Pre-analytical errors in clinical biochemistry-a comparative study

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Abstract

Introduction and Objectives: Laboratory testing is very important for diagnosing a disease, monitoring its progress and to monitor the response in patients to treatment. This study analyses the effects of reinforcing skill training among the laboratory personnel on the frequently occurring pre-analytical errors in clinical biochemistry samples.

Materials And Methods: Retrospective analysis of biochemistry laboratory records, of a tertiary care hospital, between two time points, i.e., April - October 2016 (7 months) and April- October 2017 (7 months) were compared. The laboratory personnel had undergone reinforcement training in between the two phases. Data analysis was done by using Epi-info Software version 3.4.3. Frequencies and percentages, which are part of descriptive statistics, were calculated.

Results: We received a total of 2, 77,438 patient samples (1, 30, 647 samples in 2016 and 1, 46,791 samples in 2017) for a period of 14 months. For the year 2016, the total number of pre-analytical errors was 1,215 (0.93%) and for the year 2017 it was 1,110 (0.76%). Based on the occurrence of the pre-analytical errors, the order recorded was: - haemolysis (77%), insufficient quantity of sample (8%), errors during sample transport (6%), errors during specimen handling (5%), and wrong tube collection (4%).

Conclusion: In order to safeguard patient interest and improve as well as ensure proper medical and testing services to patients we have to maintain proper quality control in all the phases of analysis and take efforts to reinforce and improve the skills of the laboratory personnel, especially in the pre-analytical phase. For achieving this, regular audits and proper monitoring is necessary.

Keywords: Pre-analytical errors, Turnaround time (TAT), Total testing process (TTP), India.

Introduction

The definition of good quality in laboratory medicine can simply be defined as the guarantee that each and every step in the total testing process (TTP) is performed correctly, thus assuring a proper informed medical decision which results in effective patient care. Lundberg, (1981), introduced the concept of the 'brain-to-brain loop' for completely describing the testing process. TTP can be defined as the generation of any lab test result which involves the following steps: ordering of a test, collection of the sample, identification and verification of the identity of patient, sample transportation preparation, analysis, reporting, and action.[1]

Even though the 'brain-to-brain loop' idea was first introduced about 40 years ago, it is still very essential and important in ensuring quality for concerned physicians as well as patients. The medical scene has changed considerably and has led to many improvements in the quality of laboratory services.[2]There has been a marked reduction in the analytical error rate in the past decade, due to improvements in the standardisation of analytical techniques, better quality reagents, and efficient instrumentation, and also due to progress in information technology as well as quality control and quality assurance methods.[3]

Quality indicators (QIs) in our laboratories focus mainly on the efficiency of analytical processes.[4] According to a study done by Plebani *et al.*, (2010); most of the errors were seen outside of the analytical phase. Such studies strongly suggest that the pre and post-analytical steps are even more vulnerable to risk of errors.[5]Although all the studies and evidences are pointing at the multitude of errors that continue to occur in the pre-analytical phase, it is in sharp contrast to the fact that there is lack of attention to extra-laboratory factors even as of now.

A consensus was done by a technical Committee of the International Organization for Standardization (ISO/TC 212, 2008) on the interpretation of mistakes in testing processes in laboratory. As it promotes and puts a lot of emphasis on the requirement for proper evaluation of all the procedures involved in the process of testing in laboratory, it is indeed very important.[6]

Errors in the Pre-analytical Phase

In accordance to a study done by Lippi *et al.*, (2011), it was noted that the pre-analytical errors accounted for about70% of all the errors in the laboratory diagnostics, consisting mainly of patient preparation mistakes, specimen collection errors, delayed transportation, and errors in preparation for analysis and storage.[7]

In a study done by Zaninotto *et al.,* (2012), it is stated that sample transportation needs to be considered as well, as it was identified as one of the source of pre-analytical errors that is commonly ignored and needs to be improved as there is an increasing trend towards a need for sample transport over long distances with the flourishing of laboratory facilities.[8]

