, investigating how impaired *ex-vivo* platelet aggregation is associated with the development of posttraumatic VTE is of particular interest given the central role platelets

play in thrombosis, and the lack of routine platelet targeted chemoprophylaxis regimens (16, 17).

In this study, we examined the relationship of *ex-vivo* platelet aggregation immediately after injury on presentation (pre-resuscitation), at 24 hours after injury (post-resuscitation), and the change in platelet aggregation from presentation to 24 hours after injury with subsequent development of posttraumatic VTE in a cohort of severely injured trauma patients admitted to the intensive care unit (ICU). We hypothesized that following injury, impaired *ex-vivo* platelet aggregation (*PA*) is associated with the development of posttraumatic VTE.

Methods:

Patient Enrollment

Whole blood was collected in sodium citrate (3.2%) from 1,024 injured patients on arrival to the emergency department as part of a longitudinal prospective observational study examining coagulation and inflammation after injury at a Level 1 Trauma Center (2010-2019) (11, 13, 18-20). Patients who required the highest-level trauma activation were included, unless in-custody, pregnant, pediatric, suffering from >20% body surface area burns, transferred from another facility, did not require ICU level care, or were atraumatic. For the purposes of this post-hoc analysis, we further limited our study cohort to the subset of 133 patients who were admitted to the ICU 24 hours, had platelet aggregometry performed on presentation and at 24 hours from presentation (to be able to examine platelet aggregation both pre- and post-resuscitation), and who were admitted to the hospital for 3 days (to be able to examine those at risk of development of VTE) (Supplemental Figure 1). Patients were additionally excluded if they were confirmed to be taking antiplatelet or anticoagulant agents prior to admission (Supplemental Figure 1). Demographics, injury and physiologic data, and clinical outcomes were prospectively collected in parallel with laboratory assays. Consent was obtained under an initial waiver of consent with subsequent informed consent from all patients, under approval by the Committee on Human Research (IRB #19-28933). This study conforms with the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines and a complete checklist has been uploaded as Supplemental Digital Content (Supplemental Table 1).

Identification and Definition of Venous Thromboembolism

Venous thromboembolism (VTE) was defined as development of radiologically confirmed pulmonary embolism (PE) or deep vein thrombosis (DVT, upper or lower extremity) diagnosed after admission. No screening protocols were used and all VTEs were considered symptomatic in this study. Chemoprophylaxis and treatment was determined by the treating clinician. We excluded isolated subsegmental pulmonary clots identified on presentation imaging from our diagnosis of pulmonary embolism, as these have been shown to have different pathophysiology and clinical implications as compared to the development of posttraumatic VTEs after presentation (21, 22).

Platelet aggregometry

Platelet aggregation (*PA)* was measured *ex-vivo* by multiple electrode platelet aggregometry (Multiplate®, Verum Diagnostica GmbH; Munich, Germany) immediately after sample collection as described previously (11, 23). Briefly, whole blood was diluted with 3mM $CaCl_2$ in warmed normal saline, incubated at 37°C and stirred for 3 minutes in a Multiplate® test cell containing two copper electrodes. Platelet aggregation was stimulated by the surface receptor stimulants adenosine diphosphate (ADP; final concentration 6.5μM; via P2 receptors), thrombin receptor activating peptide-6 (thrombin; final concentration 32μM; via PAR receptors), and collagen (final concentration 3.2μg/mL; via GpIa/IIa and GpVI receptors). Platelet aggregation was quantified in aggregation units (AUC) by measuring the average of the change in electrical impedance at the two electrodes in each test cell over a 6-minute interval. The manufacturer provided reference ranges for the 6-minute aggregation in response to platelet stimulation by each stimulant are: ADP (36-101U), thrombin (75-137U), and collagen (24-79U). Platelet aggregation was measured on presentation and at 24 hours for all patients included in this study. The delta in platelet aggregation from presentation to 24 hours was calculated by subtracting the platelet aggregation in response to each stimulant at 24 hours from the platelet aggregation response to each stimulant at presentation.

