Preanalytic Factors Associated With Hemolysis in Emergency Department Blood Samples

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• *Context.*—Hemolysis of emergency department blood samples is a common occurrence and has a negative impact on health care delivery.

***Objectives.*—To determine the effect of preanalytic factors (straight stick, intravenous [IV] line, needle gauge, location of blood draw, syringe versus vacuum tube use, tourniquet time) on hemolysis in emergency department blood samples.**

***Design.*—A single 65 000-visit emergency department's electronic health record was queried for emergency department potassium results and blood draw technique for all samples obtained in calendar year 2014, resulting in 54 531 potassium results. Hemolyzed potassium was measured by hemolysis index. Comparisons of hemolysis by sampling technique were conducted by χ^2 tests.**

***Results.*—Overall hemolysis was 10.0% (5439 of 54 531). Hemolysis among samples obtained from straight stick was significantly less than among those obtained with IV line (5.4% [33 of 615] versus 10.2% [4821 of 47 266], *P* < .001). For IV-placed blood draws, antecubital location had a statistically significant lower overall hemolysis compared with other locations: 7.4% (2117 of 28 786) versus 14.6% (2622 of 17 960) (*P* < .001). For blood drawn with a syringe compared with vacuum, hemolysis was 13.0% (92 of 705) and 11.0% (1820 of 16 590), respectively (*P* = .09, not significant). For large-gauge IV blood draws versus smaller-gauge IV lines, a lower hemolysis was also observed (9.3% [3882 of 41 571] versus 16.7% [939 of 5633]) (*P* < .001). For IV-drawn blood with tourniquet time less than 60 seconds, hemolysis was 10.3% (1362 of 13 162) versus 13.9% for more than 60 seconds (532 of 3832), *P* < .001.**

***Conclusions.*—This study confirmed previous findings that straight stick and antecubital location are significantly associated with reduced hemolysis and indicated that shorter tourniquet time and larger gauge for IV draws were significantly associated with lower hemolysis.**

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In the United States, approximately 130 million patients visit emergency departments (EDs) each year; during these visits, 54 million blood samples are collected, and of these, 30 million undergo electrolyte analysis (23% of all visits).[1] Emergency department blood samples are reported to have a particularly high incidence of hemolysis (6%–30%).[2–4] Emergency department hemolysis tends to be much higher than the 2% benchmark established by the American Society for Clinical Pathology, which is especially problematic given the high-volume and overcrowded nature of EDs.[5] Hemolysis can significantly affect the reliability of test results, often requiring repeat testing, which can increase patient discomfort, ED costs, and throughput time. In hemolyzed specimens, chemistry test results might not reflect the patient's clinical condition, either because of red blood cell contents spilling into the serum or plasma (actually changing the plasma concentration of the analytes) or because of red blood cell contents interfering with the testing methodology (eg, hemoglobin can discolor a solution and impact spectrophotometric absorbance measurements at certain wavelengths). Commonly, the analytes most affected by this first interference mechanism are increased concentrations of potassium, lactate dehydrogenase, and aspartate aminotransferase. Many other analytes may be increased or decreased based on the testing methodology; most laboratories assess a specimen's degree of hemolysis and, based on the laboratory director's instructions, have protocols to address resulting hemolyzed specimens. Even if they are generated at a fairly low rate (~2%), hemolyzed samples can substantially impact the delivery and quality of health care.[1,6,7]

The reasons for high hemolysis are likely multifactorial, with the conditions leading to hemolysis typically arising during the preanalytic phase of the testing process.[6] Our study focused on altering preanalytic, in vitro, mechanical causes of hemolysis. There are many pathologic causes of in vivo hemolysis arising from genetic conditions (eg, red blood cell membrane defects, hemoglobinopathies) or other

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factors (drug and/or immune mediated or mechanical, such as microangiopathic hemolytic anemia). However, despite the variety of these hemolytic anemias, their incidence is relatively low in our ED population, which we conclude based on the successful (nonhemolyzed) redraws we observed in almost all of our cases. We did not seek to exclude these types of patients from our analysis; therefore, some baseline level of hemolysis is perpetually expected.