Quality Indicators (QIs) in the pre-analytic Phase

For a better understanding, the pre-analytical phase should be subdivided into a 'pre-pre-analytical phase' and a 'true' pre-analytical phase, which can be done within the laboratory walls after sample has been received. The initial phase, consisting of procedures like, test requisition, and patient sample collection and its identification which consists of procedures which usually are not performed in the laboratory or by laboratory personnel. [9]

The second phase involves samples preparation for analysis (centrifugation, and sorting). In a patient-centred setup, QIs should be designed as such, so as to include every step of the pre-analytical phase, including proper selection of tests, which is very important in studies and to ensure proper clinical response as shown in Table 1.[10]

Materials And Methods

A retrospective comparison was done using data from the laboratory records for a duration of 14 months – 2 phases of 7 months each -, i.e., April - October 2016 (7 months) and April - October 2017 (7 months) in the clinical biochemistry section of a tertiary care hospital. During these periods there was preparation for the process of accreditation by the National Accreditation Board of Laboratories (NABL). The personnel assigned with sample collection were trained by the technical support team of Becton Dickinson and company (BD). The vacutainers suppliers for the centralized laboratory conducted hands on training on the standard techniques for phlebotomy procedures. The topics covered under phlebotomy training included selection of veins, common sites for phlebotomy, inappropriate sites for vein puncture, tourniquet application, cleaning the site, performing the draw of blood and end of draw. At the sample accession end, the lab personnel were trained thoroughly regarding the sample rejection criteria. The choice of appropriate colour coding of vacutainers tubes and transport of the specimen was also explained.

The clinical biochemistry lab is equipped with the VITROS 5600 auto-analyzer (Ortho-Clinical Diagnostics, Inc., Rochester, NY, USA). Phlebotomies for the inpatients are performed by the respective clinical department staff, whereas outpatient samples are collected at a centralized collection centre by lab staff. The data collection procedure involves reviewing of blood samples received from the inpatient and outpatient units. The samples collection is done using evacuated tubes (Becton Dickinson vacationers). All the specimens collected from their respective units are then delivered to the lab by the concerned paramedical staff.

In our laboratory, routine biochemistry testing is provided. Upon receiving the specimens, the lab supervisor visually tries to detect any error. The rejection criteria for the blood

Table 1: Quality indicators in the pre-analytic phase (10)

QI-1: Appropriateness of test request	of requests with clinical question (%)
QI-2: Appropriateness of test request	of appropriate tests with respect to the clinical question (%)
QI-3: Examination requisition	of requests without physician's identification (%)
QI-4: Examination requisition	of unintelligible requests (%)
QI-5: Identification	of requests with erroneous patient identification (%)
QI-6: Identification	of requests with erroneous identification of physician (%)
QI-7: Test request	of requests with errors concerning test input (%)
QI-8: Samples	of samples lost'not received (%)
QI-9: Samples	of samples collected in inappropriate containers (%)
QI-10: Samples	of samples haemolysed (haematology, chemistry) (%)
QI-11: Samples	of samples clotted (haematology', chemistry') (%)
QI-12: Samples	of samples with insufficient volumes (%)
QI-13: Samples	of samples with madequate sample-anticoagulant ratio (%)
QI-14: Samples	of samples damaged in transport (%)
QI-15: Samples	of improperly labelled samples (%)
QI-16: Samples	of improperly stored samples (%)
Adapted from Sciacovelli et al., 2009) [10]	

samples were as follows: 1) Incorrect volume 2) Patient misidentification, 3) Inappropriate tube, 4) After centrifugation haemolysis (visual haemolysis) and further on the run of the sample if the HI index is found to be> or = 15%) [11] and 5) Lipemic samples. Entries are made in the rejected sample log book when an error is come across. There is a weekly review of the data that is generated. The data from the specified durations, before as well as after the training was obtained retrospectively from the recorded log books in the biochemistry central laboratory and analysed for this study.

After the samples were allowed to clot for 30 min, the samples were then centrifuged in the lab. The samples are then analysed for routine clinical chemistry analytes using Vitros 5600 auto analyser that automatically measured HI using a spectrophotometric technique in all blood samples. The samples were considered haemolysed at an HI ≥15 (equivalent to 0.15 g/L of free haemoglobin). This level was the lowest detectable value of haemolysis.