Viscoelastic Assays

Viscoelastic testing was performed using the TEG-5000 Thrombelastography Hemostasis Analyzer according to manufacturer instructions (24) given the central role of platelet function in clot formation and breakdown. Briefly, on presentation (pre-resuscitation) and at 24 hours (post-resuscitation), citrated kaolin TEG (CK-TEG) was run in parallel with platelet aggregometry described above using a parallel sample of citrated whole blood. CK-TEG values generated include activated clotting time (ACT, min), angle (rate of clot strength increase, degrees), maximum clot strength achieved (MA, millimeters) and percent clot lysis 30 and 60 minutes after reaching MA (LY30 and LY60, %).

Statistical Analysis

Study patient characteristics, laboratory data, and outcomes are presented as mean (± standard deviation), median (interquartile range[IQR]), or percentage; univariate comparisons were made using Student's *t* test for normally distributed data, Wilcoxon rank sum or Kruskal-Wallis testing for nonparametric data, and Fisher's exact test for proportions. Based on previously published data of platelet aggregation in trauma patients (11, 23) (mean of 50 and SD ±20 aggregation units [AUs] in response to ADP), our study had 80% power to detect an effect size of 15 AUs between those with and without VTE with alpha set at 0.05. Bivariate and multivariable logistic regression tested the association of platelet aggregation and viscoelastic measures and clinical risk factors for the development of VTE. The multivariable logistic regression models were built testing risk factors for VTE and known potential confounders in the relationship of platelet aggregation with development of VTE (including platelet count, chemoprophylaxis timing, transfusions, length of stay, shock, and injury severity). The Akaike Information Criterion was used to further assess and optimize model fit on sensitivity analyses to examine the

relative importance of the covariates included (25). The odds ratios reflect the estimated effect for each 10 unit change in platelet aggregation units (AUC) to allow for clinically meaningful interpretation of the results, given the known median and interquartile ranges in this population and in prior studies of trauma patients (13, 19, 23). An [alpha] < 0.05 was considered significant. Analyses were carried out in Stata version 15.1 (StataCorp, College Station, TX).

Results

The patients were young with a median age of 38, primarily male (88%), severely injured with an injury severity score of 25 (IQR 14-33), and an in-hospital mortality rate of 12% (Table 1). The majority (69%) were bluntly injured, and 49% suffered a traumatic brain injury (Table 1). Transfusion of blood products was performed in 65% of patients in the initial 24 hours, and 23% received a platelet transfusion (Table 1). Out of the 133 patients, VTE was detected in 18 patients including 8 with PE, 9 with DVT, and one with both (Table 1 and Supplemental Table 2). The median time to development of VTE was 6 days with a range of 2-29 days- 33% occurred in the first 2-5 days, 23% within 5-10 days, and 44% in 10-30 days (Supplemental Figure 2).

Patients who developed VTE had significantly higher injury severity scores (29 [14-30] vs 22 [27-34]) longer hospital (28 [21-28] vs 16 days [9-24]) and ICU lengths of stay (14 [7-23] vs 5 [3-11] days), and fewer ventilator-free days (18 [5-24] vs 25 [20-27] days, Table 1, all $p<0.05$). Presentation platelet counts were not different between those with and without development of VTE ($260x10^9$/L [$209 x10^9$/L - $322 x10^9$/L] vs $263x10^9$/L [$207 x10^9$/L - $299 x10^9$/L]), but at 24 hours, those who subsequently developed VTE had significantly lower platelet counts ($126 x10^9$/L [$106 x10^9$/L - $153 x10^9$/L] vs $164 x10^9$/L [$127 x10^9$/L – $201 x10^9$/L], $p=0.01$, Table 1). A significantly lower proportion of patients who subsequently developed VTE had initiation of VTE chemoprophylaxis by 72 and 96 hours ($p<0.05$, Table 1). When examining injury mechanisms, there were non-significant trends for increased rates of multiple rib fractures, and hemo- or pneumothorax among those who developed VTE, and a significantly higher proportion of patients with multiple long bone fractures ($p<0.01$, Table 1).