Heyer et al[2] published a meta-analysis and systemic review on best practices around laboratory sample hemolysis. They sought to examine 7 preanalytic practices that could reduce the hemolysis: (1) sample collection by a phlebotomist versus ED medical staff, (2) collection via straight stick versus intravenous (IV) line, (3) use of the antecubital versus other location, (4) syringe versus vacuum draw (IV only), (5) 21-gauge IV line or smaller versus larger sizes (IV draws only), (6) tourniquet time ($\leq$1 minute versus longer), and (7) low partial vacuum tubes versus other tubes. Only 2 of these practices had high-level evidence satisfactory enough to meet their support criteria: use of a straight stick to obtain the sample and, if collecting blood via IV access, use of the antecubital location. The remaining 5 practices could not be supported by the Heyer et al[2] meta-analysis: 2 because no studies of the practice were available (phlebotomists versus ED medical staff and tourniquet time) and 3 because of insufficient data (syringe versus vacuum IV draw, use of 21-gauge needles or smaller versus other sizes, and use of low partial vacuum tubes versus other tubes). Their findings are supported by a report published by the Emergency Nurses Association, outlining best practices and factors associated with increased ED blood sample hemolysis.[8] The objective of our study was to further determine the effect of a set of preanalytic factors from the Heyer et al[2] study (use of straight stick, needle gauge, location of blood draw, syringe versus vacuum tube use, tourniquet time) on hemolysis. Our primary hypothesis was that these preanalytic factors are significantly associated with different incidences of hemolysis.

DESIGN

This was a secondary analysis of a larger performance improvement project that was targeted toward reducing hemolysis in ED specimens and conducted in an urban tertiary care center with an affiliated ED residency program and annual census of approximately 65 000 visits. This study was funded by a Cooperative Agreement with the Centers for Disease Control and Prevention (project number 5U47OE000053-02) and aided in the study design. Because this initiative was part of a quality improvement project, our institutional review board deemed it exempt from consent procedures.

We routinely collect a "rainbow draw" in the ED, that is, multiple tubes of various types to allow flexibility for the ordering provider when placing laboratory orders. Most stat chemistry tests are performed on a Becton Dickinson (BD, Franklin Lakes, New Jersey) 6-mL lithium heparin tube, without gel. Syringes are also from BD. The same tubes were used throughout this phase of our work. All ED-obtained samples in which potassium analysis was completed during calendar year 2014 were included. The hospital electronic health record Epic (Verona, Wisconsin) was queried for all ED patients with laboratory potassium results. The method of sample collection (use of straight stick, needle gauge, location of blood draw, syringe versus vacuum tube use, and tourniquet time) was determined from data in the documentation flow sheet in the laboratory collection module in the ED nursing narrator of Epic. Currently, there is no standard practice of when to use ultrasound when guiding IV access placement, but it is increasingly being used for blood draws from patients in whom IV access is difficult to establish in an ED setting. At the time of the study, documentation of each of the preanalytic factors was a standard part of ED care, although each field was not mandatory and therefore not always filled out, resulting in missing fields. Our ED does not use phlebotomists, and at the time of this study in our ED, the use of small tubes was not feasible (this potential process improvement was being considered as a performance improvement project at a later date). Moreover, no studies that examined hemolysis under the Heyer et al[2] categories 2, 3, 4, and 5 based solely on straight-stick use have been published. Therefore, we also examined the Heyer et al[2] practices 2 through 5 in the setting of straight-stick draws alone. All specimens were processed using a Roche Cobas 8000 (Roche Diagnostics, Indianapolis, Indiana). For all such samples sent to the core laboratory, the hemolysis index (HI), a quantitative measure of the extent of hemolysis, is determined as a quality indicator. The HI on the Roche Cobas 8000 is a quantitative, continuous measurement of the amount of hemoglobin in a plasma or serum sample. The HI correlates linearly with plasma hemoglobin concentration up to a value of at least 300 (data not shown). The HI value is determined by a dual-wavelength spectroscopic absorbance measurement without use of additional reagents on the Cobas system. Other instrument vendors use similar approaches, but in some cases a semiquantitative scale, rather than a continuous one, is used. On our Roche platforms, an HI above 300 is reported as grossly hemolyzed and the sample is rejected. If the HI is more than 80 but less than or equal to 300, the numeric result is reported with a comment that the specimen was hemolyzed and the result should be interpreted with caution. The HI with comment group was not subdivided. Individual laboratories should establish their own hemolysis cutoffs, based on the degree of interference they observe and the medical impact of that interference in particular patient settings. However, we recognize that subdividing our groupings and performing additional analysis could provide further insight regarding different collection techniques or equipment. We may revisit the data in the future for additional analyses.

The incidence of hemolysis was determined by taking the total number of initial specimens reported as hemolyzed and dividing by the total number of initial specimens (hemolyzed or not). Any association of blood collection methodology and incidence of hemolysis (either grossly hemolyzed or hemolyzed with comment) was assessed with χ^2 tests. For pairwise comparisons in categories with more than 2 groups, we used the Bonferroni correction to evaluate statistical significance and account for multiple comparisons. For comparison of results in contingency tables in which expected cell counts were less than 5, we used the Fisher exact test rather than the χ^2 test. We used a 2-sided type I error threshold of 0.05 for all statistical tests. All analyses were conducted in SAS v.9.4 (Cary, North Carolina).

