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ORIGINAL ARTICLE

The impact of blood sampling technique, including the use of peripheral intravenous cannula, on haemolysis rates: A cohort study

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Aims: To explore the relationship between blood sampling techniques and haemolysis.

Background: Haemolysis rates of blood samples have been thought to be influenced by the method of collection. There is a lack of research evidence available to clearly show the comparative risk of haemolysis across different blood sampling methods, including venepuncture and use of peripheral intravenous cannulas.

Design: A prospective cohort study. Reporting followed the STROBE checklist.

Methods: A trained observer was used to record blood sampling techniques over a 10-week period between April and June 2019. These records were then linked to pathology haemolysis results. Multivariable logistic regression was used to model patient and blood draw characteristics affecting haemolysis.

Results: Most of the blood samples were not haemolysed (n = 324, 87.1%). Multivariable analysis showed haemolysis was associated with increased tourniquet duration and if the level of tube was less than half full. Univariable analysis showed haemolysis was associated with increased age of the patient, the difficulty of cannulation/ venepuncture and increased number of attempts. No difference was found in the haemolysis rate related to the qualification of the blood collector.

Conclusion: There was no significant difference in haemolysis rates associated with sampling blood from a PIVC compared with venepuncture. Research should be undertaken to determine whether education on the factors influencing haemolysis is useful in decreasing haemolysis rates.

Relevance to clinical practice.

There was no association with increased haemolysis rates when drawing blood via venepuncture compared with a peripheral intravenous cannula. Haemolysis of blood samples was associated with increased tourniquet duration, if level of the tube was less than half-filled, increased age of the patient and difficulty of blood draw.

Awareness of the risk of haemolysis associated with specific blood sampling methods may assist clinicians to improve care.

KEYWORDS
accuracy, blood sampling, cannulation, emergency, haemolysis, intravenous, nurses, nursing, venepuncture

'What does this paper contribute to the wider global clinical community?':

- There was no association with increased haemolysis rates when drawing blood via venepuncture compared with a peripheral intravenous cannula.
- Haemolysis of blood samples was associated with increased tourniquet duration, if the level of the tube was less than half-filled, increased age of the patient and difficulty of blood draw.
- Awareness of the risk of haemolysis associated with specific blood sampling methods may assist clinicians to improve care.

1 | INTRODUCTION

The ability to accurately and reliably test patient blood samples is integral to quality provision of care (Lippi et al., 2019). Inaccurate or corrupted samples can lead to misdiagnosis, but more usually the need to resample at considerable discomfort to patients, time and cost to healthcare and organisations (Abbas et al., 2017; Phelan et al., 2018). Health professionals understand the importance of obtaining accurate blood samples; however, there is variability in the techniques or methods that they use to achieve this (Bentley et al., 2016). Some of the variation in practice and policies regarding blood sampling is in part due to limited research on the safety of the practice for blood sampling, such as from using a peripherally inserted intravenous cannula (Coventry et al., 2019). Policies from Australian Government Health Departments show a paucity of high-level evidence supporting the development of their guidelines for withdrawing blood from a peripheral intravenous cannula (PIVC) (Jacob et al., 2020).

One common reason that blood samples may become unusable is due to haemolysis. Haemolysis is a leading cause of specimen rejection in clinical laboratories accounting for up to 70% of unsuitable specimens (Abbas et al., 2017; Lippi et al., 2008; Pilbeam et al., 2013). Haemolysis is caused by damage to the erythrocytes cell membrane resulting in the release of haemoglobin and intracellular component into the surrounding plasma (Abbas et al., 2017). Knowing the methods of blood sampling, which are least likely to lead to haemolysed samples, is important; however, there is currently little research available to clearly show the comparative risk of haemolysis across a variety of blood sampling methods, including venepuncture and use of a PIVC, to provide clinicians and policymakers with clear guidance in this area.

