

## Phlebotomy- A gateway to laboratory diagnostics

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### ABSTRACT

**Objective:** To assess the knowledge, practice and attitude about venipuncture amongst Nursing staff of a tertiary care hospital.

**Material and Methods:** This questionnaire-based survey was done at Rawal General and dental hospital, Islamabad, from January to April 2022. The Questionnaire was developed as per the CLSI H3-A6 and WHO's venipuncture guidelines. It was distributed to nursing staff by hand. Most were filled inside the classes in front of the teacher.

**Results:** The frequency of correct responses in the knowledge section varied from 4(4.6%) about the number of tube inversions to 79(90.8%) about wearing of gloves before taking a sample while 47(54%) selected the correct option of identifying a patient. Regarding the standard phlebotomy protocols, the response was quite low which was 26(29.9%), 18(20.7%) and 8(9.2%) about the correct angle of needle insertion, what to inspect of the supplies and the location of applying the tourniquet respectively. Recapping with two hands is still done by 13% and 79% still put the sample by holding tube in the other hand. In spite of all this 66% still think that one can learn venipuncture by practicing the techniques without going through a proper course.

**Conclusion:** Nursing staff in Rawal General and dental hospital is not fully aware about basic laboratory protocols and their importance. This unawareness can lead to generation of erroneous lab reports. Formal education, training with summative assessments should be a part of the curriculum of nursing courses followed by rigorous implementation monitoring.

**Keywords:** Awareness, Nurses, Phlebotomy, Venipuncture, Sample

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### INTRODUCTION

The importance of medical laboratory tests, especially the blood examination as the baseline investigation as well as the specified tests for proper diagnosis is undeniable [1].

The quality of results of the blood examinations majorly depends upon the skills and knowledge of the phlebotomist. It is important that any medical laboratory should have a well-trained phlebotomist, as even minor

negligence in the standard operating procedure of blood sampling may give false results leading to misdiagnosis and improper management of the disease [2]. In most of the hospitals in Pakistan, it has been observed that blood sample collection in admitted patients is mostly done by the nurses, who are not properly trained according to the standard guidelines of blood sampling set by WHO [3]. Moreover, there are no standard operating procedures of blood sampling and infection control practices displayed in the wards thus leading to many pre-analytical errors. The most common error is that the blood sample is hemolysed due to improper vigorous mixing of blood or when the blood is collected through intravenous catheter of size less than 20G [3,4]. It has also been observed that many samples that come for the blood

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cultures are already contaminated with the normal flora of the skin of the patient due to lack of awareness of proper patient preparation before blood collection [5]. Many other studies from Asian and African countries have also shown the inappropriate knowledge of health care professionals especially related to patient preparation, tourniquet application time, order of draw, identification of the appropriate collecting tubes required for different blood tests, sample transportation and storage [1,6]. To avoid all these errors and to provide accurate and reproducible results, it is of vital importance to regularly evaluate the knowledge of phlebotomy procedures of our nursing staff to update their training according to the standardized procedures set by World Health Organization [7].

Since we were facing the problems related to pre-analytical errors in our reports, we conducted this study to assess the knowledge, skill and attitude regarding awareness on the blood sampling procedure among the nurses and to provide references for improving the sampling practice at our hospital.

## MATERIAL AND METHODS

This descriptive cross-sectional was conducted at the Department of Microbiology, RG&DH Islamabad from January to April 2022. Sample size was calculated through WHO calculator using the following formula

$$N = z^2 \frac{P(1-P)}{d^2}$$

Where P= Expected proportion in population based on previous studies [8] and with 95% level of confidence, the calculated sample size was 73. Due to possible lack of response, we oversampled by 16%, making the final sample size 86. Non-probability consecutive sampling technique was used to collect data. The questionnaire was based on KAP (Knowledge, Attitude and Practice) having 26 questions on knowledge, 9 questions on practice and 3 questions on attitude regarding awareness of blood sampling procedure among nurses. For the knowledge questions, incorrect responses were given a 0 score, while 1 point