RESULTS

Our electronic health record query identified 54 531 individual ED potassium results during the study period.

Table 1. Summary of Emergency Department Laboratory Blood Sample Hemolysis (N = 54 531)		
Factor	No.	%
Nonhemolyzed	49 092	90.0
Combined (hemolyzed with comment and gross hemolysis)	5439	10.0
Hemolyzed with comment	4380	8.0
Gross hemolysis	1059	1.9

The combined hemolysis (grossly hemolyzed plus hemolyzed with comment) was 10.0% (5439 of 54 531), hemolysis with comment 8.0% (4380 of 54 531), and grossly hemolyzed 1.9% (1059 of 54 531) (Table 1). Combined hemolysis (hemolyzed with comment plus grossly hemolyzed) among samples obtained from straight stick was significantly lower than hemolysis among samples obtained from IV lines (5.4% [33 of 615] versus 10.2% [4821 of 47 266], $P < .001$; Table 2). For samples obtained by straight stick, use of the antecubital location had a statistically significant lower combined hemolysis compared with other peripheral locations: 2.9% (1 of 345) versus 8.8% (19 of 217) ($P = .01$; Table 3). Samples obtained from the antecubital location in patients with IV access lines had a statistically significant lower combined hemolysis than samples obtained from other peripheral locations (7.4% [2117 of 28 786] versus 14.6% [2622 of 17 960], $P < .001$; Table 4). In the descriptive analyses, we included missing levels as a comparison group to evaluate whether hemolysis was different among observations without available data (Tables 3 and 4). For samples obtained through an IV line and using a 6-mL lithium heparin plasma vacuum tube, combined hemolysis was lower than for those associated with syringe-drawn samples, but the difference was not statistically significant (11.0% [820 of 16 590] versus 13.0% [92 of 705], $P = .09$; Table 5). For large-gauge straight-stick blood draws (16–20 gauge) versus smaller-gauge straight-stick draws, a combined higher hemolysis was observed: 12.5% (14 of 112) versus 3.8% (19 of 500) ($P < .001$; Table 6). Samples obtained from large-gauge IV lines (16–20 gauge) had a lower combined hemolysis than those obtained from smaller-gauge IV lines (9.3% [3882 of 41 571] versus 16.7% [939 of 5633], $P < .001$; Table 7). Straight-stick combined hemolysis by both gauge and location showed significant differences ($P < .001$) as follows: antecubital and 16 to 20 gauge, 6.6% (5 of 76); peripheral and 16 to 20 gauge, 21.4% (6 of 28); antecubital and smaller gauges, 1.5%; and peripheral and smaller gauges, 3.7% (7 of 187) (Table 8). There were no grossly hemolyzed samples in this category. Similarly, with use of smaller-gauge needles, use of the antecubital location was also associated with a lower combined hemolysis ($P < .001$; Table 8). For samples obtained from IV lines, combined hemolysis by both gauge and location showed significant differences ($P < .001$) as follows: antecubital and 16 to 20 gauge, 7.2% (1969 of 27 233); peripheral and 16 to 20 gauge, 13.4% (1885 of 14 106); antecubital and smaller gauges, 9.5% (148 of 1551); and peripheral and smaller gauges, 19.4% (737 of 3795) (Table 9). Gross hemolysis was also statistically significantly different between certain categories, including lowest gross hemolysis for antecubital location for both large and small IV draws (Table 9).

For blood drawn from an IV line with recorded tourniquet time less than 60 seconds, the combined hemolysis was 10.3% (1362 of 13 162) versus 13.9% (532 of 3832) for patients with tourniquet time more than 60 seconds ($P < .001$) (Table 10). Hemolysis was not statistically different when ultrasound was used for IV and blood sample collection: 9.9% (5261 of 53 010) with no ultrasound versus 11.7% (178 of 1521) ($P = .02$) with ultrasound (Table 11).

DISCUSSION

The primary findings of this study, conducted in a large single-center population, indicate that multiple factors were significantly associated with hemolysis. We confirmed the 2 primary findings of the Heyer et al[2] meta-analysis indicating that use of straight stick and use of an IV line in the antecubital location to draw blood were significantly associated with reduced hemolysis. In addition, our findings indicate that shorter tourniquet time and larger gauge among IV-line draws were significantly associated with lower hemolysis; these practices were not identified as significant factors in the systematic review of Heyer et al.[2] Finally, there was no association with use of syringe versus vacuum tube for sample collection and hemolysis. These findings could provide important insights into means of reducing hemolysis in the ED setting.