1.1 | Background

Peripheral intravenous cannulas (PIVCs) are used internationally as an intervention for patients in acute health services to assist with the management of conditions and are the most inserted vascular access device (Carr et al., 2016). Most PIVCs are inserted to administer intravenous fluids and medications or to obtain pathology blood tests (Alexandrou et al., 2015; Craige et al., 2017; Fry et al., 2016; Wong et al., 2018). It is estimated that up to 80% of hospitalised patients will require intravenous therapy at some point during their inpatient hospital stay (Yagnik et al., 2017). Although blood samples are traditionally drawn from peripheral venepuncture, increasingly PIVCs are being used for blood sampling (Carr et al., 2016). A prevalence study found that over 51% of nurses surveyed in Australia used PIVCs for blood sampling (Davies, 2019) and the practice is also common in other countries (Craige et al., 2017).

There is a great degree of variance in practice regarding obtaining blood samples from intravenous cannulas internationally, between health services, states of Australia and individual nurses (Davies, 2019). The method of sample collection, including collection procedures, handling and storage, is thought to account for up to 93% of errors associated with the blood diagnostic process (Lippi et al., 2006). Some health services have guidelines against the practice of blood sampling from PIVC, and others do not have guidelines (Jacob et al., 2020). Systematic literature reviews have been undertaken to find existing evidence in relation to the safety and accuracy of drawing blood from PIVC (Coventry et al., 2019; Jeong et al., 2019; Lesser et al., 2020). The reviews found the practice of taking blood from PIVCs was safe (Jeong et al., 2019; Lesser et al., 2020) or recommended further research into the practice (Coventry et al., 2019).

Arguments for obtaining blood samples from PIVC include minimisation of invasive and painful venepuncture, convenience of access and appropriateness for certain populations. PIVCs are a recommended way to collect blood from any patient in emergency situations, who is at increased risk of bleeding, have limited vascular access or those requiring multiple blood sampling (Seemann & Reinhardt, 2000). It is common practice for blood to be taken from existing PIVC in paediatric patients due to the trauma involved in venepuncture (Department of Health WA, 2017), but this practice has not routinely occurred in adult populations.

One of the main reasons provided in the literature for not using PIVCs for blood collection was an increased risk of haemolysis. Some researchers have found increased rates of haemolysis in blood samples taken from newly inserted IVs (Coventry et al., 2019; Lowe et al., 2008), and existing IVs (Grant, 2003), as opposed to those taken through the traditional means of venepuncture.

2 | THE STUDY

2.1 | Aims

The aim of this prospective cohort study was to explore the practice of blood sampling from venepuncture and PIVC in acute healthcare settings. The primary outcome measure was haemolysis of the blood sample.

2.2 | Design

The project involved a prospective cohort study where researchers observed the methods used to sample blood from patients and analysed haemolysis reported in results. The project was developed in consultation with hospital staff from the emergency department, quality department, nursing administration, pathology and university researchers. The reporting of this study followed the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) checklist for cohort studies (Supplementary File 1).

2.3 | Sample/Participants

The project was undertaken in a large metropolitan hospital in Australia. The hospital has around 300 patients per day (100,000 patients annually) emergency department presentations. Blood sampling in the department was routinely undertaken by pathology-trained collectors, medical doctors and nursing staff. Health professionals included in the study were medical personnel, nurses and pathology collectors. Health professionals were notified of the study by the project team at three ward meetings prior to the commencement of the study and by signs placed in the department. Adult patients who presented to the department and required blood sampling between April and June 2019 were included in the study. Paediatric patients, patients presenting with a primary diagnosis of mental illness and patients who were unable to provide consent were excluded.

2.4 | Data collection

A member of the research team attended the emergency department to observe the practice of blood collection, either from a PIVC or from venepuncture. Observations were undertaken over a 10-week period, for an average of 4 hr per day for 5 days per week. The same researcher undertook all observations. As only one observer was in the department, not all blood samples collected in the department were observed due to the number of patients requiring blood samples at the same time. Only blood samples that were observed were included in the study. The observer used a hard copy data collection tool to record the data on blood sampling technique. The tool was developed for the project to record blood collection techniques and handling practices (see Appendix S1). The tool was assessed for face validity prior to use by three clinical staff and three academics. Blood samples were collected by the usual method of the health professional and processed according to standard health service policies. The data tool included collection of the patient's medical record number. Medical record numbers for patients whose blood sampling was observed were transferred onto a hard copy form and provided to the pathology laboratory. No information on the method of blood collection was provided to the pathology laboratory staff. The pathology laboratory assessed haemolysis through the use of a fully automated spectrophotometry system and provided information on which samples were haemolysed and the level of haemolysis (0–4). Medical records for patients with haemolysed samples were accessed to determine whether the blood sample was repeated within six hours.