There were no significant differences in platelet aggregation parameters on presentation between those who subsequently developed VTE and those who did not (Table 2). However, at 24 hours, mean platelet aggregation responses to ADP and collagen were significantly lower in those who developed VTE (19 [12-25] vs 28 [20-40], and 17 [15-25] vs 32 [22-48] aggregation units [AU], both $p<0.05$), with a similar but non-significant trend for platelet aggregation in response to thrombin (47 [37-75] vs 66 [49-81] AU, $p=0.06$, Table 2). Given the known wide variability in platelet responsiveness to *ex-vivo* stimulation across individuals (26), we also examined the change in platelet aggregation for each individual from presentation to 24 hours. We found that the magnitude of the change (delta) in platelet aggregation in response to each of the surface stimulants (ADP, collagen, and thrombin) was significantly larger (more negative) in those who developed VTE (all $p<0.05$, Table 2). We additionally examined viscoelastic measures of clot formation and breakdown, which

demonstrated that those who developed VTE had significantly lower percent clot lysis at 24 hours (p<0.05, Table 2).

We then performed multivariable logistic regression to further characterize the association of platelet aggregation with the development of VTE, controlling for key potential confounders. On presentation, there was a weak association of increasing platelet aggregation with increased odds of development of VTE, reaching statistical significance for platelet aggregation in response to thrombin (p=0.05), and similar but non-significant trends for platelet aggregation in response to ADP and collagen (Figure 1 and Supplemental Table 3). There were also no significant associations of either viscoelastic clot strength or clot lysis on presentation with subsequent development of VTE (Figure 2 and Supplemental Table 3).

At 24 hours however, there were some, although not all, significant associations of platelet aggregation and clot lysis with development of VTE. While there was a significant trend for the association of impaired *ex-vivo* platelet aggregation in response to collagen with development of VTE, there was no significant association with platelet aggregation in response to ADP or thrombin (Figure 1 and Table 3). Importantly, the relationship of development of VTE with platelet aggregation was most pronounced when examining the change (delta) in platelet aggregation from presentation to 24 hours. The larger the magnitude of decrease in platelet aggregation in response to ADP, collagen, and thrombin stimulation, the higher the odds of development of VTE were (OR 1.3-1.4 per 10u decrease in AUC, all p<0.05, Figure 1 and Table 3). Further, impaired clot lysis at 24 hours was also an important risk factor for the development of VTE with an adjusted odds ratio of 2.5 for every 1% decrease in clot lysis (p<0.05, Figure 2 and Table 3).

Discussion

In this study, we examined the associations between post-injury *ex-vivo* platelet aggregation and risk of development of posttraumatic VTE in a cohort of severely injured trauma patients admitted to the intensive care unit. Our examination identified that the magnitude of decrease in *ex-vivo* platelet aggregation in the initial 24 hours of hospitalization for severely injured patients is a significant risk factor for development of VTE, independent of platelet count and known VTE clinical risk factors. This finding is of importance, as it may have implications for future studies examining targeted approaches to identifying patients at higher risk for development of VTE who may benefit from earlier initiation of chemoprophylaxis, and/or of the addition of platelet targeted chemoprophylaxis to current heparin-based regimens. Further, the use of the change (delta) in platelet aggregation may reduce challenges of the known wide variation in responses to *ex-vivo* assays of platelet aggregation across individuals and assist with a more personalized approach to assessing platelet function.

Evidence for the role of platelets in the pathobiology of VTE has been explored across disciplines (27-32), though fewer studies have examined this specifically in trauma patients (15, 33). Increased understanding of both the hemostatic and immune properties of platelets suggests that across diseases, damage associated molecular patterns triggers

platelet-monocyte and platelet-neutrophil interactions leading to pro-inflammatory and hypercoagulable states, promoting endothelial injury and thrombotic complications (28, 31, 34, 35). These platelet-immune cell interactions cause release of cytokines, platelet microparticles, as well as increased tissue factor and thrombin production (31, 33). Procoagulant platelet microparticle release may be an important mechanism by which activated platelets after injury mediate the risk of hypercoagulable complications based on human and murine studies (33). Platelets also have important effects on the downregulation of fibrinolysis via release of plasminogen activator inhibitor-1 and alpha antiplasmin-2, thereby regulating clot breakdown (36, 37).