Howanitz and colleagues[9,10] recently published data on hemolysis in hospital laboratory specimens. Hemolysis among the 722 laboratories studied ranged from less than 1% to 36%, consistent with previous publications reporting hemolysis as high as 30% for specimens obtained in EDs.[2,4] To compare our findings with similar studies is challenging. Although Soderberg et al[4] found much higher rates of hemolysis in EDs (30%) than in our study, this was in the setting of a non-US health care model, which may have been linked to other variables affecting these rates. Rates were also found to be lower in EDs staffed by primary health care physicians as opposed to emergency physicians, suggesting physician-linked preanalytical factors may have played a role.[4] Soderberg and colleagues[4] postulated that increased use of IV draws may have caused the higher hemolysis incidence in patients triaged to be seen by emergency medicine physicians. However, their data-gathering methods did not include information on sample collection methodology. In our work, this information was collected and used to assess preanalytical

Table 2. Hemolysis by Blood Sample Method: Intravenous Line Versus Straight Stick[a]			
Factor	Intravenous, No. (%) (n = 47 266)	Stick, No. (%) (n = 615)	P[b]
Not hemolyzed	42 445 (89.8)	582 (94.6)	<.001
Hemolyzed with comment	3870 (8.2)	26 (4.2)	<.001
Gross hemolysis	951 (2.0)	7 (1.1)	.12
Hemolyzed with comment and gross hemolysis	4821 (10.2)	33 (5.4)	<.001

[a] Sample method was missing for 265 cases.
[b] Pearson χ^2 test.

Table 3. Hemolysis by Straight Stick Placement Location[a]

Factor	Antecubital, No. (%) (n = 345)	Peripheral, No. (%) (n = 217)	Missing, No. (%) (n = 53)	P
Not hemolyzed	335 (97.1)[b]	198 (91.2)[c]	49 (92.5)	.01[d]
Hemolyzed with comment	9 (2.6)	13 (6.0)	4 (7.5)	.07[d]
Gross hemolysis	1 (0.29)[b]	6 (2.8)[c]	0 (0.0)	.03[e]
Hemolyzed with comment and gross hemolysis	10 (2.9)[b]	19 (8.8)[c]	4 (7.5)	.01[d]

[a] A significance level of .017 was used for pairwise ad hoc comparisons.
[b] Significantly different from peripheral.
[c] Significantly different from antecubital.
[d] Pearson χ^2 test.
[e] Fisher exact test.

Table 4. Hemolysis by Intravenous Placement Location[a]

Factor	Antecubital, No. (%) (n = 28 786)	Peripheral, No. (%) (n = 17 960)	Missing, No. (%) (n = 520)	P[b]
Not hemolyzed	26 669 (92.6)[c,d]	15 338 (85.4)[e]	438 (84.2)[e]	<.001
Hemolyzed with comment	1776 (6.2)[c,d]	2025 (11.3)[e]	69 (13.3)[e]	<.001
Gross hemolysis	341 (1.2)[c,d]	597 (3.3)[e]	13 (2.5)[e]	<.001
Hemolyzed with comment and gross hemolysis	2117 (7.4)[c,d]	2622 (14.6)[e]	82 (15.8)[e]	<.001

[a] A significance level of .017 was used for pairwise ad hoc comparisons.
[b] Pearson χ^2 test.
[c] Significantly different from peripheral.
[d] Significantly different from missing.
[e] Significantly different from antecubital.

Table 5. Hemolysis by Blood Sample Technique Through Intravenous Line (Vacuum Tube Versus Syringe)[a]

Factor	Vacuum Tube, No. (%) (n = 16 590)	Syringe, No. (%) (n = 705)	P[b]
Not hemolyzed	14 770 (89.0)	613 (87.0)	.09
Hemolyzed with comment	1370 (8.3)	72 (10.2)	.07
Gross hemolysis	450 (2.7)	20 (2.8)	.84
Hemolyzed with comment and gross hemolysis	1820 (11.0)	92 (13.0)	.09

[a] Sample technique was missing for 29 971 cases.
[b] Pearson χ^2 test.

Table 6. Hemolysis by Gauge of Straight Needle[a]

Factor	Other Size, No. (%) (n = 500)	16–20 Gauge, No. (%) (n = 112)	P[b]
Not hemolyzed	481 (96.2)	98 (87.5)	<.001
Hemolyzed with comment	13 (2.6)	13 (11.6)	<.001
Gross hemolysis	6 (1.2)	1 (0.89)	.78
Hemolyzed with comment and gross hemolysis	19 (3.8)	14 (12.5)	<.001

[a] Gauge was missing for 73 cases.
[b] Pearson χ^2 test.