2.5 | Ethical considerations

Ethical approval was received from both the participating health service (No. 1832) and the university (No. 18384JACOB). An information sheet was given to patients describing the study, and verbal consent was obtained from both the patient and the health professional obtaining the blood sample. All patients consented to the observation of their blood being collected, but several health professionals (n=five) declined to be observed.

2.6 | Data analysis

Data were entered into Microsoft Excel (Microsoft) and then exported into SPSS Version 26.0.0.1 (IBM SPSS Statistics). Categorical data were described using frequencies with percentages, and continuous data were described using medians and interquartile ranges expressed as the 25th to 75th percentile. Haemolysed blood samples were decided a priori to be dichotomised to 'not haemolysed' (haemolysis level 0) and haemolysed (including haemolysis level 1 to level 4). Comparisons of demographic characteristics of the cohort and characteristics of the blood draw between not haemolysed and haemolysed blood samples were performed using chi-square or Fisher's exact for categorical variables and the Mann–Whitney test for medians for continuous variables.

Logistic regression was used to estimate the odds ratio (OR) and the 95% confidence interval (CI) for all demographic characteristics and characteristics of the blood draw by not haemolysed compared with haemolysed blood samples. Models were adjusted for age (as a continuous variable) and gender. All variables with $p < .157$ (Sauebrei et al., 2020) in initial tests of association in univariable

TABLE 1 Demographic characteristics of participants.

	All patients (N = 373)		Not haemolysed (n = 325, 87.1%)		Haemolysed (n = 48, 12.9%)		
Variable	**n (%)**	**Median (IQR)**	**n (%)**	**Median (IQR)**	**n (%)**	**Median (IQR)**	***p*-value**
Age (years)	373 (100)	62.0 (42.5–76.0)	325 (100)	62.0 (41.5–75.0)	48 (100)	70.0 (52.5–81.8)	.006[a]
Observations							
Systolic blood pressure	328 (87.9)	139.0 (125.0–154.8)	284 (87.4)	138.0 (125.0–154.0)	44 (91.7)	140.0 (125.0–156.0)	.77[a]
Diastolic blood pressure	328 (87.9)	78.5 (68.2–88.8)	284 (87.4)	78.5 (69.0–89.0)	44 (91.7)	77.5 (66.2–85.0)	.38[a]
Heart rate	342 (91.7)	76.5 (66.0–88.0)	298 (91.7)	77.5 (67.8–88.0)	44 (91.7)	75.5 (70.0–84.2)	.48[a]
Temperature (Celsius)	314 (84.2)	36.5 (36.1–37.0)	274 (84.3)	36.5 (36.1–37.0)	40 (83.3)	36.5 (36.2–37.0)	.30[a]
Haematocrit	369 (98.9)	0.4 (0.4–0.4)	323 (99.4)	0.4 (0.4–0.4)	46 (95.8)	0.4 (0.4–0.4)	.76[a]
		n (%)	**n (%)**				
Gender							
Female		210 (56.3)	188 (89.5)		22 (10.5)		.12[b]
Male		163 (43.7)	137 (84.0)		26 (16.0)		
Admission diagnosis							
Abdominal		117 (31.4)	103 (88.0)		14 (12.0)		.98[c]
Neurological		43 (11.5)	35 (81.4)		8 (18.6)		
Cardiac		60 (16.1)	53 (88.3)		7 (11.7)		
Pain		17 (4.6)	15 (88.2)		2 (11.8)		
Respiratory		54 (14.5)	45 (83.3)		9 (16.7)		
Fall		16 (4.3)	14 (87.5)		2 (12.5)		
Infection		15 (4.0)	14 (93.3)		1 (6.7)		
Genitourinary		18 (4.8)	16 (88.9)		2 (11.1)		
Fracture		2 (0.5)	2 (100.0)		0 (0.0)		
Reproductive		15 (4.0)	14 (93.3)		1 (6.7)		
Other		16 (4.3)	14 (87.5)		2 (12.5)		

[a]Mann-Whitney U test.

[b]chi-square test.