However, the changes in platelet hemostatic functions in the setting of TIC are heterogenous and dynamic with evidence that tissue injury may precipitate platelet activation and hypercoagulability, while hypoperfusion may trigger release of soluble mediators that may inhibit platelets or cause cytoskeletal and structural defects (23, 38-42). Understanding the true *in-vivo* biology of platelets after injury is further complicated by limitations of *ex-vivo* assays of platelet function and divergent findings of increased platelet activation yet impaired aggregation in *ex-vivo* assays from trauma patients (10, 13, 43). Important to our findings, impaired *ex-vivo* platelet aggregation in the setting of injury may not necessarily equate to a deficit of platelet hemostatic function *in-vivo*- but rather a functionally exhausted phenotype in which the sampled platelets have already been activated and degranulated at local sites of injury *in-vivo*, and therefore have reduced capacity to further aggregate when stimulated *ex-vivo*. We hypothesize from the results of our study that the decrease in *ex-vivo* platelet aggregation from presentation to 24 hours reflects a functionally exhausted platelet phenotype that is part of a hypercoagulable state *in-vivo* contributing to subsequent risk of development of VTE. Patients with the largest decrease in *ex-vivo* platelet aggregation following resuscitation may be those who experienced robust platelet activation and associated degranulation, leading to a population of circulating yet functionally exhausted platelets. This hypothesis is supported by prior studies of *ex-vivo* platelet function in trauma patients (10, 13), including one prior study based on an analysis of the Pragmatic, Randomized, Optimal Platelet and Plasma Ratios (PROPPR) study database which identified similar post-resuscitation deficits in *ex-vivo* platelet aggregation in trauma patients who developed VTE compared to those who did not (15). However, further studies are needed to test this hypothesis, given the lack of direct data examining platelet surface receptor markers of activation or platelet microparticle plasma profiles in this cohort.

Importantly, we also found a significant independent association between decreased viscoelastic clot lysis at 24 hours and increased risk of development of VTE, but not for overall clot strength nor for clot lysis on presentation. Under our hypothesis that patients with severe injury experience robust platelet activation, degranulation, and downstream functional exhaustion, it is plausible that platelet degranulation releasing plasminogen activator inhibitor-1 and alpha antiplasmin-2 participates in the downregulation of fibrinolysis. Several studies have detected a range of aberrations in fibrinolysis pathways after major trauma, with important associations with mortality for both patient with hyperfibrinolysis as well as those with impaired fibrinolysis or fibrinolytic shutdown (44, 45). To our knowledge, few studies have examined the relationship of impaired fibrinolysis with posttraumatic VTE development, though the Consortium of Leaders in the study of

Traumatic Thromboembolism (CLOTT) Study investigators have emerging data examining impaired fibrinolysis as a significant risk factor for the development of posttraumatic VTE (21). In a study by Moore and colleagues, no association was identified between impaired fibrinolysis on presentation and development of posttraumatic VTE development, though data at 24 hours were not examined (46). Overall, because platelets are essential regulators of fibrinolysis, further examinations of the role platelets play in impaired fibrinolysis and associated worse outcomes after injury including the development of VTE is warranted (46, 47).

