Concordance between Activated Partial Thromboplastin Time and Antifactor Xa Assay for Monitoring Unfractionated Heparin in Hospitalized Hyperbilirubinemic Patients

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ABSTRACT

BACKGROUND: Activated partial thromboplastin time (aPTT) and antifactor Xa (anti-Xa) monitoring methods for unfractionated heparin (UFH) often disagree. The extent of discordance for those with elevated bilirubin remains unclear. Our objective was to evaluate concordance between activated aPTT and anti-Xa methods for hyperbilirubinemic patients on UFH.

METHODS: This was a retrospective cohort study of 26 patients hospitalized at Rhode Island Hospital between August 2014 and September 2014. Patients had at least one bilirubin measurement >5 mg/dL. After categorizing lab values, percent agreement and kappa were used to examine concordance between aPTT and anti-Xa.

RESULTS: Overall percent agreement between aPTT and anti-Xa was 50%. A nontherapeutic aPTT and therapeutic anti-Xa accounted for 98% of all disagreement. Specifically, 76.7% of disagreement was due to a subtherapeutic aPTT and a therapeutic anti-Xa. Unweighted kappa was 0.141 (95%CI: 0.048–0.235).

CONCLUSION: Concordance between aPTT and anti-Xa values was poor in hyperbilirubinemic patients.

KEYWORDS: Activated partial thromboplastin time, antifactor Xa heparin assay, unfractionated heparin, hyperbilirubinemia, Rhode Island

INTRODUCTION

Intravenous unfractionated heparin (UFH) is an anticoagulant frequently used to treat thromboembolic diseases.[1] Despite its beneficial anticoagulant effect, UFH is recognized as a high-risk medication by the Institute of Safe Medication Practices due to associated medication errors and adverse drug events (ADEs), including serious or fatal bleeding episodes.[2, 3] Laboratory monitoring of UFH guides dosing to achieve therapeutic levels and avoid ADEs due to over- or under-dosing. Monitoring of UFH was traditionally done using activated partial thromboplastin time (aPTT) since it was widely available and inexpensive.[4] More recently, institutions have transitioned to using antifactor Xa levels (anti-Xa).[4]

Data suggest that the aPTT and anti-Xa tests are not equivalent measures due to distinct limitations of each.[5-8] There is no single absolute numerical reference range for aPTT because it can vary between institutions,[1] which is a significant limitation that interferes with accurate assessment of a patient's intrinsic heparin activity across care settings.[1] Variation in aPTT occurs due to differences in collection, sample preparation, reagents, and instruments used between institutions.[9] Studies have shown that aPTT is also a more variable assay when compared to anti-Xa due to biologic variables.[9] For example, several conditions can affect the aPTT assay, such as factor deficiency, renal disease, and liver disease.[4] Even though anti-Xa is less affected by these laboratory and biological factors, it still can be affected by others like hyperbilirubinemia and hypertriglyceridemia.[4, 9, 10]

At Rhode Island Hospital, physicians identified hyperbilirubinemia as a particular concern due to its potential to interfere with valid assessment of intrinsic heparin activity. The threshold for hyperbilirubinemia to interfere is dependent on the lab reagent used, but many institutions use an upper total bilirubin level of 20 mg/dL.[10] Nonetheless, clinicians at our institution reported discordant values of anti-Xa and aPTT for patients with elevated bilirubin levels of just 5 mg/dL. In some cases, clinicians believed that the interference with assessment of heparin activity via anti-Xa had been obscured by hyperbilirubinemia, which may motivate improper dosing, increasing the risk for significant bleeding and thromboembolic events. Due to a deficit in published literature examining the concordance between anti-Xa and aPTT values in patients with hyperbilirubinemia, we aimed to examine the agreement between anti-Xa and aPTT in these patients.

METHODS

Setting and Participants

This was an exploratory single-center retrospective observational cohort study conducted at Rhode Island Hospital in Providence, RI. Data was collected for all patients on UFH intravenous infusion protocol from August 8, 2014 to September 8, 2014, a time period during which both anti-Xa and aPTT methods were available to clinicians for monitoring as the hospital transitioned to exclusively using anti-Xa for UFH protocol. We collected baseline data from the medical record admission history, including age, sex, height, actual body weight, and other relevant characteristics. Patients

included had ≥1 order for continuous intravenous UFH that was administered during their inpatient stay, a total bilirubin level >5 mg/dL, and both an aPTT and anti-Xa measurement at least once during their stay. At our institution, the normal range for bilirubin is 0.2-1.3 mg/dl. We therefore chose bilirubin >5mg/dL as the threshold for hyperbilirubinemia as it is approximately three times the upper limit of normal. Patients were excluded from the analysis if they did not have both aPTT and anti-Xa measured at least once on the same day. Patients were also excluded if their treating physician did not follow the hospital-approved UFH dosing nomogram. The study was approved by the Lifespan-Rhode Island Hospital Institutional Review Board.