Data Analysis

Data entry was done in Microsoft excel sheet and data analysis was done using Epi-info Software version 3.4.3.To express the findings, we used descriptive statistics like percentage and frequencies

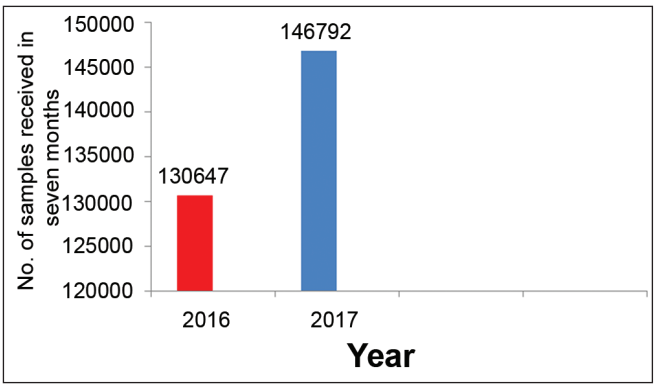

Fig. 1: Total number of samples received in the study periods

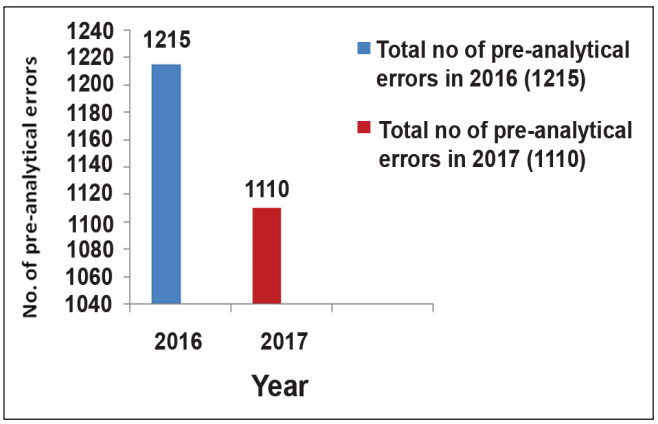

Fig. 2: Numbers of pre-analytical errors in 2016 and 2017

Table 2: Month wise distribution of samples received by clinical biochemistry during the study periods

S. No.	Month	Year→ No. of samples received (n) 2016	2017
1	APRIL	16,107	19,339
2	MAY	17,907	19,948
3	JUNE	20,303	18,224
4	JULY	20,299	22,635
5	AUGUST	20,295	22,239
6	SEPTEMBER	18,086	22,088
7	OCTOBER	17,650	22,318
	Total	1,30, 647	1,46,792
	Average/ month	18,664	20,970

Results

Total number of samples received by the laboratory for a period of 14 months was 2, 77, 438, i.e., (1, 30, 647 in 2016 and 1, 46, 791 in 2017) as shown in (Fig. 1).

The month-wise distribution of the samples collected in April – October, 2016 shows there were more samples from June to August (average 20, 299) as compared to those collected during the same months in 2017 which were more (average 22,320) between July and October (Table 2.)

Fig. 2 shows that the pre-analytical errors documented during the study period were 1,215 (0.93%) in 2016, and 1,110 (0.76%) in 2017. There was a marginal decrease in pre-analytical errors in 2017 as compared to 2016.

Table 3 and Fig. 3 shows that, the pre-analytical errors distribution was lesser in 2017 than in 2016 in all the months during the study periods.

Tables 4 and 5 depicts that hemolysed samples were the most both in 2016 and 2017 (Fig. 4) followed by quantity not sufficient, among all the types of pre-analytical errors.

On comparison of all the types of pre-analytical errors in the two years, it shows that hemolysed samples was the most frequent pre-analytical error seen, while the other errors like quantity not sufficient, sample transport, handling and wrong tube were negligible. As expressed in (Fig. 5), there was a marginal decrease in all the errors in 2017 compared to 2016.

Table 6 indicates that the total number of samples received were significantly higher in 2017 ($p<0.05$) than in 2016. Most of the pre-analytical errors documented in this study were non-significant ($p>0.05$) between the two study phases except for sample transport, which showed was significantly different ($p<0.05$) between 2016 and 2017.