Limitations and future directions

This analysis has important limitations to consider, including its retrospective nature and limited sample size. The detection and diagnosis of VTE in this study was not strictly protocolized, but rather determined by clinical judgement. Therefore, our VTE rate of 14% is likely an underestimate, given prior studies using screening imaging have shown VTE in over a quarter of critically injured patients (2). The patients included in this study were composed from a cohort of severely injured patients admitted to the ICU for at least 24 hours and to the hospital for at least 3 days, and therefore the results may not be generalizable to a broader population of trauma patients, including those who expire early and those have relatively minor injuries and shorter length of stay. However, we chose to focus on this subgroup given they are at highest risk for the development of posttraumatic VTE. Although few of the patients diagnosed with PE in this study were known to have concurrent DVT, routine screening LE duplex was not performed in patients who had a diagnosis of PE. Recent data does show that some pulmonary clots in trauma patients may represent in situ thromboses, rather than embolic events, and that these have different pathophysiology than PE (21). To account for this, subsegmental pulmonary clots present on admission imaging were not included in our definition of VTE. To control for potential confounders in our multivariable regression models, we adjusted for multiple factors that influence platelet aggregation such as platelet count, injury severity, and shock that may also be associated with the development of VTE. Despite this, it is possible there is unmeasured confounding in our analysis. Furthermore, while we controlled for time to initiation of chemoprophylaxis in our models, we were not able to account for missed doses or variations in dosing and frequency. Finally, the associations of platelet aggregation with development of VTE in our study appear uniform across the surface receptor pathways tested, supporting a global or upstream mechanism mediating the relationship of post-injury platelet biology with the development of posttraumatic VTE. We hypothesize this association is due to an exhausted platelet phenotype, in which impaired *ex-vivo* aggregation is reflective of excessive *in-vivo* platelet activation and aggregation triggered by prothrombotic mediators released in the setting of extensive tissue injury (13, 38). In this setting, platelets that have been excessively activated and stimulated to degranulate *in vivo* may no longer capable of normal aggregation *ex-vivo* (14). However, this hypothesis of functional exhaustion requires further investigation including analysis of surface receptor markers of platelet activation and of platelet degranulation to provide more supporting evidence. Additionally, further study characterizing the downstream effects of altered platelet function on fibrinolysis are also warranted as this may be an important pathway through which platelets promote hypercoagulability and increased risk of posttraumatic VTE, given platelet activation can

include the degranulation of platelet derived plasminogen activator inhibitor-1 and alpha antiplasmin-2.

In conclusion, this study demonstrated strong independent associations between impairments in platelet aggregation and clot lysis at 24 hours after presentation with an increased risk of development of posttraumatic VTE. We hypothesize that it is possible this reflects a functionally exhausted platelet phenotype, and believe that future studies should examine biologic markers of platelet activation and degranulation in parallel with *ex-vivo* aggregation assays to further characterize the platelet biology associated with the development of posttraumatic VTE. This is an important line of investigation, as these data could be used to further test whether a targeted approach to adding platelet-based chemoprophylaxis to standard-of-care heparin-based chemoprophylaxis is safe and beneficial in patients at high risk of development of posttraumatic VTE.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements:

Disclosure information:

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Media Summary:

Altered platelet aggregation is common following injury, but the relationship with venous thromboembolism is unknown. We identify that the decrease in platelet aggregation from presentation to 24 hours post-resuscitation is independently associated with development of posttraumatic venous thromboembolism.

Figure 1.
Coefficient Plot of Association of Platelet Aggregation on Presentation, at 24 hours, and Change from 0 to 24 hours with Development of Posttraumatic Venous Thromboembolism (VTE). X axis is the odds ratio per 10 aggregation units, adjusted for length of stay, initiation of VTE chemoprophylaxis within 72 hours, platelet count, injury severity score, shock index, and whether patient was transfused in initial 24 hours.

Association of Viscoelastic Clot Strength and Lysis at Presentation and at 24 hours with Development of Venous Thromboembolism
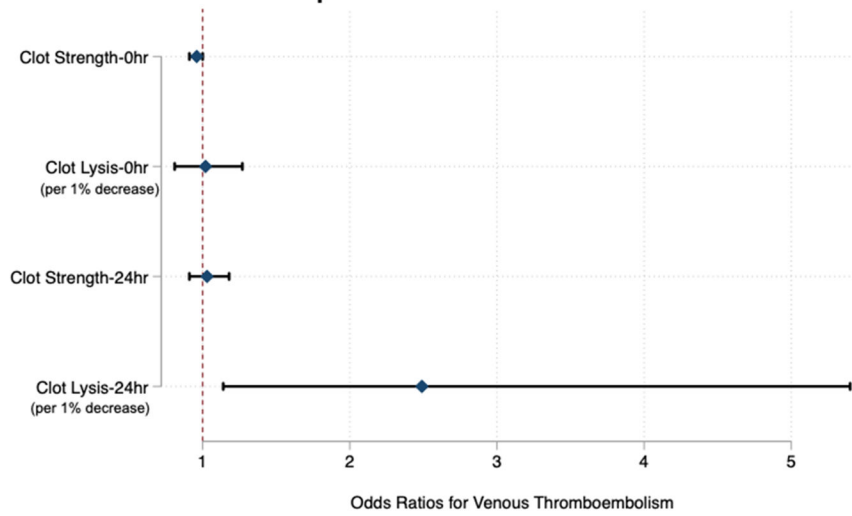
Figure 2.