was assigned for choosing the correct answer. The expected minimum and maximum total knowledge score were 0 and 26, respectively. Attitude towards blood sampling procedure was measured by 3 questions. A statement with options yes and no were given 1 and 0, respectively. The expected maximum total attitude score was 3. Practice was scored 1 for standard practice and 0 for other. Participants' KAP levels were defined as "good" or "poor" based on Bloom's cut off point. Participants with knowledge scores above 60% were regarded as having good knowledge, while those with score below 60% were considered having poor knowledge. Participants with attitude scores of 59% and below were considered as having an unacceptable attitude, while those within the range of 60-79% moderate and score above 80% were regarded as having a good attitude. For practice section, participants with scores >80% and <80% were classified as taking acceptable and unacceptable blood sampling measures, respectively.

The collected data were analyzed using SPSS version 22.0. Descriptive statistics were used to summarize the demographic characteristics of the sample data. The KAP assessment was conducted out by assigning scores to the variables. Bivariate statistics (Pearson's Chi square) was conducted to check the association of participant's knowledge scores with their attitude and practice scores.

## RESULTS

A total of 86 questionnaires were distributed. There were 57(66%) females and 29(34%) males. Almost all of the investigated nurses were aware that patient identification should be confirmed prior to venous blood sampling. However, the frequency of nurses who knew the right procedures to identify a conscious patient was 47(54%). Few, 18(20.7%) knew what to check about the phlebotomy supplies. Although 72.4% of the nurses knew proper tourniquet releasing time, the correct rates on the tourniquet applying location were quite low, 8(9.2%). In addition, a good number of the investigated nurses knew that gloves should be worn during phlebotomy and the proper time

to put on gloves 79 (90.8%) & 71(81.6%) respectively. The knowledge about correct angle of insertion of the needle was known to very few, 26(30%). The rest of the results on knowledge of the pre-sampling phase are shown in Table-I.

The knowledge of mix by inverting blood collection tubes is shown in Table-II. The tube inversions were lower than those of recommended times. In the practice section (Table-III), the frequency of nurses who still recap the needle with two hands is 12(13.8%) while 69(79.3%) put the sample by holding tube in the other hand. The correct rates on where to dispose needles were relatively high 72 (82.8%) while the rest used the medical waste bin for disposal. A good percentage perform hand hygiene before putting on gloves i.e 73(83.9%).

During sampling 45(51.7%) of the nurses were not practicing the right order of draw during multi-tube sampling. The rest of the answers' detail is shown in the table.

In the Attitude section (Table-IV) quite a high number of nurses (58-66%) believed that not performing hand hygiene when wearing gloves, taking the sample even before the antiseptic is dry and no need for a formal phlebotomy training was ok.

The P-values were 0.329 and 0.152, indicating no significant association of Knowledge with Attitude and Practice for the given study.

**Table-I: Knowledge of the investigated nurses about the pre-sampling phase.**

Questions	Wrong answer	Correct answer
<b>K1.</b> Patient identity should be confirmed prior to venous blood sampling? True False	3(3.4%)	84(96.6%)
<b>K2.</b> What is the right procedure to identify a patient who is conscious? Ask a patient to give his/her full name Nurse states a patient's full name or bed number Check a patient's bed tag	40(46%)	47(54%)
<b>K3.</b> What should be inspected about the supplies? (multi-choice) Expiry dates of phlebotomy devices Looseness or defects of the tube cap Appropriate tubes according to the test requests All of the above	69(79.3%)	18(20.7%)
<b>K4.</b> Which vein is preferred for venipuncture Median cubital vein / median vein Basilic vein Other veins	24(27.6%)	63(72.4%)
<b>K5.</b> Where is the proper location to apply a tourniquet? 3.5 - 5.0 cm above the venipuncture site 5.0 - < 7.5 cm above the venipuncture site 7.5 - 10.0 cm above the venipuncture site 10.0 - < 12.5 cm above the venipuncture site	9(90.8%)	8(9.2%)
<b>K6.</b> How long can tourniquet application last? ≤ 60 seconds >60 seconds	24(27.6%)	63(72.4%)
<b>K7.</b> Gloves should be worn during phlebotomy? True False	8(9.2%)	79(90.8%)
<b>K8.</b> When is the proper time to put on gloves? Before assembling supplies Before performing venipuncture	16(18.4%)	71(81.6%)
<b>K9.</b> While taking blood sample, the needle should enter at an angle of 45-50° 35-40° 15-30°	61(70.1%)	26(29.9%)

