Original Article

Observational Study to Determine Factors Associated with Blood Sample Haemolysis in the Emergency Department

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Abstract

Introduction: Haemolysis of blood samples is a common problem encountered in the Emergency department (ED). It leads to inaccurate blood results and has cost implications as blood samples very often have to be retaken. The purpose of our study was to determine which factors in blood sampling were associated with higher rates of haemolysis. Materials and Methods: An observational convenience sample of all patients presenting to the ED requiring blood urea and electrolyte (UE) analysis were eligible for our study. Questionnaires were distributed to the doctors and nurses conducting blood sampling to determine the method used and outcome data were collected after the samples were processed. Results: Out of 227 UE samples analysed, 45 (19.8%) were haemolysed. Various factors, including method (IV cannulation or venepuncture), system (syringe or vacutainer), operator, rate of blood flow, difficulty of cannulation/venepuncture and source of blood (arterial or venous), were analysed, but their effects on haemolysis were not statistically significant (*P* >0.05). However, the use of the vacutainer system was associated with the highest rates of haemolysis [adjusted odds ratio (OR), 6.0; 95% confidence interval (CI), 2.3 to 15.1]. Conclusion: We found blood sampling with the vacutainer system to have increased rates of haemolysis. This could potentially change attitudes towards equipment used for blood sampling in the ED.

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Key words: Chemistry testing, Emergency department, Sample haemolysis, Venepuncture, Venous cannulation

Introduction

Haemolysis of blood samples leads to inaccurate results and often necessitates a repeat sample.[1-3] Escalating workloads and finite resources are an increasing problem in many Emergency departments (EDs),[4] where many conditions have time-dependent outcomes, and accurate and quick blood results are thus important. Erroneous blood results lead to unnecessary delays and additional costs by necessitating obligatory repeat samples. Repeat blood sampling can also cause unnecessary pain to patients.

Haemolysis can occur in-vivo and in-vitro. In-vitro haemolysis affects assay results by underestimating albumin, alkaline phosphatase (ALP) and sodium, and overestimating alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatine kinase (CK), and especially potassium levels.[2,5] In the ED, inaccurate potassium levels can lead to potential misdiagnosis and dangerous management as treatment protocols for both hyperkalaemia and hypokalaemia are drastically different.

Factors that have been suggested to cause increased sample haemolysis include pressure differences and needle size,[6,7] prolonged time between sample collection and analysis,[8,9] size of collection tubes,[10] difficulty of blood drawing,[11] and the use of a vacutainer system.[12,13]

We aimed to determine which factors of the blood sampling process were associated with the highest rates of haemolysis. Results of this study could potentially assist us in modifying phlebotomy methods in our ED to minimise blood sample haemolysis.

Materials And Methods

Questionnaire

A questionnaire was designed with relevance to current phlebotomy methods and equipment available in our ED, and distributed amongst the potential blood sampling operators (consultants, registrars, medical officers, nurses

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and medical/nursing students). This recorded their personal method of sampling a particular UE blood sample with reference to the following variables:

1) Method [venepuncture or intravenous (IV) cannulation]
2) System [arterial blood gas (ABG) sampling or arterial puncture, syringe, vacutainer]
3) Size of needle from either syringe (23G or 21G) or IV cannulation (24G, 22G, 18G or 16G)
4) Operator (consultant, registrar, medical officer or student)
5) Blood flow (fast, moderate or slow)
6) Difficulty of venepuncture/cannulation (easy, moderate or hard)
7) Source (venous or arterial)
8) Sample volume in millilitres (mL)
9) Time sample taken
10) Time sample processed by the laboratory. One questionnaire was completed for every blood sample obtained.

Samples

No limitations were placed on the operators in terms of number of blood samples taken and personal methods used. All patients presenting to the ED requiring blood UE analysis were eligible for our study regardless of demographics, comorbidities or presenting complaint. The patients' label was included in the questionnaire for ease of retrospective identification via the UE results from the biochemical laboratory to determine the main outcome, whether the blood UE sample was haemolysed or not. Sample lysis was determined by the biochemistry laboratory with the use of a standardised validated method determined by the laboratory. Actual UE results were not relevant for our study.

Table 1. Characteristics of Study Samples

Characteristic	Option	*n*	%
Method	IV cannula	168	74.0
	Venepuncture	59	26.0
System	Syringe	146	64.3
	Vacutainer	81	35.7
Size of needle	</=21G	86	37.9
	>21G	141	62.1
Operator	Consultant	18	7.9
	Registrar	18	7.9
	Medical officer	137	60.4
	Student/ Nurse	54	23.8
Blood flow	Fast	92	40.5
	Moderate	102	44.9
	Slow	33	14.5
Difficulty of venepuncture/ cannulation	Easy	146	64.3
	Moderate	52	22.9
	Hard	29	12.7
Source	Venous	219	96.9
	Arterial	7	3.1
Lysis	Yes	45	19.8
	No	182	80.2

IV: intravenous; G: gauge

Statistical Analysis

All analyses were done using JMP (release 5.1). The differences in sample processing intervals and sample volumes with regard to haemolysis rates were determined using a 2-sample *t*-test. The associations between the various categorical factors and the rate of haemolysis were assessed via Chi-square/Fisher's exact tests. A multivariate logistic regression model was performed. $P < 0.05$ was considered statistically significant.

Results

A total of 227 blood samples and completed questionnaires were collected and included in our study for analysis, out of which 45 (19.8%) were haemolysed. Table 1 documents the blood sample characteristics as gathered by the various operators on the questionnaires.