Coefficient Plot of Association of Viscoelastic Clot Strength and Lysis at presentation (0 hours) and at 24 hours with Development of Posttraumatic VTE. X axis is the odds ratio per 10 aggregation units, adjusted for length of stay, initiation of chemoprophylaxis with in 72 hours, injury severity score, shock index, and whether patients were transfused in initial 24 hours.

Table 1.

Baseline Characteristics of Patients with and without Development of Venous Thromboembolism

	No VTE (n=115)	Yes VTE (n=18)	Total (n=133)	Number with missing data	P-value
Age (years)	37 (26-52)	44 (29-53)	38 (26-52)	0	0.41
Sex				0	0.27
Female	12.2%	22.2%	13.5%		
Male	88%	78%	87%		
BMI (kg/m^2)	26.8 (23.0-30.2)	27.6 (24.6-31.4)	26.8 (23.3-30.6)	3	0.33
Mechanism				0	0.27
Penetrating	33%	17%	31%		
Blunt	67%	83%	69%		
Injury Severity Score	22 (14-30)	29 (27-34)	25 (14-33)	0	<0.01
AIS Head	3 (0-4)	3 (2-5)	3 (0-4)	2	0.30
GCS	13 (7-15)	14 (9-15)	13 (7-15)	1	0.61
Heart Rate	96 (24)	100 (35)	96 (26)	1	0.57
Systolic Blood Pressure (mmhg)	132 (32)	128 (39)	131 (33)	1	0.70
Shock Index	0.9 (0.3)	0.9 (0.4)	0.9 (0.3)	1	0.48
pH	7.3 (7.3-7.4)	7.3 (7.2-7.3)	7.3 (7.3-7.4)	23	0.08
Base Excess (meq/ml)	−2.5 (−7.0-0.2)	−5.7 (−7.6--2.8)	−2.6 (−7.4--0.1)	22	0.11
INR	1.1 (1.0-1.2)	1.1 (1.1-1.2)	1.1 (1.0-1.2)	5	0.48
pTT (seconds)	28 (25-30)	30 (26-34)	28 (25-31)	5	0.07
Platelet count (presentation), x10^9/L	263 (209-322)	260 (207-299)	263 (209-310)	0	0.59
Platelet count (24 hours), x10^9/L	164 (127-201)	126 (106-153)	157 (124-193)	0	0.01
Proportion with initiation of chemoprophylaxis				0	
24 hours	15%	0%	13%		0.08
48 hours	39%	17%	36%		0.07
72 hours	64%	39%	61%		0.04
96 hours	78%	44%	73%		<0.01
120 hours	84%	78%	83%		0.51
Length of stay (to 28 days)	16 (9-24)	28 (21-28)	17 (10-25)	0	<0.01
ICU length of stay (to 28 days)	5 (3-11)	14 (7-23)	7 (3-14)	0	<0.01
Ventilator-free days (to 28 days)	25 (20-27)	18 (5-24)	25 (15-27)	6	<0.01
Multiple Rib Fractures (3+)	21%	33%	22%	4	0.24
Hemo- or pneumothorax	35%	50%	37%	4	0.29
Pelvic fracture	17%	28%	19%	4	0.33
Multiple long bone fractures (2+)	12%	39%	16%	4	<0.01
Spinal Cord Injury	15%	17%	16%	4	1.00
Traumatic Brain Injury	48%	56%	49%	0	0.62
Transfused any product (24h)	62%	83%	65%	0	0.11
pRBCs transfused (24h)	2.0 (0.0-4.0)	3.0 (2.0-7.0)	2.0 (0.0-5.0)	0	0.04
Transfused platelets (24h)	22%	29%	23%	0	0.54
Platelet units transfused (24h)	0.0 (0.0-0.0)	0.0 (0.0-1.0)	0.0 (0.0-0.0)	0	0.48