Table 7. Hemolysis by Gauge of Intravenous Line[a]

Factor	Other Size, No. (%) (n = 5633) Summary	16–20 Gauge, No. (%) (n = 41 571) Summary	P[b]
Not hemolyzed	4694 (83.3)	37 689 (90.7)	<.001
Hemolyzed with comment	700 (12.4)	3170 (7.6)	<.001
Gross hemolysis	239 (4.2)	712 (1.7)	<.001
Hemolyzed with comment and gross hemolysis	939 (16.7)	3882 (9.3)	<.001

[a] Gauge was missing for 62 cases.
[b] Pearson χ^2 test.

factors associated with hemolysis. Lippi et al[11] found ED hemolysis rates more consistent with those reported in our study (3%–12.4%). Their review cited various preanalytical factors and references that examined some factors comparable with the work of Heyer et al[2] and our work. Some findings presented here are consistent, keeping in mind that hemolysis is defined differently in each laboratory setting. In spite of continuing reports of high ED hemolysis rates, a Q-Probes survey by the College of American Pathologists[10] also demonstrated that 70% of laboratories reported little to no progress in addressing the problem of hemolyzed specimens.

Table 8. Hemolysis by Straight-Stick Location and Gauge[a]					
Factor	Antecubital/16–20 Gauge, No. (%) (n = 76)	Peripheral/16–20 Gauge, No. (%) (n = 28)	Antecubital/Other Gauge, No. (%) (n = 269)	Peripheral/Other Gauge, No. (%) (n = 187)	P[b]
Not hemolyzed	70 (92.1)[c]	22 (78.6)[c]	265 (98.5)[d,e,f]	174 (93.0)[c]	<.001
Hemolyzed with comment	5 (6.6)	6 (21.4)[c,f]	4 (1.5)[e]	7 (3.7)[e]	<.001

[a] Location or gauge was missing in 55 cases. No gross hemolysis was observed with available location and gauge data performed with stick. A significance level of .008 was used for pairwise ad hoc comparisons.
[b] Pearson χ^2 test.
[c] Significantly different from antecubital/other gauge.
[d] Significantly different from antecubital/16–20 gauge.
[e] Significantly different from peripheral/16–20 gauge.
[f] Significantly different from peripheral/other gauge.

The finding that straight sticks were associated with lower hemolysis confirms previous study results.[3,12] The mechanism of this association is likely multifactorial and might be related to the physical structure of the IV catheter and the straight-stick needle. Manufacturers have indicated that softer materials used in IV catheters can stay open under positive pressures (eg, during infusion) but can collapse and crimp under negative pressures (vacuums) associated with blood draws, generating turbulent flow and hemolysis.[3] The smooth tip and solid/rigid inner lumen of a straight-stick needle versus a tapered tip and soft IV catheter could also contribute to less hemolysis because the former generates less turbulent flow. The barrier to more widespread implementation of straight-stick sample collection might be related to the fact that the likelihood that ED patients will require an IV line is uncertain at the time the sample is to be drawn. Therefore, in some EDs, when it is determined that blood samples are required, an IV catheter is inserted both for access and for collection of samples. Published reports comparing a single stick with an IV line versus 2 sticks (one for the blood draw and another for the IV line) typically show that use of a straight stick reduces hemolysis.[11] Our results suggest that use of a straight needle should be the preferred practice, because it is associated with a reduction in the hemolysis rate, but we have not evaluated the effect of this practice on patient preference or satisfaction. Intuitively, most patients should prefer 1 needle stick to 2, and identifying practices to reduce a second needle stick should be a priority.[13]

For sample collection resulting in reduced hemolysis, some EDs rely on phlebotomists, who draw blood only via a straight stick; medics and nurses can be also be instructed to use straight-stick blood draws only.[14,15] However, Dietrich[13] reported similar hemolysis among blood samples obtained from IV lines, which suggests that factors other than solely the use of a straight stick to obtain a sample might be influencing hemolysis. This is the fundamental issue: whether ED blood samples should be drawn from a single stick when placing an IV line, which many nurses, medics, and patients prefer, or whether 2 sticks should be used (one to obtain the samples via a straight-stick method and a second to place an IV line). The original Dietrich[13] publication supported the use of "one poke" but did not elaborate the specific source of the lower incidence of hemolysis. However, one source of the lower incidence might have been the use of lower-volume vacuum tubes during acquisition of samples from the IV line (H. Dietrich, oral communication, 2016).[16]