Table 3: Number of pre-analytical errors documented during the two study periods

S. No.	Year →	2016		2017	
	Month	No. of samples received (n)	Pre-analytical errors n (%)	No. of samples received (n)	Pre-analytical errors n (%)
1	APR	16,107	207 (1.30)	19,339	168 (0.86)
2	MAY	17,907	187 (1.04)	19,948	163(0.81)
3	JUN	20,303	169 (0.80)	18,224	159 (0.87)
4	JUL	20,299	171 (0.84)	22,636	158 (0.69)
5	AUG	20,295	196 (0.96)	22,239	156 (0.70)
6	SEP	18,086	216 (1.19)	22,088	155 (0.70)
7	OCT	17,650	141 (0.79)	22,318	151 (0.67)
	Total	1,30, 647	1,215 (0.93)	1,46,792	1,110 (0.76)
	Average pre-analytical errors/month		174		159

The data presented is frequency with percentage in parenthesis.

Table 4: Distribution of the errors documented under various categories in 2016 (7 months)

Sl. No.	Month 2016	No. of samples received	Pre- Analytical Errors (n=1215)				
			Hemolysed n (%)	Quantity Not Sufficient n (%)	Sample transport n (%)	Specimen handling n (%)	Collection in wrong tube n (%)
1	APR	16,107	145 (0.90)	20(0.12)	19(0.12)	15(0.09)	8(0.04)
2	MAY	17,907	138 (0.77)	17(0.09)	15(0.08)	12(0.06)	5(0.02)
3	JUN	20,303	128 (0.63)	17(0.08)	13(0.06)	6(0.03)	5(0.02)
4	JUL	20,299	111 (0.55)	14(0.07)	11(0.05)	4(0.01)	6(0.02)
5	AUG	20,295	119 (0.60)	13(0.06)	12(0.05)	3(0.01)	9(0.04)
6	SEP	18,086	162 (0.89)	10(0.06)	11(0.06)	20(0.11)	6(0.03)
7	OCT	17,650	108 (0.61)	6(0.03)	5(0.02)	12(0.06)	10(0.05)
	TOTAL	**1,30,647**	**911 (0.69)**	**97 (0.07)**	**86 (0.07)**	**72 (0.05)**	**49 (0.04)**

The data presented is frequency with percentage in parenthesis.