	No VTE (n=115)	Yes VTE (n=18)	Total (n=133)	Number with missing data	P-value
Mortality at discharge	10%	22%	12%	0	0.23

VTE- venous thromboembolism; AIS- abbreviated injury score; GCS- Glasgow coma scale; HR- heart rate; SBP- systolic blood pressure; shock index=HR/SBP; INR- international normalized ratio; pTT- partial thromboplastin time; chemoprophylaxis defined as initiation of low-molecular weight or unfractionated heparin; pRBCs=packed red blood cells; ICU-intensive care unit

Table 2.

Bivariate Comparisons of Platelet Aggregation and Viscoelastic Measures of Patients with and without Development of Venous Thromboembolism

	No VTE (n=115)	Yes VTE (n=18)	Total (n=133)	P-value
ADP AUC, presentation	55 (37-70)	53 (47-72)	54 (38-71)	0.31
Collagen AUC, presentation	48 (33-63)	50 (38-68)	49 (34-63)	0.43
Thrombin AUC, presentation	97 (81-115)	87 (80-144)	96 (80-117)	0.70
ADP AUC, 24h	28 (20-40)	19 (12-25)	26 (19-39)	<0.01
Collagen AUC, 24h	32 (22-48)	17 (15-25)	29 (18-46)	<0.01
Thrombin AUC, 24h	66 (49-81)	46 (37-75)	66 (46-80)	0.06
Delta ADP AUC, presentation to 24h	19 (7-36)	29 (17-58)	21 (9-40)	0.03
Delta Collagen AUC, presentation to 24h	13 (−1-25)	30 (14-53)	16 (0-26)	<0.01
Delta Thrombin AUC, presentation to 24h	31 (11-49)	43 (34-90)	34 (15-51)	<0.01
Clot strength (CK-MA), presentation	65.2 (61.8-68.4)	64.1 (60.4-67.9)	65.2 (61.8-68.3)	0.52
Clot lysis (CK-LY30,%), presentation	0.5 (0.1-2.0)	0.2 (0.0-1.6)	0.5 (0.0-1.9)	0.31
Clot strength (CK-MA), 24h	64.5 (61.2-67.1)	65.2 (61.4-68.4)	64.5 (61.2-67.2)	0.78
Clot lysis (CK-LY30,%), 24h	0.9 (0.3-1.8)	0.2 (0.0-1.0)	0.8 (0.2-1.8)	0.02

ADP AUC- adenosine diphosphate stimulated platelet aggregation, Collagen-collagen stimulated platelet aggregation, Thrombin AUC- thrombin stimulated platelet aggregation. Delta- Change platelet aggregation from presentation to 24 h.

Table 3.

Multivariable Logistic Regression Results for Association of Platelet Aggregation and TEG values at 24h with Development of VTE