Heyer et al[2] also identified antecubital location as significantly associated with lower hemolysis rates when blood is obtained from an IV line.[2] Our study confirmed this finding, suggesting a 2-fold increase in hemolysis among samples collected from peripheral sites other than the antecubital location (Table 4). The antecubital site provides a larger-bore vein and is the preferred site according to the Emergency Nurses Association guidelines.[8] In addition, other factors might be associated with use of peripheral site blood draws. These include provider practice preferences and patient characteristics (eg, inability to access an antecubital vein, obesity), which could also explain differences in rates of hemolysis. Our study supports the use of antecubital blood draws when possible.

In our study, shorter tourniquet time was associated with a lower rate of hemolysis (Table 10). Heyer and colleagues[2] were not able to identify suitable reports of investigations of this association. Shorter tourniquet time might be associated with a lower hemolysis rate because the blood has less opportunity to remain in stasis. Factors that shorten tourniquet time are ease of the blood draw, patient characteristics, and the presence of fewer complications.

Table 9. Hemolysis by Intravenous Line Location and Gauge[a]					
Factor	Antecubital/16–20 Gauge, No. (%) (n = 27 233)	Peripheral/16–20 Gauge, No. (%) (n = 14 106)	Antecubital/Other Gauge, No. (%) (n = 1551)	Peripheral/Other Gauge, No. (%) (n = 3795)	P[b]
Not hemolyzed	25 264 (92.8)[c,d,e]	12 221 (86.6)[d,e,f]	1403 (90.5)[c,e,f]	3058 (80.6)[c,d,f]	<.001
Hemolyzed with comment	1648 (6.1)[c,d,e]	1496 (10.6)[d,e,f]	128 (8.3)[c,e,f]	529 (13.9)[c,d,f]	<.001
Gross hemolysis	321 (1.2)[c,e]	389 (2.8)[d,e,f]	20 (1.3)[c,e]	208 (5.5)[c,d,f]	<.001
Hemolyzed with comment and gross hemolysis	1969 (7.2)[c,d,e]	1885 (13.4)[d,e,f]	148 (9.5)[c,e,f]	737 (19.4)[c,d,f]	<.001

[a] Location or gauge was missing in 581 cases. A significance level of .008 was used for pairwise ad hoc comparisons.
[b] Pearson χ^2 test.
[c] Significantly different from peripheral/16–20 gauge.
[d] Significantly different from antecubital/other gauge.
[e] Significantly different from peripheral/other gauge.
[f] Significantly different from antecubital/16–20 gauge.

Table 10. Hemolysis by Tourniquet Time[a]

Factor	<60 s, No. (%) (n = 13 162)	>60 s, No. (%) (n = 3832)	P^b
Not hemolyzed	11 800 (89.7)	3300 (86.1)	<.001
Hemolyzed with comment	1038 (7.9)	392 (10.2)	<.001
Gross hemolysis	324 (2.5)	140 (3.7)	<.001
Hemolyzed with comment and gross hemolysis	1362 (10.3)	532 (13.9)	<.001

[a] Tourniquet time was missing in 37 537 cases.
[b] Pearson χ^2 test.

Further inquiry evaluating the physiologic mechanisms for this association is needed.

An additional finding of our study was that, among samples obtained from IV lines, larger-gauge needles were associated with lower rates of hemolysis (Table 7). Our results indicated a lower hemolysis proportion using a category (16–20 gauge) of larger-gauge needles for IV draws. It is possible this effect is evident within this group (ie, 16–20 gauge needles) as well, which may be important for prospective study (Table 7). Larger needles are likely used preferentially in patients from whom samples are easy to obtain. In addition, the larger needles probably have lower turbulence at their terminal tip, where presumed shearing forces contribute to hemolysis. Interestingly, among straight-stick samples in our analysis, hemolysis rates were higher with use of lower-gauge needles (Table 6). The interpretation of these results is not clear, but this was a relatively small subsample and the mechanism for this finding requires further validation. Moreover, our study also demonstrated that, for blood samples obtained through an IV line, the use of syringes or vacuum tubes did not have a statistically significant effect on hemolysis, similar to the findings of Heyer et al[2] (Table 5). Lastly, we also evaluated the use of ultrasound to guide placement of peripheral IV lines and obtain blood samples. Documentation of ultrasound use is part of the IV placement documentation template in our EMR. Typically, ultrasound is used only in scenarios of difficult-to-place IV catheters, and it is being used increasingly for IV and blood draws for patients with difficult access. (Ultrasound-placed IV lines indicate difficult IV access.) Although the hemolysis rate was slightly higher in all categories in the ultrasound-placed IV lines, the difference was not statistically significant (Table 11).