Table 2 reveals the various characteristics with their associated rates of haemolysis and odds ratio (OR) with a 95% confidence interval (CI). The size of the needle, operator, and perceived rate of blood flow and difficulty of cannulation had poor associations with sample haemolysis rates ($P > 0.05$). Both the use of vacutainer and IV cannulation appear to be associated with the highest rates of haemolysis [OR, 4.5 (2.3, 9.0) and OR, 4.4 (1.5, 13)] respectively. However, after adjustment with a logistic regression model with haemolysis as the outcome, the use of the vacutainer was associated with significantly higher rates of haemolysis [adjusted OR, 6.0 (2.3, 15.1)].

There was no association between sample processing intervals and sample volume with haemolysis rates, as indicated in Table 3. The mean processing interval time that resulted in haemolysis was 65.2 minutes (SD, 27.7 minutes), as compared to the non-haemolysis interval mean of 59.7 minutes (SD, 25.8 minutes). The mean sample volume that resulted in haemolysis was 3.9 mL (SD, 1.8 mL), as compared to the non-haemolysis volume mean of 4.7 mL (SD, 3.1 mL).

Discussion

In this study, we found that the use of the BD vacutainer

Table 2. Various Factors Related to Blood Sampling and their Associated Rates of Haemolysis

Characteristic	Option	Sample lysed (%)	OR (95%CI)
Method	Venepuncture	4 (6.8)	
	IV cannula	41 (24.4)	4.4 (1.5, 13.0)
System	ABG, Syringe	16 (11.0)	
	Vacutainer	29 (35.8)	4.5 (2.3, 9.0)
Size of needle	</=21G cannula	15 (17.4)	
	>21G cannula	30 (21.3)	1.3 (0.6, 2.5)
Operator	Registrar	2 (11.1)	
	Medical officer	22 (16.1)	1.5 (0.3, 7.1)
	Consultant	4 (22.2)	2.3 (0.4, 14.4)
	Student/Nurse	17 (31.5)	3.7 (0.8, 17.8)
Blood flow	Fast	14 (15.2)	
	Moderate	23 (22.5)	1.6 (0.8, 3.4)
	Slow	8 (24.2)	1.8 (0.7, 4.7)
Difficulty of	Hard	4 (13.8)	
venepuncture/	Easy	27 (18.5)	1.6 (0.8, 3.4)
cannulation	Moderate	14 (26.9)	1.8 (0.7, 4.7)
Source	Arterial	1 (14.3)	
	Venous	44 (20.1)	1.5 (0.2, 12.9)

CI: confidence interval; G: gauge; IV: intravenous; OR: odds ratio

Table 3. Relationship between Sample Volume and Processing Interval, and Sample Haemolysis

Factor	Haemolysis (n)	Mean	SD	OR (95%CI)
Sample volume (mL)	Yes (45)	3.9	1.8	0.9 (0.8, 1.0)
	No (182)	4.7	3.1	
Interval (min)	Yes (45)	65.2	27.7	1.0 (1.0, 1.0)
	No (182)	59.7	25.8	

OR: odds ratio; SD: standard deviation

system (BD Medical, Franklin Lakes, NJ USA) was associated with the highest rates of haemolysis. This is similar to previous studies that have also suggested that the vacutainer system, especially when used with intravenous (IV) cannulas, may increase sample lysis.[12,13] The BD vacutainer system uses evacuated blood collection tubes, which draw in a blood sample through a closed vacuum system into the collection tubes.

In planning our study, many other factors associated with the various methods of phlebotomy were initially presumed to have an influence on haemolysis. In previously reported studies, an inverse relationship between needle diameter and rate of haemolysis[2,7,13,14] was found. However in our study, we did not find a relationship between needle diameter and haemolysis rates. Haemolysis rates were also found to be operator-independent, which could indicate the importance of phlebotomy equipment and methods rather than the experience of the operator. In contrast to previous studies, we also found that sample volumes[10] and processing intervals[8,9] had no effect on haemolysis rates.

We also had the opportunity to determine the preferred method of phlebotomy in our ED via the questionnaires. It appears that operators preferred smaller gauge IV cannulation with a syringe to draw blood. However, the majority of the operators were medical officers, hence these data may not be an accurate reflection of senior ED staff.

One of the limiting factors of this study was the subjective assessment of the rate of blood flow and difficulty of phlebotomy.[11] We did not have a standard objective measure to gauge blood flow and difficulty of phlebotomy, so these parameters were determined subjectively by the operators. Sample volumes drawn using the syringe method would be objectively recorded, however subjective judgements were used to estimate sample volumes in vacutainer tubes, which do not have volume markings.

Another consideration besides sample lysis for the use of vacutainers is the issue of prevention of needle-stick

injury,[15,16] A proposed advantage of closed vacuum systems is that there is no need to transfer samples (as in the needle and syringe method), reducing the risk of needle-stick injuries. However, in our ED, we propose that if a syringe is used to draw a blood sample, whether from an IV cannula hub or from a venepuncture, a needle-less method should be used for sample transfer. This involves uncapping a collection tube and squirting a blood sample directly into a tube from the syringe, without using a needle, reducing the risk of needle-stick injury. Care should be taken, however, to prevent blood spillage.

We intend to follow up with a programme to reduce haemolysis rates in the ED based on the findings of this study. We intend to conduct an educational programme addressing known factors associated with sample lysis. This will include recommending the use of a syringe rather than the vacutainer system in the ED for blood sampling, especially when blood is drawn directly from an IV cannula hub. Sample lysis rates will then be measured in a post-intervention study.

Conclusion

Vacutainers were found to result in higher rates of blood haemolysis compared to obtaining blood samples with a syringe. We intend to follow up with a programme to reduce haemolysis rates in the ED based on the findings of this study. This could potentially change attitudes towards equipment used for blood sampling in the ED.

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