[c]Fisher's exact test.

logistic regression models were included in the multivariable logistic regression models. These multivariable models were simplified in a stepwise fashion by removing the variable with the least significant p-value and refitting the models until only variables with *p*-values < .05 were retained. As a final check, all excluded variables were retested one at a time. A *p*-value < .05 was considered significant.

2.7 | Validity and reliability

Data were collected using an audit tool designed by the authors. An extensive review of the literature and consultations with clinical experts provided face validity of the audit tool. Pilot testing was conducted with four clinical experts to ensure usability of the tool.

Haemolysis was tested by spectrophotometry using a Beckman Coulter AU680 Chemistry Analyzer. The machine was routinely used by the pathology facility and was regularly calibrated according to the pathology protocols. This method of analysis provides a higher reliability for haemolysis levels than commonly used visual analogue scales.

Data collection was standardised through the use of a single clinical expert, and any questions related to data entry were discussed with the research team, and decisions were based on consensus. Data analysis was completed separately and confirmed by two authors.

3 | RESULTS/FINDINGS

During the study period (April 2019 to June 2019), data were collected from 373 participants (female, n = 210, 56.3%; males, n = 163, 43.7%). Median age of the participants was 62 (IQR: 42.5–76) years,

TABLE 2 Number of cannulation attempts

Number of cannulation attempts	n	(%)
1	287	76.9
2	53	14.2
3	12	3.2
4	8	2.1
5	2	0.5

and most of the patients had an abdominal (n = 117, 31.4%), cardiac (n = 60, 16.1) or a respiratory (n = 54, 14.5%) admission diagnosis. Median vital signs were all within normal limits. Most of the blood samples were not haemolysed (n = 324, 87.1%) compared with the haemolysed samples (n = 48, 12.9%). The only demographic characteristic that was significantly associated with haemolysis was age of the participant (median age 62 compared with 70 years, $p = .006$) (see Table 1).

Characteristics of the blood draw showed that the majority were obtained on insertion of the PIVC (n = 323, 86.8%). Most cannulation/venepuncture insertions were classified as easy (n = 266, 73.9%), with the most common insertion site being the cubital fossa (n = 294, 79.9%). The tourniquet was in place for a median of two minutes (range one to 12 min), and the median number of cannulation/venepuncture attempts was 1 (range one to five attempts). Twenty per cent of the cohort required more than one attempt, and one patient required 5 attempts to draw blood for collection (see Table 2). The PIVC size most commonly used was 20 g (n = 271, 81.6%), the most common venepuncture needle size was 21G (n = 23, 56.1%), the average syringe size was 10 mL (n = 282, 95.6%) and vacutainers were mostly not used (n = 293, 79.0%). The level of the blood tube was mostly filled greater than half full (331, n = 91.2%), and blood tubes were often not rotated at completion (n = 219, 58.7%). No blood samples were found to be re-draw within six hours of the original sample, indicating no change to clinical practice was required due to haemolysis.

The variables that were significantly different between the not haemolysed group and the haemolysed group were tourniquet duration total ($p = .01$), ease of cannulation/venepuncture ($p = .007$), number of cannulation attempts ($p = .03$), amount of blood collected ($p = .002$) and level of tube filled ($p < .001$). There was no significant difference in haemolysis associated with the use of PIVC for sampling as compared with venepuncture (see Table 3).

After using a multivariable model to adjust for age and gender, the variables associated with a haemolysed blood sample were tourniquet duration (OR: 1.22; 95% CI: 1.03–1.44, $p = .02$) and if the level of the blood tube was less than half full (OR: 8.18; 95% CI: 3.62–18.4, $p < .001$). That is, for every extra minute the tourniquet was in situ the odds were 22% more likely for the sample to be haemolysed. If the blood tube was less than half full compared with greater than half full, the odds of a haemolysed sample were 8.18-fold increased (see Table 4).

4 | DISCUSSION

This study found that haemolysis of blood samples was not related to whether blood was collected via either an intravenous cannula or direct venepuncture. The most important factors identified by multivariable analysis associated with haemolysis in this study included the length of time the tourniquet was tightened and the level at which the blood collection tube was filled. Other variables identified through univariable models included the difficulty of the cannulation/ venepuncture was, the number of attempts made to access the vein and the age of the patient from which the blood was collected. All of these factors may occur simultaneously in a single patient when practitioners have difficulty in accessing a vein. These findings concur with Phelan et al., (2018) who found that multiple factors are associated with haemolysis.