One criticism brought up in the Heyer et al[2] publication was that gauge and location might have been confounding variables. We evaluated the effects of location and gauge for samples obtained from IV lines and those obtained via straight stick. We found that antecubital location and large gauge were associated with lower hemolysis compared with peripheral location and large-gauge IV lines (Table 9). The lower hemolysis rates were also maintained for antecubital and lower-gauge IV lines versus peripheral sites and lower-gauge needles, indicating that gauge size alone seems to be the variable more closely related to hemolysis.

We evaluated the effects of gauge and location on hemolysis by the straight-stick method (Table 8). No gross hemolysis was identified with available location and gauge for straight stick. For samples obtained via straight stick (large gauge and antecubital location versus large gauge and peripheral location), there seemed to be increased hemolysis with a more peripheral location of the draw. A similar finding was that a lower hemolysis rate was found with antecubital location and other, smaller-gauge straight-stick needles (Table 8). However, our results indicated a higher incidence of hemolysis among straight-stick samples with larger-gauge needles (16 to 20 gauge) (Table 6). The meaning of this finding is not clear; however, it might represent the influence of other confounding factors or our limited sample size related to this comparison.

Hemolysis most often results from damage to red blood cells during collection. Preanalytic processes (both technique and equipment) play a significant role in hemolysis rates.[2,7] As stated above, the reported rates of hemolysis among blood samples obtained in EDs range from 6.8% to 19.8%, with some as high as 30%; these levels are markedly elevated compared with those in other hospital departments.[4] The American Society for Clinical Pathology established a benchmark of 2% or lower for hemolysis rates among laboratory blood samples, and most collection sites outside the ED achieve this level or better.[5] The fast-paced work environment of the ED and a patient-centered desire to avoid a second stick have led to the practice in some EDs of obtaining samples when IV access is established, but this practice, among others, contributes to higher hemolysis rates.[5,11] Laboratory medicine has identified ED hemolysis as a significant quality issue because of its impact on efficiency. Unfortunately, the national scope of the effect of hemolysis rates on ED patients remains unknown, but it is of utmost importance in a policy environment inundated with quality metrics and in the search for evidence that will form the basis for process improvement initiatives that will enhance outcomes and reduce costs.

There continue to be gaps in information that may be contributing to a lack of widespread adoption of ED practice guidelines around laboratory sample hemolysis.[2,8] Improving the quality of blood samples received from ED settings has been stressed previously, primarily for efficiency reasons.[2] Although efficiency is clearly a critical quality metric in this setting, other quality outcome domains are

Table 11. Hemolysis When Using Ultrasound to Obtain Blood Sample

Factor	No Ultrasound, No. (%) (n = 53 010)	Ultrasound, No. (%) (n = 1521)	P^a
Not hemolyzed	47 749 (90.1)	1343 (88.3)	.02
Hemolyzed with comment	4240 (8.0)	140 (9.2)	.09
Gross hemolysis	1021 (1.9)	38 (2.5)	.11
Hemolyzed with comment and gross hemolysis	5261 (9.9)	178 (11.7)	.02

[a] Pearson χ^2 test.

likely to be impacted. As quality outcome metrics are often related, it is difficult to affect one domain without affecting others such as clinical effectiveness, timeliness, patient-centered care, patient safety, and equity. These domains would likely improve with a consistent decrease in the proportion of hemolyzed samples received from the ED. However, until similar studies are performed, including the collection of outcome data related to these quality domains, the advantage for laboratories and the health care systems they operate in to adopt these best practices will remain hypothetical. Multi-laboratory projects aimed at performing the labor-intensive collection of outcome data that provide evidence for a statistically significant association between use of best practices and clinical outcomes are needed for these best practices to be adopted on a much wider scale.

There are several strengths and weaknesses of the study to consider with interpretation of the findings. A significant strength of the study is the large number of samples collected, allowing comparisons of different factors identified as potentially associated with hemolysis. In addition, our electronic health record allowed systematic data collection of many of these preanalytical variables on sample collection techniques. Several limitations must also be considered. The results are derived from a single institution, which may limit their external validity. In addition, several variables had significant missing data elements, which may include systematically (rather than randomly) different values from samples in which all data were documented. Although ideally we would collect each of these in a systematic manner, there are cases in clinical practice in which they cannot be obtained because of competing priorities. We do appreciate that from a research perspective, a complete data set is ideal, but as discussed previously, we do not feel that these missing data systematically bias our primary conclusions. Finally, consistent with any observational retrospective study, results may reflect underlying confounders and cause-and-effect relationships cannot be inferred. Other factors might affect hemolysis beyond the variables captured in the current study: for example, provider variability, equipment type, and patient characteristics that were not identified for our analysis.