Measures

Since we did not expect the aPTT and anti-Xa tests to be assessed using the same plasma sample (at exactly the same measurement time), we calculated the mean daily value of each test by patient. The mean daily lab values for aPTT and anti-Xa were then recoded from continuous variables to two dichotomous variables indicating whether the lab value for each test was in the therapeutic range or out of the therapeutic range for a given day. The first dichotomous variable was equal to 1 if the aPTT value was >=70 and <=100 seconds, 0 if otherwise.[11] The second dichotomous variable was equal to 1 if anti-Xa was >=0.3 and <=0.7 units/mL, 0 otherwise.[11] For a secondary analysis, values of the dichotomous variables were recoded to multilevel categorical variables where 0 indicated subtherapeutic lab values (<70 seconds for aPTT and <0.3 units/mL for anti-Xa), 1 indicated therapeutic values (as above), and 2 indicated supratherapeutic values (>100 seconds for aPTT and >0.7 units/mL for anti-Xa).

Statistical Analysis

To describe the relationship between mean daily anti-Xa and aPTT levels, we plotted the anti-Xa values versus the aPTT values with a means-centered 95% confidence ellipse. Assuming a bivariate normal distribution, the ellipse shows where 95% of the data in a scatter plot should lie on average. Confidence ellipses also serve as visual indicators of correlations, where more circular ellipses indicate that two variables are uncorrelated and more diagonal ellipses indicates stronger correlations.[12,13] To complement the plot and describe the linear association between anti-Xa and aPTT, we additionally calculated the coefficient of determination (R^2), thus allowing for interpretation of the correlation between measures independent of the scale of the plot.[12]

After creating categorical variables, we calculated the observed agreement between the aPTT and anti-Xa tests. Cohen's unweighted kappa (κ) was also used to assess agreement between the dichotomous variables and both unweighted and weighted κ were used to assess agreement between the ordinal variables.[5,6,8,14,15] Kappa is a more robust measure than percent agreement because it accounts for agreement due to chance. Kappa was interpreted using previously established benchmarks.[16] We calculated 95% confidence intervals for κ using an analytic method for the dichotomous variables[17] and 1,000 bootstrap replications for ordinal variables.[18,19] Statistical significance was based on a two-sided type 1 error of 0.05.

RESULTS

We identified 86 individuals with 1,236 lab measurements as our initial study population. Their mean (SD), median, and range of daily total bilirubin level during the inpatient stay was 5.4 (8.3) mg/dL, 1.2, and 0.2 to 34.2, respectively. Of 483 total days of patient stay for all patients in the cohort, a mean daily aPTT and anti-Xa levels were both available on 108 days (22.4%). There were 42 patients (49%) with complete information for anti-Xa, 53 patients (62%) with complete information for aPTT, and 26 patients (30%) with complete information for both. At baseline, patients were older, predominantly male, and typically admitted for acute coronary syndrome (**Table 1**). During the inpatient stay, the mean (SD), median, and range of daily total bilirubin level for these 26 patients was 5.2 (9.0) mg/dL, 0.8, and 0.2 to 34.2, respectively; these values stand in contrast to those at admission (baseline), which were 1.1 (1.3) mg/dL, 0.5, and 0.2 to 5.3.