**Table-II. Knowledge of mix by inverting times for blood collection tubes.**

<b>K21.</b> How many times the red cap (non-additive serum tube) should be inverted for mixing after blood collection?		
Mixing not required		
1-4		
more than 5	48(55.2%)	39(44.8%)
<b>K22.</b> How many times the yellow cap (tube with clot activator and gel plasma separator) should be inverted for mixing after blood collection?		
Mixing not required	83(95.4%)	4(4.6%)
1-4		
more than 5		
<b>K23.</b> How many times the Lavender cap (EDTA tube) should be inverted for mixing after blood collection?		
Mixing not required		
1-4		
more than 5	69(79.3%)	18(20.7%)
<b>K24.</b> How many times the green cap (Heparin tube) should be inverted for mixing after blood collection?		
Mixing not required		
1-4		
more than 5	80(92.0%)	7(8.0%)
<b>K25.</b> How many times the <b>blue</b> cap (1:9 Sodium citrate tube) should be inverted for mixing after blood collection?		
Mixing not required		
1-4	35(40.2%)	52(59.8%)
more than 5		
<b>K26.</b> How many times the <b>grey</b> cap (Glycolysis inhibitor tube) should be inverted for mixing after blood collection?		
Mixing not required		
1-4		
more than 5	73(83.9%)	14(16.1%)

**Table-III: Practice questions responses.**

Items	n (%)
Where do you dispose the needle after phlebotomy?	
Medical waste dustbin	15(17.2%)
Sharps container	72 (82.8%)
How many times have you taken a blood sample?	
>20 times	57(65.5%)
<20 times	30(34.5%)
How do you recap a needle?	
I don't recap	28(32.2%)
With single hand	47(54%)
With two hands	12(13.8%)
After taking the blood sample, how do you put the blood in the tube?	
By keeping the tube in a rack	18(20.7%)
By holding tube in one hand and putting the sample by the other	69(79.3%)
Do you touch the venipuncture site after disinfecting it?	
Never	69(79.3%)
Sometimes	13(14.9%)
Always	5(5.7%)
In admitted patients how do you take blood sample?	
From a new venipuncture	50(57.5%)
From existing peripheral venous access site	11(12.6%)
From first introducing an I.V device before connecting the cannula	26(29.9%)
Do you perform hand hygiene before putting on gloves?	
Yes	73(83.9%)
no	14(16.1%)
When taking sample for multiple tests for (a) Blood Culture (b) Red top (LFTs)	

and (c) lavender top (CP). Which tube do you inoculate on first, 2nd & 3rd number?	
A, B & C	45(51.7%)
B, A & C	41(47.1%)
C, B & A	1(1.1%)
Do you allow the antiseptic to dry before taking blood?	
Yes	66(75.9%)
no	21(24.1%)

**Table IV. Attitude questions' responses**

Items	Yes%	No%
<b>A1</b> It is OK NOT to perform hand hygiene if we are wearing sterile gloves before venipuncture.	37(42.5)	50(57.5)
<b>A2</b> When in a hurry we may take a sample, even before an antiseptic is dry.	30(34.5)	57(65.5)
<b>A3</b> One can learn venipuncture by practicing the techniques without going through a proper course.	30(34.5)	57(65.5)

**Table-V: Table of significance.**

Knowledge	Attitude			Practice	
	Good	Moderate	Unacceptable	Acceptable	Unacceptable
Good	12(21.4%)	3 (13.0%)	3 (37.5%)	3 (42.9%)	15 (18.8%)
Poor	44(74.6%)	20(87.0%)	5 (62.5%)	4 (57.1%)	65 (81.3%)
	p-value = 0.329			p-value = 0.152	

## DISCUSSION

The knowledge of pre analytical process of laboratory tests plays a vital role in quality assurance of test results leading to diagnostic approach. In our country, the facility of trained phlebotomists is not available in most of the hospitals so blood sampling procedure is usually done by nursing staff, post graduate residents and house officers. The awareness of patient identification, proper vacutainers, tourniquet application, needle handling, order of draw and proper mixing of blood sample to avoid hemolysis is crucial for quality laboratory results. It is a bitter reality that most of the healthcare workers are unaware of proper protocol of blood sampling technique [8,9]. We have conducted this study to analyze the proficiency of nursing staff in our hospital to analyze their blood sampling technique to improve the quality of our laboratory test results.