Tourniquet duration was found to be a significant factor in haemolysis with the greater the time the tourniquet was in situ the greater the chance of haemolysis. Tourniquet time is measured from the time the tourniquet was applied until it was released following the collection of the blood sample. Prolonged tourniquet time has previously been thought to increase haemolysis Lippi et al., (2008). The average tourniquet time for non-haemolysed samples in this study was 120 seconds. This is a longer period than previous studies by Phelan et al., (2018) who found that tourniquet time greater than 60 seconds increased rates of haemolysis.

The level to which the vacuum blood collection tube was filled was found to be related to haemolysis. This has been identified in studies with paediatric patients where the increased pressure in the collection tube due to the low blood volume was thought to increase the haemolysis of blood specimens (Hu et al., 2020). Hu et al., (2020) identified that blood samples collected in which the pressure in the specimen container was removed by uncapping the tube resulted in significantly lower haemolysis rates. While vacuum blood collection tubes are routinely used to guarantee clean specimens and increase safety, they require an adequate volume of blood being drawn and the tube should be filled using the negative pressure (Clinical & Laboratory Standards Institute, 2017). For patients with difficult blood draws, it may be difficult to fulfil the standard for adequate blood volumes, which may have increased the haemolysis rate. Phelan et al., (2018) found that collecting smaller volumes of blood using small volume vacuum blood collection tubes in emergency departments significantly decreased the rates of haemolysis.

The number of cannulation/venepuncture attempts was found to influence haemolysis rates. High numbers of accessing attempts were found to increase the haemolysis rate of the blood sample. Many hospitals have policies regarding the number of attempts at cannulation/venepuncture a health practitioner may undertake in accessing a vein (Department of Health WA, 2017; Sou et al., 2017). This is in an effort to decrease pain to patients and the increased risk of phlebitis, thrombosis and catheter-related infections from multiple vein accesses (Sou et al., 2017). Despite these policies, health practitioners in this study were found to have

TABLE 3 Characteristics of the blood draw

Variable	All patients (N = 373)		Not haemolysed (n = 325, 87.1%)		Haemolysed (n = 48, 12.9%)		*p*-value
	n (%)	Median (IQR)	n (%)	Median (IQR)	n (%)	Median (IQR)	
Tourniquet duration total (min)	372 (99.2)	2.0 (2.0–4.0)	324 (99.7)	2.0 (2.0–3.0)	48 (100)	3.0 (2.0–5.0)	.01[a]
Number of cannulation/venepuncture attempts	362 (97.3)	1.0 (1.0–1.0)	315 (96.9)	1.0 (1.0–1.0)	47 (97.9)	1.0 (1.0–2.0)	.03[a]
Amount of blood collected	297 (79.6)	10.0 (10.0–10.0)	258 (79.4)	10.0 (10.0–10.0)	39 (81.3)	10.0 (6.0–10.0)	.002[a]
Time between collection and transfer to tube (min)	372 (99.2)	1.0 (1.0–2.0)	324 (99.7)	1.0 (1.0–2.0)	48 (100)	1.0 (1.0–1.0)	.32[a]
Time between collection and transfer to chute (min)	359 (96.2)	7.0 (5.0–10.0)	312 (96.0)	7.0 (5.0–10.0)	47 (97.9)	7.0 (5.0–9.0)	.35[a]
Time between collection and processing (min)	372 (99.2)	68.0 (57.2–84.0)	324 (99.7)	68.0 (57.0–83.8)	48 (100)	68.0 (60.2–90.8)	.25[a]
	n (%)		n (%)		n (%)		
Blood taken							
From established PIVC	9 (100)		8 (88.9)		1 (11.1)		.92[c]
On insertion of PIVC	323 (100)		280 (86.7)		43 (13.3)		
Venepuncture	40 (100)		36 (90.0)		4 (10)		
Ease of cannulation/venepuncture							
Difficult	31 (8.6)		25 (80.6)		6 (19.4)		.006[c]
Moderate	63 (17.5)		48 (76.2)		15 (23.8)		
Easy	266 (73.9)		240 (90.2)		26 (9.8)		
Insertion site							
Dorsum hand and wrist	59 (16.0)		47 (79.7)		12 (20.3)		.15[c]
Forearm	15 (4.1)		14 (93.3)		1 (6.7)		
Cubital fossa	294 (79.9)		260 (88.4)		34 (11.6)		
Cannula size (mm)							
22 and 24	11 (3.3)		8 (72.7)		3 (27.3)		.11[c]
20	271 (81.6)		233 (86.0)		38 (14.0)		
18	50 (15.1)		47 (94.0)		3 (6.0)		
Needle size (mm)							
23 and 25	18 (43.9)		16 (88.9)		2 (11.1)		>.99[c]
21	23 (56.1)		21 (91.3)		2 (8.7)		
Syringe size (ml)							
10	282 (95.6)		245 (86.9)		37 (13.1)		.68[c]
20	13 (4.4)		11 (84.6)		2 (15.4)		
Vacutainer used							
Yes	78 (21.0)		68 (87.2)		10 (12.8)		.97[b]
No	293 (79.0)		255 (87.0)		38 (13.0)		