Figure 1 depicts the distribution of mean daily anti-Xa versus aPTT levels. The R^2 was 0.32, indicating modest correlation. **Table 2** shows that when the aPTT and anti-Xa levels disagree, the mismatch almost always occurs (98% of disagreement) because the anti-Xa level is therapeutic and the aPTT level is not. We observe the same result in **Table 3**, which shows that disagreement usually occurs (77% of

Figure 1. Mean Daily anti-Xa Versus Mean Daily aPTT Levels with 95% Confidence Ellipse

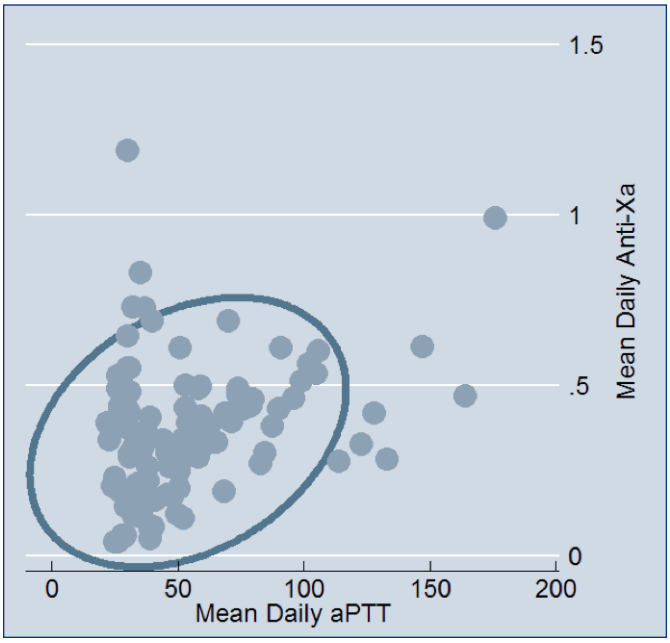

Table 1. Demographic and Clinical Characteristics of Patients at Admission

CHARACTERISTIC	(N=26)
Age at Admission – yr [mean (SD)]	**66.5 (15.5)**
Median	64.5
Range	23 to 88
Sex – no. (%)	
Male	17 (65.4)
Female	9 (34.6)
Weight – kg [mean (SD)]	**84 (23.4)**
Median	82.1
Range	43.6 to 137
International Normalized Ratio – mean (SD)	**1.2 (0.2)**
Median	1.1
Range	1 to 1.6
Total Bilirubin – mg/dL [mean (SD)]	**1.1 (1.3)**
Median	0.5
Range	0.2 to 5.3
Unfractionated Heparin Indication – no. (%)	
Acute Coronary Syndrome	12 (46.2)
Pulmonary Embolism and/or Deep Vein Thrombosis	5 (19.2)
Atrial Fibrillation or Flutter	2 (7.7)
Atrial Thrombus	1 (3.9)
Congestive Heart Failure	1 (3.9)
Extracorporeal Membrane Oxygenation	1 (3.9)
Ischemic Colitis	1 (3.9)
Ischemic Stroke	1 (3.9)
Portal Vein Thrombosis	1 (3.9)
Necrotic Bowel status post Short Bowel Resection	1 (3.9)
Hepatic Impairment – no. (%)	
Yes	1 (3.9)
No	25 (96.1)
Factor Deficiency – no. (%)	
Yes	0 (0)
No	26 (100)
Vitamin K Deficiency – no. (%)	
Yes	0 (0)
No	26 (100)
Factor Xa Use – no. (%)	
Yes	2 (7.7)
No	24 (92.3)
Vitamin K Antagonist Use – no. (%)	
Yes	2 (7.7)
No	24 (92.3)
Direct Thrombin Inhibitor Use – no. (%)	
Yes	0 (0)
No	26 (100)
Antiphospholipid Syndrome – no. (%)	
Yes	1 (3.9)
No	25 (96.1)
Triglyceride Value >360 mg/dL – no. (%)	
Yes	1 (3.9)
No	25 (96.1)

Table 2. Percent Agreement of Mean Daily aPTT and anti-Xa Therapeutic Classifications, Dichotomous Indicator

		aPTT		
		Non-therapeutic	Therapeutic	Total
Anti-Xa	Non-therapeutic	41 (37.9%)	1 (0.9%)	42
	Therapeutic	53 (49.1%)	13 (12%)	66
	Total	94	14	108

Table 3. Percent Agreement of Mean Daily aPTT and anti-Xa Therapeutic Classifications, Multilevel Indicator

		aPTT			
		Sub-therapeutic	Therapeutic	Supra-therapeutic	Total
Anti-Xa	Sub-therapeutic	34 (31.5%)	1 (0.9%)	2 (1.9%)	37
	Therapeutic	46 (42.6%)	13 (12%)	7 (6.5%)	66
	Supra-therapeutic	4 (3.7%)	0 (0%)	1 (0.9%)	5
	Total	84	14	10	108

disagreement) when the aPTT level is subtherapeutic and the anti-Xa level is in the therapeutic range.