CONCLUSIONS

This study confirms previous findings that the use of straight needles and the antecubital location is significantly associated with reduced hemolysis, supporting the original conclusion of Heyer and colleagues.[2] In addition, our findings indicate that shorter tourniquet time (less than 60 seconds) and the use of larger-gauge needles for IV draws were significantly associated with lower hemolysis. No association was found between syringe versus vacuum tube sample collection in regard to incidence of hemolysis. These findings confirm and support best practices in the ED to reduce hemolysis and improve efficiencies in the acquisition of blood samples.

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References

1. National Hospital Ambulatory Medical Care Survey: 2011 emergency department summary tables. Centers for Disease Control and Prevention Web site. www.cdc.gov/nchs/data/ahcd/nhamcs_emergency/2011_ed_web_tables.pdf. Accessed April 25, 2016.

2. Heyer NJ, Derzon JH, Winges L, et al. Effectiveness of practices to reduce blood sample hemolysis in EDs: a laboratory medicine best practices systematic review and meta-analysis. *Clin Biochem*. 2012;45(13–14):1012–1032.

3. Grant MS. The effect of blood drawing techniques and equipment on the hemolysis of ED laboratory blood samples. *J Emerg Nurs*. 2003;29(2):116–112.

4. Soderberg J, Jonsson PA, Wallin O, Grankvist K, Hultdin J. Haemolysis index—an estimate of preanalytical quality in primary health care. *Clin Chem Lab Med*. 2009;47(8):940–944.

5. Lowe G, Stike R, Pollack M, et al. Nursing blood specimen collection techniques and hemolysis rates in an emergency department: analysis of venipuncture versus intravenous catheter collection techniques. *J Emerg Nurs*. 2008;34(1):26–32.

6. Phelan MP, Reineks EZ, Schold JD, Kovach A, Venkatesh A. Estimated national volume of laboratory results affected by hemolyzed specimens from emergency departments. *Arch Pathol Lab Med*. 2016;140(7):621.

7. Phelan MP, Schold J, Reineks E, Meldon S, Cornish N, Procop G. Projecting the potential impact of reductions in hemolysis among US emergency departments. Abstract and poster presented at: Annual Meeting of the American College of Medical Quality; March 25–28, 2015; Alexandria, Virginia.

8. Proehl JA, Bradford JY, Leviner S, et al. Clinical practice guideline: prevention of blood specimen hemolysis in peripherally-collected venous specimens. Emergency Nurses Association Web site. www.ena.org/practice-research/research/CPG/Documents/HemolysisCPG.pdf. Accessed April 29, 2016.

9. Howanitz PJ, Lehman CM, Jones BA, Meier FA, Horowitz GL. Practices for identifying and rejecting hemolyzed specimens are highly variable in clinical laboratories. *Arch Pathol Lab Med*. 2015;139(8):1014–1019.

10. Howanitz PJ, Lehman CM, Jones BA, Meier FA, Horowitz GL. Clinical laboratory quality practices when hemolysis occurs. *Arch Pathol Lab Med*. 2015; 139(7):901–906.

11. Lippi G, Plebani M, Di Somma S, Cervellin G. Hemolyzed specimens: a major challenge for emergency departments and clinical laboratories. *Crit Rev Clin Lab Sci*. 2001;48(3):143–153.

12. Kennedy C, Angermuller S, King R, et al. A comparison of hemolysis rates using intravenous catheters versus venipuncture tubes for obtaining blood samples. *J Emerg Nurs*. 1996;22(6):566–569.

13. Dietrich H. One poke or two: can intravenous catheters provide an acceptable blood sample?: a data set presentation, review of previous data sets, and discussion. *J Emerg Nurs*. 2014;40(6):575–578.

14. Straszewski SM, Sanchez L, McGillicuddy D, et al. Use of separate venipunctures for IV access and laboratory studies decreases hemolysis rates. *Intern Emerg Med*. 2011;6(4):357–359.

15. Barnaby DP, Woolowitz A, White D, et al. Generalizability and effectiveness of butterfly phlebotomy in reducing hemolysis. *Acad Emerg Med*. 2016;23(2):204–207.

16. Phelan MP, Reineks EZ, Kovach AC. Intravenous catheters and blood samples [letter to editor]. *J Emerg Nurs*. 2017;42(2):102–103.