In our study, we did a questionnaire-based survey involving 86 trained nurses, 39% having more than 5 years of work experience. Questions were designed to assess the participant's knowledge and practical approach regarding pre sampling, sampling and post sampling phase.

Before taking blood sample, confirmation of patient identity is as significant as the

sampling technique. Almost all nurses (96.6%) were aware of the importance of proper identification however, only 54% selected the correct identifier that should be used for a fully conscious person. When we compared our results with other studies done around the globe, we found consistent results in other questionnaire-based studies in Pakistan and China, in which 64% and 58.8% nurses knew the correct procedure of patient identification respectively [8,10]. Research done in a tertiary care hospital in Turkey showed higher number of nurses (78%) who are unaware of proper identification procedure [4]. Two more studies performed in India and Croatia found 86.6% and 70% phlebotomists respectively who knew the proper identification process, which is much higher than our results [12,13].

In pre sampling phase, we asked questions to evaluate the knowledge of study participants about the correct site for venipuncture and correct method of sanitization and use of gloves before taking blood sample. We found that 72.4% participants knew the preferred vein for blood sampling i.e. median cubital vein.

When we compared our results with other studies, we came to know that knowledge of suitable site for blood collection is more or



less the same. As a study done in Sri Lanka in 2021 showed 83% participants with correct response [14]. Another study done at a tertiary care hospital's nurse in Turkey found 89% subjects with the correct answer<sup>11</sup> while in a study done in China, 87.2% of investigated nurses knew the preferred site [10].

While concerning proper method of sanitization which includes type of antiseptic used and use of gloves before taking blood sample, we came to know that 73.6% subjects knew the correct concentration of alcohol i.e. 70% should be used for sanitization. Almost all study subjects (90.8%) knew the use of gloves in phlebotomy. Same results were obtained in a study from India in which 88% participants were aware of proper use of gloves during phlebotomy and 84.6% answered correctly about the antiseptic [12]. Another questionnaire-based survey done in Turkey showed 82.5% subjects who knew about the correct use of antiseptic [15]. While another research done in the same country showed that majority (92%) wore clean gloves before phlebotomy procedure [11]. Our results were a little higher than a study done in China in 2018 in which 67% nurses knew the proper method of wearing gloves during phlebotomy.<sup>10</sup> Probably because that was a multicenter study and a little old too compared to ours.

In the sampling phase, we focused on the knowledge and skill of subjects about recommended procedure of tourniquet application, the method acquired after application of antiseptic at venipuncture site and precautions to avoid needle stick injuries. We were also concerned to check the awareness of our study participants about an important component of phlebotomy procedure, the correct order of draw.

In our study, majority of nurses, 90.8%, did not know the correct location for tourniquet application and 72.4% gave correct responses about tourniquet application time. When we compared our responses with other studies, we found that 62.4% nurses who participated in a study conducted in Sahiwal, Pakistan knew about the correct position of tourniquet application [8]. When we analyzed other studies

done in Pakistan and in neighboring countries, our results regarding tourniquet application time are more or less the same i.e 74%, 78%, 65.6% and 84% respectively [12,10,8,11] (evaluation of phlebotomy in India, China (parent article), Pakistan and Turkey).

In our survey, we also checked the phlebotomist routine after applying antiseptic whether they give proper time of alcohol to dry before taking blood sample or if they touch the venipuncture site to feel the vein shaft after disinfecting the site. Only 19.5% participants knew the recommended time which should be given to dry the disinfectant, while on the other hand, 79% subjects answered that they never touch the venipuncture site after disinfectant application.