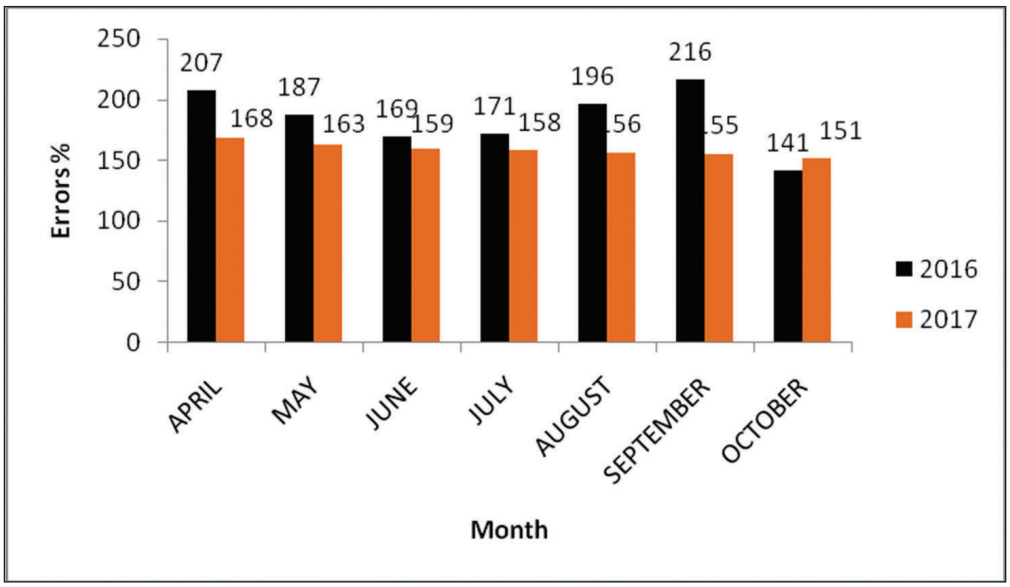

Fig. 3: Comparison of pre-analytical errors in 2016 and 2017

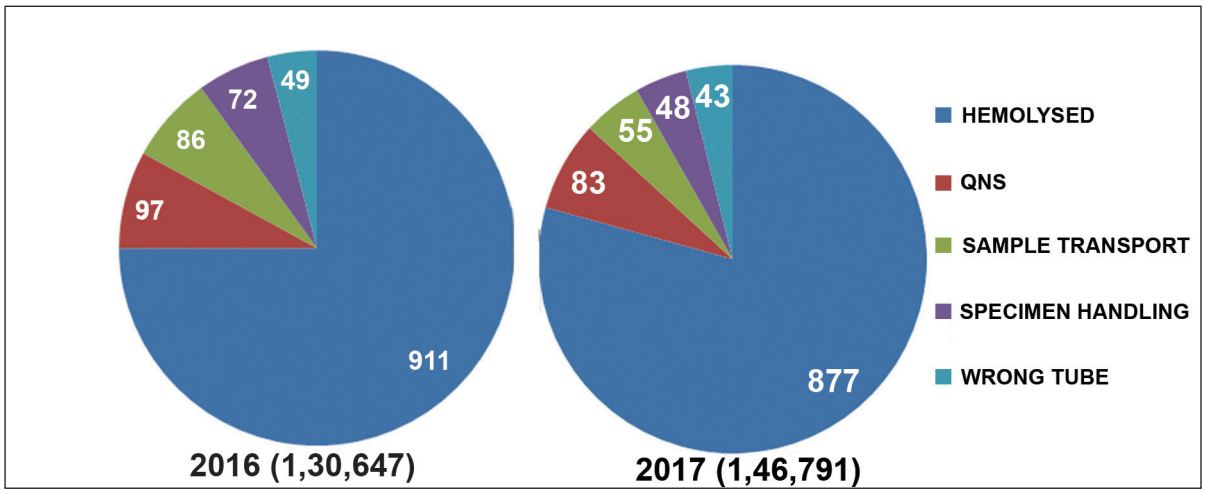

Fig. 4: Comparison of the pre-analytical errors under all categories of errors between 2016 and 2017

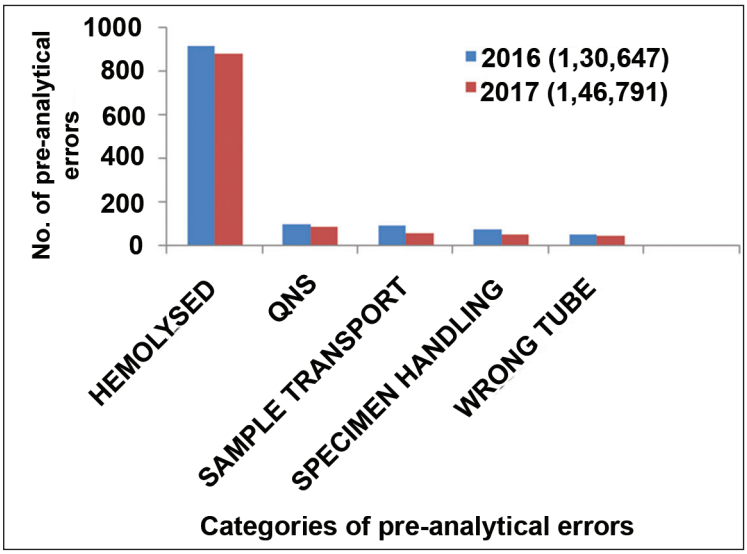

Fig. 5: Comparison of the pre-analytical errors under all categories of errors between 2016 and 2017

Table 5: Distribution of the errors documented under various categories in 2017 (7 months)