As shown in **Table 4,** the observed agreement between aPTT and anti-Xa levels was 50.00%. If each lab test had produced a value randomly, we would expect the aPTT and anti-Xa levels to agree 41.77% of the time. The unweighted kappa value was 0.141 (95%CI: 0.048–0.235), indicating poor agreement. Using the ordinal categorical indicator variables, the observed agreement between sub-, supra-, and therapeutic levels was 44.4%. We would expect the aPTT and anti-Xa levels to agree 35% of the time by chance. The unweighted kappa value associated with this was 0.145 (bias-corrected 95%CI: 0.070-0.173), indicating poor agreement.

The linear weighted kappa for the ordinal categorical indicator variables was 0.15 (bias-corrected 95%CI: 0.079–0.161), indicating poor agreement (**Table 5**). Similarly, the quadratic weighted kappa for the ordinal categorical indicator variables was 0.15 (bias-corrected 95%CI: 0.096-0.253).

DISCUSSION

We report the results of an exploratory retrospective cohort study of aPTT and anti-Xa UFH laboratory monitoring methods for anticoagulation in hyperbilirubinemic patients at a large academic medical center. The monitoring methods agreed approximately half of the time with a nontherapeutic aPTT and therapeutic anti-Xa accounting for nearly all of the disagreement

Table 4. Unweighted Kappa for Agreement Between aPTT and anti-Xa Measurements

A. Dichotomous Indicator Variable				
Observed Agreement	Expected Agreement	Kappa	95%CI	*P*-value
50.00%	41.77%	0.141	0.048-0.235	<0.01
B. Multilevel Categorical Indicator Variable				
Observed Agreement	Expected Agreement	Kappa	95%CI	*P*-value
44.44%	35.00%	0.145	0.070-0.173	<0.01

Table 5. Weighted Kappa for Agreement Between aPTT and anti-Xa Measurements

A. Linear Weights				
Observed Agreement	Expected Agreement	Kappa	95%CI*	*P*-value
69.44%	64.11%	0.149	0.049-0.269	<0.01
B. Quadratic Weights				
Observed Agreement	Expected Agreement	Kappa	95%CI*	*P*-value
81.94%	78.67%	0.153	0.009-0.313	<0.01

**Confidence intervals bias-corrected and calculated using 1000 bootstrap replications.*

and 76.7% of disagreement due to a subtherapeutic aPTT and therapeutic anti-Xa. To our knowledge, this is one of the first studies examining the discordance between aPTT and anti-Xa among hyperbilirubinemic patients.

Even though guidelines recommend calibrating aPTT levels to anti-Xa levels, this practice has been questioned.[5, 11, 20-22] Previous studies have shown that anti-Xa and aPTT assays poorly correlate and that about 50% of measurements are discordant, but did not examine the effect of hyperbilirubinemia on coagulation assays.[5-8] Those findings and our own results support the hypothesis that anti-Xa and aPTT often disagree, with disagreement among those with hyperbilirubinemia similar to those without. Our findings align with the laboratory study of Lippi et al., who concluded that bilirubin up to 20mg/dl does not significantly affect coagulation testing.[23] Kostousov et al. conducted an in vitro study of extracorporeal membrane oxygenation (ECMO) patients with hyperbilirubinemia and found that elevated bilirubin increased the aPTT level and decreased the anti-Xa level.[24]

A limitation of our study was that aPTT and anti-Xa levels were not measured at the same exact time, but measuring the two levels at the exact same time could produce different results. Our study was also limited by its single-center setting, small sample size, and the unclear cause of hyperbilirubinemia for many patients. A major strength of our study was its use of recent data and the application of statistical methodologies that have not been previously used to examine the discordance between aPTT and anti-Xa.

In conclusion, our study supports the evidence that aPTT and anti-Xa monitoring methods often disagree, but offers important new information to suggest that disagreement in patients with elevated total bilirubin is not dramatically different from that documented in the overall hospitalized population. Clinicians should continue to use anti-Xa to assess coagulation status since it is less affected by laboratory and biological factors than aPTT. Alternatively, some studies have suggested the use of endogenous thrombin potential (ETP) as a more direct measure of heparin activity,[7, 25] but this assay is not widely available and requires further investigation to assess its clinical utility.

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