When we analyzed other studies for comparison, we came to know that a study done in a tertiary care hospital in Pakistan reported 52% phlebotomists who gave proper time (30 seconds) for drying of alcohol.<sup>8</sup> Another study carried in Lahore, which evaluated the pre analytical errors in post graduate trainees found 35.2% residents with knowledge of drying time of alcohol.<sup>9</sup> Another questionnaire based survey in India showed 88.6% participants with correct response.<sup>12</sup> These results are in contrast to our results that emphasizes on the importance of refresher training courses for nurses and phlebotomists in our set up.

When we assessed the knowledge of participants whether they retouch the cleaned site or not in other researches, we found that 91% subjects in a survey performed in India did not touch the phlebotomy site after disinfection [12]. Another study conducted in Sri Lanka with 100 participants, one third of study subjects admitted that they palpate the site after disinfection [14]. A study done in Izmir; Turkey showed that 96.4% subjects do not touch the sanitized site [15]. These results clearly show the high standard of training in this country.

Needle stick injury (NSI) is a significant risk factor to health care workers. Awareness of accidental prick should be the important component of nursing training program. Recapping of used needle after taking blood sample, transfer of blood sample in the

vacutainer tube and dealing with uncooperative patients without any support are important factors which may lead to needle stick injury [12]. A study done in Karachi showed 53.7% nurses who were exposed to NSI [16]. When evaluating this skill, we found only 20.7% nurses who used correct procedure of blood transfer to vacutainer tubes and half of the subjects (57.5%) knew the protocol if they get an accidental prick. A study in India declared 13.3% participants who got NSI [12]. In the study done in Karachi, Pakistan in 2023 found 59.7% nurses with low level of knowledge regarding NSI [16]. Research done in Turkey in 2020 showed that correct safety procedure for blood sampling was adopted only by 38% study participants [11].

The awareness of correct order of draw has an important role in minimizing the pre analytical errors. In our study, 47% phlebotomists were aware of correct order of draw. On comparison with other studies, we found similar results in a survey done in tertiary care hospital in Pakistan showing 49% correct responses [8]. Two other studies done in Asian countries showed dissimilar results with quite less number of nurses with correct knowledge about order of draw, 18% and 15.5% [12,10]. The study done in Turkey also found only 22% nurses with correct response [15]. This deficiency was also seen in studies on postgraduate trainees. Only 24% and 55% participants gave correct answer [17,19].

In post sampling phase, an important thing to avoid is hemolysis of blood sample. The common factors leading to hemolysis of blood sample are the use of needle with improper gauge, forceful pressing of plunger while transferring blood and insufficient knowledge of proper method of blood mixing. In our study, we checked the awareness regarding proper skill of inversion of tubes for mixing of blood sample and calculated the mean percentage of correct responses which came out to be 25.7%.

While a study conducted in Pakistan found only 8.8% nurses who knew about appropriate method of mixing of additive with blood sample [8]. Another study done in Turkey showed a mere 6% of nurses which were aware of correct mixing protocol [11]. However, an

analysis done in Europe found 47% subjects with correct response [13]. Probably missed especially in hours of heavy workload.

The P-values being insignificant shows that in spite of knowledge non-adherence to the SOPs needs strict measures including inspection and clinical audits to make sure that the SOPs are followed. Further studies for the effect of strictness are needed for this purpose.

## CONCLUSION

This study shows that significant number of nursing staff in our setup are not fully aware of basic protocols of venipuncture and their importance, which may lead to generation of not only erroneous lab reports but also agonizing the patients in case they have to revisit for repeat sampling. Formal education and training including summative assessments should be a part of curriculum of the nursing courses. Last but not the least it is the strictness on implementation which is most importantly required.

## CONFLICT OF INTEREST

Authors declare no conflict of interest.

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Declared none

## AUTHORS CONTRIBUTION

**Shehla Ambreen Alizai:** Literature search, study design and concept, questionnaire design, data collection, drafting

**Rabia Sadaf:** Discussion writing

**Maliha Atif:** Introduction writing

**Kanwal Shehzadi:** Statistical analysis

**Naima Noor and Muhammad Saeed:** Data collection

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