Sl. No.	Month 2017	No. of samples received	Pre- Analytical Errors (n=1215)				
			Hemolysed n (%)	Quantity Not Sufficient n (%)	Sample transport n (%)	Specimen handling n (%)	Collection in wrong tube n (%)
1	APR	19,339	131 (0.70)	14(0.07)	10(0.05)	8(0.04)	5(0.02)
2	MAY	19,948	127 (0.63)	13(0.06)	9(0.04)	8(0.04)	6(0.03)
3	JUN	18,224	127 (0.69)	12(0.06)	8(0.04)	6(0.03)	6(0.03)
4	JUL	22,636	125 (0.55)	11(0.05)	7(0.05)	7(0.03)	7(0.03)
5	AUG	22,239	125 (0.56)	10(0.04)	6(0.05)	6(0.02)	6(0.04)
6	SEP	22,088	122 (0.55)	11(0.04)	8(0.06)	6(0.03)	8(0.04)
7	OCT	22,318	120 (0.54)	12(0.05)	7(0.03)	7(0.03)	5(0.02)
8	TOTAL	1,46,791	877 (0.69)	83 (0.07)	55 (0.07)	48 (0.05)	43 (0.04)

The data presented is frequency with percentage in parenthesis.

Table 6: Comparison of the pre-analytical errors between the 2016 and 2017

Sl. No.	Parameters	2016 (n=7)	2017 (n=7)	T	p
1.	No. of samples received	18663.86 ± 1657.97	20970.29 ± 1765.32	–2.52	<0.05*
2.	Hemolysed	130.14 ± 19.51	125.29 ± 3.59	0.648	0.529
3.	Quantity Not Sufficient	13.86 ± 4.74	11.86 ± 1.35	1.074	0.304
4.	Sample transport	12.29 ± 4.27	7.86 ± 1.35	2.617	<0.05*
5.	Specimen handling	10.29 ± 6.24	6.86 ± 0.9	1.439	0.176
6.	Collection in wrong tube	7.00 ± 2.00	6.14 ± 1.07	1.00	0.337

Data is expressed as mean ± standard deviation of the number of samples over a duration of 7 months. n=7 months. Statistical test used:'t' test. Level of significance: '*' - $p<0.05$ was considered significant.

Discussion

With all the innumerable advances in science and technology and the recent innovations, laboratory diagnostics has been altered from labouring and inconvenient testing methods to fully automated science that has helped achieve accuracy. However, the lab working and testing is dependent on the cooperation of other departments, mainly clinical, for properly filled requisition forms and samples for analysis. According to a lot of studies, just by ensuring accuracy in the analytical phase of the testing process in a clinical laboratory, accuracy in results cannot be achieved. Pre- analytical (the phases before the sample reaches the lab) and post–analytical (the phase after the sample is analyzed) phases are both equally important.

In accreditation standards and, also considerations of laboratory quality expertise, there has been an increase in the recognition of the importance of the pre and post (extra-analytical) phases in laboratory medicine. In May 2010, a meeting was organised of 40 medical laboratory opinion leaders, in May 2010, to discuss issues and challenges faced by laboratory medicine.[12]

One group was given the responsibility of assessment of risk and control of error sources in the laboratory. These groups followed two recently published CLSI guidelines for risk management concerning extra-analytical issues and examined two specific questions in this area.[13,14] These questions were:-

1. "Proper examination of all the factors processes or conditions in the total testing process that contribute to risk of harm to the patient?"The group, using CLSI guidelines on non-conforming laboratory events, identified the following activities: ordering of test, sample collection, labelling/patient identification,

transport, handling, and quality of sample (pre-analytical phase); and result interpretation (including calculation errors), data entry, and results communication (post-analytical phase).[15]The most problematic area in the risk management was tackling the human factors.

2. "It is a known fact that even a single wrong result can compromise not only the lab credibility but patient health as well. Are there any special precautions that can be implemented to manage or eliminate such risks?" The solution lies in gaining cooperation of all stakeholders, simplification and proper standardization of processes, using relevant technology as best as we can, monitoring of all the steps and activities in the TTP continually in order to gain improvement in quality.