(Continues)

TABLE 3 (Continued)

	n (%)	n (%)	n (%)	
Level of tube filled				
Less than half	32 (8.8)	17 (53.1)	15 (46.9)	<.001[b]
Greater than or equal to half	331 (91.2)	300 (90.6)	31 (9.4)	
Blood tubes rotated after transfer				
No	219 (58.7)	189 (86.3)	30 (13.7)	.57[b]
Yes	154 (41.3)	136 (88.3)	18 (11.7)	
Haemolysed				
None	325 (87.1)	325 (100)		
Level 1	29 (7.8)		29 (60.4)	
Level 2	9 (2.4)		9 (18.8)	
Level 3	3 (0.8)		3 (6.3)	
Level 4	7 (1.9)		7 (14.6)	

[a]Mann-Whitney U test.
[b]chi-square test.
[c]Fisher's exact test.

between one and five attempts to access a vein before successful blood collection.

The gauge of the cannula for IV blood draw was not found to affect the haemolysis rates. This is in contrast to Phelan et al., (2018) who found that large gauge cannulas for IV draws decreased haemolysis rates. Other studies have also shown that the size of the cannula affected haemolysis with smaller gauges (>21) seen to increase the risk (Dugan et al., 2005; Kennedy et al., 1996; Tanabe et al., 2003).

The location of the vein used for the blood draw was not found to be a significant factor affecting haemolysis rates of blood samples. This contrasts with previous findings, which found that the location of the draw influenced haemolysis rates with antecubital locations reducing the rate (Phelan et al., 2018). The insertion site of a cannula has been thought to influence the ability to aspirate blood through a PIVC (Gagne & Sharma, 2017). Lippi et al., (2014) also found that haemolysis was increased when blood samples were taken from a PIVC distal to a median sized vein, but this was not found to affect haemolysis rates of the blood samples in the current study.

The rates of haemolysis found in this study (12.9%) were higher than those reported in the literature for patients from other emergency departments. Phelan et al., (2018) reported rates of 10% in an urban tertiary emergency department in the USA, and Craige et al., (2017), 6.8% in two emergency departments in university teaching hospitals. This contrasts with the 22.4% previously reported by Pilbeam et al., (2013) for emergency departments in Australia. The different rate of reported haemolysis may be related to the population group, with higher rates of haemolysis seen in patients with difficult access to the veins. The difficulty of the blood draw was found to influence the haemolysis rate in this study. Difficulty of the blood draw was determined based on the clinical decision of the observing nurse and based on their perceived ability to aspirate blood, the number of attempts required to gain blood, the size of the vein able to be accessed, and whether previous attempts had been made to cannulate the patient. Phelan et al., (2018) reported that emergency department haemolysis tends to be consistently higher than benchmarks of 2% haemolysis set by the American Society for Clinical Pathology. Another possible reason for the discrepancy is the way that the haemolysis of samples is measured and assessed. For our study, we used an automated technique. Other studies have also used the automated technique (Corbo et al., 2007; Dietrich, 2014; Munnix et al., 2011; Wollowitz, 2013), and others, the visual techniques (Grant, 2003; Lowe et al., 2008; Seemann & Reinhardt, 2000).