ADP Stimulated *PA*, 24hr	Odds Ratio	CI-Low	CI-High	P-Value
ADP *PA*, (per 10 AUC units)	0.86	0.55	1.34	0.51
Length of Stay (days)	1.09	1.02	1.17	0.01
Initiation of chemoprophylaxis within 72 hours	0.27	0.08	0.86	0.03
Platelet count (24 hours)	0.99	0.98	1.01	0.35
Injury Severity Score	1.02	0.97	1.07	0.43
Shock Index	1.46	0.27	7.96	0.66
Transfused in initial 24 hours	1.21	0.23	6.44	0.82
Area under ROC curve=0.84				
Collagen Stimulated *PA*, 24h	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Collagen *PA*, (per 10 AUC units)	0.63	0.40	1.00	0.05
Length of Stay (days)	1.10	1.02	1.17	0.01
Initiation of chemoprophylaxis within 72 hours	0.40	0.12	1.30	0.13
Platelet count (24 hours)	1.00	0.98	1.01	0.86
Injury Severity Score	1.02	0.97	1.08	0.40
Shock Index	0.55	0.09	3.51	0.52
Transfused in initial 24 hours	1.55	0.26	9.37	0.63
Area under ROC curve=0.84				
Thrombin stimulated *PA*, 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Thrombin *PA*, (per 10 AUC units)	0.91	0.68	1.23	0.55
Length of Stay (days)	1.11	1.03	1.19	0.01
Initiation of chemoprophylaxis within 72 hours	0.25	0.08	0.84	0.02
Platelet count (24 hours)	0.99	0.98	1.01	0.50
Injury Severity Score	1.03	0.98	1.09	0.27
Shock Index	1.59	0.29	8.75	0.59
Transfused in initial 24 hours	0.99	0.18	5.43	0.99
Area under ROC curve=0.84				
Delta-ADP presentation to 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Delta ADP, (per 10u)	1.31	1.03	1.65	0.03
Length of Stay (days)	1.10	1.02	1.18	0.01
Initiation of chemoprophylaxis within 72 hours	0.45	0.13	1.54	0.20
Platelet count (24 hours)	0.99	0.97	1.00	0.15
Injury Severity Score	1.04	0.98	1.09	0.19
Shock Index	0.80	0.11	5.88	0.83
Transfused in initial 24 hours	0.89	0.16	5.10	0.90
Area under ROC curve=0.86				
Delta-Collagen presentation to 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Delta Collagen, (per 10u)	1.36	1.07	1.73	0.01
Length of Stay (days)	1.10	1.02	1.18	0.01

ADP Stimulated *PA*, 24hr	Odds Ratio	CI-Low	CI-High	P-Value
Initiation of chemoprophalaxis within 72 hours	0.35	0.10	1.20	0.10
Platelet count (24 hours)	0.99	0.98	1.01	0.46
Injury Severity Score	1.04	0.98	1.10	0.21
Shock Index	0.82	0.10	6.50	0.85
Transfused in initial 24 hours	1.16	0.20	6.72	0.87
Area under ROC curve=0.85				
Delta-Thrombin, presentation to 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Delta- Thrombin, (per 10u)	1.41	1.11	1.78	<0.01
Length of Stay (days)	1.13	1.05	1.22	<0.01
Initiation of chemoprophalaxis within 72 hours	0.18	0.04	0.72	0.02
Platelet count (24 hours)	0.99	0.98	1.01	0.23
Injury Severity Score	1.04	0.97	1.11	0.23
Shock Index	1.21	0.18	8.11	0.84
Transfused in initial 24 hours	0.55	0.08	3.67	0.53
Area under ROC curve=0.88				
Clot Strength at 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
Clot strength, (mm)	1.03	0.91	1.18	0.62
Length of Stay (days)	1.17	1.05	1.30	<0.01
Initiation of chemoprophalaxis within 72 hours	0.31	0.09	1.07	0.06
Injury Severity Score	1.03	0.97	1.09	0.31
Shock Index	1.40	0.24	8.25	0.71
Transfused in initial 24 hours	2.97	0.49	17.88	0.23
Area under ROC curve=0.84				
Clot Lysis at 24 hours	**Odds Ratio**	**CI-Low**	**CI-High**	**P-Value**
CK-LY30, (%)	2.49	1.14	5.49	0.02
Length of Stay (days)	1.20	1.07	1.34	<0.01
Initiation of chemoprophalaxis within 72 hours	0.33	0.09	1.21	0.09
Injury Severity Score	1.03	0.97	1.09	0.33
Shock Index	0.92	0.13	6.64	0.93
Transfused in initial 24 hours	3.00	0.46	19.75	0.25
Area under ROC curve=0.88				

PA-Platelet aggregation in aggregation units (AUC), ADP- adenosine diphosphate, delta-change in platelet aggregation from 0 to 24 hours, CK-LY30-percent clot lysis at 30 minutes, shock index-heart rate/systolic blood pressure on arrival