Laboratory medicine has been recognized by lab authorities as a very complicated process and its proper management is required to minimize the risk of occurrence of errors. The Swiss cheese model of error propagation can best illustrate the management to minimize error.[16,17] The error minimizing processes can be considered same as a stack of slices of Swiss cheese, where the holes represent opportunities for the passage of an error to the next level in the system. Each slice can prevent the error from passing to next layer and act as a defensive layer through the system. The vulnerability of the system is denoted by the number and efficacy of these defensive layers.[18] For laboratory medicine, the slices represent areas such as equipment maintenance, correct training, proper supervision and quality assurance procedures. It is important to block the gaps in order to minimize the likelihood of error. We have to ensure to minimize both the human and the systemic factors to avoid errors.

In the preanalytical phase we come across many shortcomings ranging from carelessness in attitude about filling the requisition slips to the lack of proper education of staff's regarding ideal phlebotomy procedures. Scientific understanding to reduce the errors must be applied diligently in this phase by the health care system.[19] All this is imperative in order to curb the dent on laboratory procedures that occur due to human errors. The error rate information within the lab testing procedure is very diverse (0.1% to 9.3%). According to a study by Plebani and Carraro, (2006), the majority of laboratory errors arise from problems in the pre or post analytical phases.[3]

In the present study, the majority of rejections in samples were due to haemolysis. The use of vacuum tubes along with the closed system of blood collection has made sample collection easy and efficient. Lack of training for staff engaged in phlebotomy is responsible for errors in collection and transport of samples. Most common causes of haemolysis are: - blood forced through a fine needle, shaking the tubes vigorously, and sample centrifugation before clotting is complete.[20] This can be avoided by taking precautions like; to allow the anticoagulant to mix with the blood properly, vacutainers for plasma should be gently inverted a few times and red top vacationers without anticoagulant should not be shaken after sample collection. Massive haemolysis can be caused by freezing and thawing of blood specimens.

In a study by Jay *et al.*, (2008), it was observed that the majority of hemolyzed samples (>95%) were due to mistakes resulting from incorrect sampling procedure or transportation.[21] Hemolysis causes exuding of intracellular contents into the plasma, giving false high values of potassium and enzymes such as SGOT and LDH. It causes prolongation of turn-around-time (TAT) as fresh samples are needed for processing the request.[22]

The root cause analysis of the retrospective data identified these pre-analytical errors in their descending order of occurrence: Hemolysis - 77%, Insufficient quantity of sample (QNS) -8%, Errors during sample transport - 6%, Errors during specimen handling - 5%, Wrong tube collection - 4%. The corrective action undertaken to minimize the errors are as follows:

1. Regular in-house training sessions for the technicians and nurses were carried out.
2. Three sessions of phlebotomy training, once every month since 2017 by professional corporate personnel.
3. Standard operating procedures (SOPs) were developed for the different steps - which helped in the incorporation of ideal phlebotomy practices.
4. The bar-coding has significantly reduced errors like incorrect identification / improper labelling.

It is ensured that the sample is transported to the lab and the sample centrifuged at the earliest. Temperature fluctuations resulting from transport delay can causes instability of temperature-dependent analytes. Examples of temperature-sensitive diagnostic analytes include arterial blood gas (ABG) parameters, lactate, ammonia and acid phosphatase. The majority of rejections in our study were due to haemolysis, maybe due to one or more of the following: using a syringe instead of a vacuum system, drawing of blood before the disinfectant dries, or application of tourniquet for one minute.

Hemolysis is one of the most common causes of pre-analytical errors, causing considerable harm to the accuracy of analytical tests. Analytes, such as potassium, alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine, and creatine kinase (CK), are typically overestimated and analytes, such as albumin, alkaline phosphatase (ALP), chloride, γ-glutamyltransferase (GGT), glucose and sodium, are reduced when haemolytic samples are used.

Hemolysis can be prevented by: allowing alcohol to dry before collection, using an appropriate bore needle, gentle mixing of samples, avoiding syringe collection, and avoiding collection from IV line.

Conclusion

This comparative study revealed that the NABL accreditation process has benefitted in identifying and minimizing the pre-analytical variables to an appreciable extent. However all the stakeholders involved in maintaining the quality have to remain vigilant at all times as "quality is a continual process."

Limitations

Most of the pre–analytical errors documented in this study were non- significant. So this study has not benefitted much in minimizing the pre-analytical errors, as the change was not significant.

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Conflict of Interest: None.

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