There was no delay in patient care as a result of haemolysis for any of the samples in this study. While on the surface, our results are contrasting to the findings of Lippi et al., (2008) who identified haemolysis as the cause of up to 70% of unsuitable specimens, the finding of limited patient delay may not be inconsistent. Not all haemolysed blood samples are rejected by pathology as samples can still be processed except for electrolytes such as potassium and other tests influenced by haemolysis (Abbas et al., 2017). This means that while haemolysis may have occurred, it may not have caused delays in patient care. Lesser et al., (2020) also found that haemolysis of blood samples did not cause significant errors in results for the most blood test. Clinical relevance of haemolysis appears to be related to individual patients and their specific testing requirements.

No difference was found in the haemolysis rate related to the qualification of the blood collector with similar results found in haemolysis between medical, nursing and pathology staff. This contrasts with previous reports by Pilbeam et al., (2013) who suggests it is the lack of training of the blood collector that causes increased haemolysis rates in emergency departments. The decrease in haemolysis rates for pathology collectors reported by Pilbeam et al., (2013) may be due

TABLE 4 Univariable models for haemolysed group. None compared to the haemolysed group levels 1, 2, 3 and 4 combined

	Univariable models			Multivariable models		
	OR	95% CI	*p*-value	OR	95% CI	*p*-value
Age (years)	1.02	1.00–1.04	.01	1.02	1.00–1.03	.10
Gender						
Female	0.62	0.34–1.13	.12	0.65	0.32–1.30	.23
Male	1.00			1.00		
Systolic blood pressure	1.00	0.99–1.01	>.99			
Diastolic blood pressure	0.99	0.97–1.01	.34			
Heart rate	0.99	0.97–1.01	.37			
Temperature (Celsius)	1.31	0.86–2.00	.21			
Haematocrit	3.29	0.01–1467.8	.70			
Sample collected by			.58			
Medical officer	1.05	0.41–2.68	.92			
Nurse	0.68	0.22–2.11	.51			
Pathology	1.00					
Time of blood draw			.91			
18:00–23:59	1.29	0.40–4.10	.88			
12:00–17:59	1.06	0.50–2.26	.67			
06:00–11:59	1.00					
Tourniquet duration total (min)	1.23	1.06–1.44	.01	1.22	1.03–1.44	.02
Blood taken			.83			
From PIVC	1.38	0.47–4.08	.56			
On insertion	1.12	0.11–11.46	.92			
Venepuncture	1.00					
Ease of cannulation/venepuncture			.009			
Difficult	2.22	0.83–5.90	.11			
Moderate	2.88	1.42–5.85	.003			
Easy	1.00					
Number of cannulation/ venepuncture attempts	1.54	1.08–2.19	.02			
Insertion site			.15			
Dorsum hand and Wrist	1.95	0.94–4.04	.07			
Forearm	0.55	0.07–4.29	.56			
Cubital fossa	1.00					
Cannula size (mm)			.14			
22 and 24	5.88	1.00–34.39	.05			
20	2.56	0.76–8.62	.13			
18	1.00					
Needle size (mm)						
23 and 25	1.31	0.17–10.35	.80			
21	1.00					
Syringe size (ml)						
10	0.83	0.18–3.90	.81			
20	1.00					

(Continues)

TABLE 4 (Continued)

	Univariable models			Multivariable models		
	OR	95% CI	*p*-value	OR	95% CI	*p*-value
Amount of blood collected	0.90	0.80–1.01	.07			
Vacutainer used						
Yes	0.99	0.47–2.08	.97			
No	1.00					
Level of tube filled						
Less than half	8.54	3.89–18.8	<.001	8.18	3.62–18.8	<.001
Greater than or equal to half	1.00			1.00		
Blood tubes rotated after transfer						
No	1.20	0.64–2.24	.57			
Yes	1.00					
Time between collection and transfer to tube (min)	1.21	0.88–1.66	.25			
Time between collection and transfer to chute (min)	0.96	0.89–1.04	.28			
Time between collection and processing (min)	1.00	1.00–1.00	.27			

to blood collection by pathology collectors being undertaken predominately on stabilised patients who are either hospitalised or day procedures. A systematic review by Makjumula-Nkhoma et al., (2019) identified no RCT studies that have demonstrated that venepuncture training reduced haemolysis rates and other studies were unclear. Similarly, a study by Abbas et al., (2017) found no difference in rejection rates for blood samples post-phlebotomy training for health workers. No studies were found that examined the education of staff regarding blood sampling from intravenous cannulas.

No difference in haemolysis was found to exist between blood collected through a needle and syringe and a vacutainer. This is similar to Phelan et al., (2018) who reported that haemolysis of samples by syringe (13%) and vacuum (13%) was identical. Other studies have suggested that collection of blood using a vacutainer and PIVC resulted in increased haemolysis when compared to collection through a syringe and needle (Grant, 2003; Ong et al., 2008; Pilbeam et al., 2013).

This study suggests that healthcare staff, including nurses, doctors and other practitioners, can take blood effectively and that the qualifications of staff members do not alter haemolysis rates. Comparable rates of haemolysis across the PIVC and venepuncture cohorts provide evidence that whether a sample is retrieved through PIVC or venepuncture is not relevant to the risk of haemolysis occurring. However, the choice of whether to sample from PIVC or venepuncture may be significant to patient comfort.

Practices such as filling of vacuum tubes and decreasing length of tourniquet time may assist in decreasing haemolysis. This may relate to how practitioners prepare for procedures as many people put tourniquet on and then organise their trolley for the procedure. The use of smaller vacuum tubes in emergency and education of staff may also be beneficial in decreasing the number of attempts required to obtain blood samples and hence decrease haemolysis rates. Education of health professionals should be undertaken to decrease the rates of haemolysis of blood samples.

Further research should be undertaken to determine whether education on the factors influencing haemolysis rates is useful in decreasing the haemolysis rates in emergency departments. Other questions for research include whether drawing blood from a PIVC affects the life/ dwell time of the catheter or causes phlebitis or bloodstream infection.

4.1 | Limitations

Even though we observed a large number of blood samples (n = 373), it is possible this study was underpowered to detect significant results. Due to the observational nature of this study, we did not conduct a power calculation for sample size. Also, this research was undertaken at one health service, so blood collection practices may be different at other facilities. As only one observer was present in the emergency department, not all blood collection procedures were observed. Paediatric patients, patients with a primary diagnosis of mental health and patients who were unable to consent were not included. This may have led to the sample excluding those presenting with serious trauma or the highest levels of illness severity.

5 | CONCLUSION

Haemolysis of blood samples was due to multiple factors such as the level the tube was filled, length of time the tourniquet was in situ, number of needle stabs required to access the vein, difficulty

of cannulation and the age of the patient. No difference was found in haemolysis rate between blood obtained from intravenous cannulas and venepuncture or between healthcare personnel. Guidelines and policies on PIVCs should be updated and blood collection from PIVCs should be considered a possibility. Haemolysis should not be used as a justification to restrict healthcare staff from obtaining blood samples from PIVC. Understanding the factors involved in haemolysis can assist practitioners to adjust their practice to decrease the rate of sample rejection and increase patient safety.

6 | RELEVANCE TO PRACTICE

Nurses are required to undertake blood sampling as part of their role. Understanding the factors that increase haemolysis of blood samples may assist clinicians to improve care and limit the need for resampling. Being able to make evidence-based choices on the method of blood sampling may improve the safety of the practice and enable nurses to select the best method of blood sampling for the particular patient situation.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Elisabeth Jacob conceptualised the study, designed methodology, investigated the data, curated the data, wrote the original draft, supervised the study, administered the project and acquired funding. Alycia Jacob conceptualised the study, designed methodology, investigated the data, analysed the data, wrote the original draft and acquired funding. Linda Coventry conceptualised the study, designed methodology, analysed the data, wrote the original draft and acquired funding. Hugh Davies conceptualised the study, designed methodology, investigated the data, wrote, reviewed and edited the manuscript, and acquired funding. Darren Jacob conceptualised the study, collected the data collection, analysed the data, and wrote, reviewed and edited the manuscript. Mark Jenkins analysed the data, and wrote, reviewed and edited the manuscript. Margaret Husain conceptualised the study, and wrote, reviewed and edited the manuscript.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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