Analysis Of Completeness Of Blood Vacuume Tube Filling At A Clinical Laboratory Attached To Tertiary Care Center, A Cross Sectional Study

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Abstract: Background: Overfilled and under filled tube are most important cause of pre-analytical error and failure to complete requested examination in sample. The study was performed to analyse relationship between time of receiving of sample, location of sample, type of vacuum tube, method of collection, total volume required in a patient and volume incompleteness. Material And Methods: The study involved collection and patient samples data, i.e. receiving time of sample, place of collection, type of vacuum tube, blood collection method, total volume required in a patient, measurement of volume before processing. Average incompleteness of vacuum tube was calculated and average relationship between incompleteness and these parameters done.Result: Average incompleteness in OPD samples is better than Non-OPD samples. Average incompleteness is better during 9 am to 12 am than early morning hours. Incompleteness with relation to vacuum tube type is in general Plain > (EDTA/Fluoride) > Citrate. Average incompleteness better in sample collected with vacuum tube needle with holder than use of syringe. Average incompleteness increases as total volume required in a patient increases. Conclusion: Dedicated phlebotomist and use of vacuum tube needle with holder is required to bring overall improvement in completeness of blood collection. [Patel C Natl J Integr Res Med, 2022; 13(3): 10-14, Published on Dated: 10/05/2022]

Key Words: Volume incompleteness, Vacuum tube, OPD, Non-OPD

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Introduction: Measurement of blood volume is very important part of pre analytical error because overfilled and under filled tube are most important cause of pre-analytical error and failure to complete requested examination in sample as well as to provide accuracy in examination. Modern laboratory instrumentation uses very little amounts of blood, serum or plasma (i.e. typically between 2 and 20 µL per test), yet a minimum volume is required for processing samples automatically. Maintaining an optimum, sample-to-additive ratio is important for effective anti-coagulation and accurate laboratory tests. A key step in the sample handling process is ensuring that the blood draw sample volume is at least 90 percent of the stated volume on the collection tube.

In citrate vacuum tube if less blood is collected (e.g.1:7), then there is a significant increase in the a PTT results as compared to those which are obtained with the 1:9 ratio[1]. The effect of the less blood to citrate is lesser on PT and it becomes meaningful only when anticoagulant/blood ratio reaches 1:4.5 i.e. just less than half of its nominal volume. Under-filling of EDTA vacuum tube is also common problem and it leads to excess EDTA. This excess EDTA reduces PCV and affect other haematological parameters. All salts of EDTA are hyperosmolar, which causes water to leave the cells, resulting in cell shrinkage. This shrinking effect is noticed only with K3EDTA, not with K2EDTA and Na2EDTA[2]. This is because although the cells shrink with K2EDTA and Na2EDTA, the lower pH of the anticoagulant causes the cells to swell as well, thus balancing for the osmotically dependent shrinkage and the cell size is not altered. The higher the concentration of EDTA, the greater the osmotic withdrawal of water from the cells , leading to a reduction in PCV. This discrepancy will lead to a reduction in MCV and increase in MCHC[3]. Excess EDTA will increase chelation of magnesium and zinc and affect reagent enzymes used for signal generation like Alkaline phosphatase (ALP)[4].

The correct volume of sample is important considerations in the estimation of plasma glucose concentration of patients because if recommended volume is changed, glycolytic inhibition may not be optimal. Overfilling and under-filling (2.0 ± 1.0 ml) of fluoride-oxalate tubes did not affect the plasma glucose results significantly[5]. Hemolysis occur when vacuum

sample containers are not completely filled. Although the hemolysis may not be apparent to the naked eye, LDH activity can be significantly elevated by such exposure to vacuum. The most important factor is the relative ratio of the sample volume to the specified draw volume of the vacuum tube[6]. This study is aimed at analysis of variations in incompletely filled vacuum tube in relation to time and place of collections, type of tube, and method of collection and volume requirements.

Material & Methods: This study is planned for patients of New Civil Hospital, Goverment medical college Surat between April-2021 to october-2021. Blood samples received in plain, EDTA, Citrate and fluoride vacuum tube to the Clinical Biochemistry Laboratory and sample collected at OPD common collection center, New Civil Hospital, Surat are included in this study.

All samples examined were collected from patients that had been referred the laboratories for various clinical chemistry assays and therefore the study did not involve collection of any additional blood specimens and they are devoid of patient identification.

The study involved collection and patient samples data, i.e. receiving time of sample, place of collection, type of vaccum tube, blood collection method, total volume required in a patient, measurement of volume before processing.

Modification in LIS software was done to enter sample volume of each sample collected in vacuum tube. All technician were instructed to enter sample volume in each sample entry.

Venous blood samples were collected using 2 method - open collection method wirh syringe and needle and closed collection method using vacuum tube needle with holder. This involved usage of BD Vacuume tube multi sample needles, BD Vacuume tube holder and BD Vacuum serum tubes, normal hypodermic needle (22G) and disposables syringes (2ml,5ml,10ml).

The specimen was mixed by complete inversions for 8-10 times immediately after the draw. The needle was discarded into sharps container, while the (re-usable) BD Vacuum holder was re-used. Specimens thus collected were kept at room temperature for 60 min before centrifugation. The unopened tubes were centrifuged after ensuring balancing, using REMI PR-23 centrifuge at 3000 RPM for 10 minutes. After centrifugation, sample volume was measured by comparing all blood sample tube visually with appropriately marked comparative tube to find its volume and enter it in LIS.

Total 10663 samples were measured for volume incompleteness, out of them 9620 samples were collected by using syringe and needle and 1043 samples were collected by using Vacuum tube needle with holder. The data collected were analysed in spreadsheet
software.

Data Analysis: All data were exported from LIS into Microsoft excel file. For each vacuum tube dummy ID has been given. For each vacuum tube MRD number, Date and time of receiving, type of vacuum tube, method of collection, Department, Location were exported from LIS. After that for each vacuum tube designated volume was entered by formula.

Calculation Of Completeness Of Vacuum Tube Was Done By Formula:

$$\text{Completeness Of Vacuum Tube} = \frac{\text{Sample Volume}}{\text{Designated Volume}}$$

Calculation Of Incompleteness Of Vacuum Tube Was Done By Formula:

Incompleteness Of Vacuum Tube = ABS (1 – Completeness Of Vacuum Tube)

All data were sorted according to time of receiving of sample, location of sample tube, vacuum tube type and method of collection.

Average of incompleteness of vacuum tube and total number of sample were calculated using formula in micro-soft excel for each hour for total 24 hour, for each location, for vacuum tube type and for method of collection using formula in micro-soft excel.

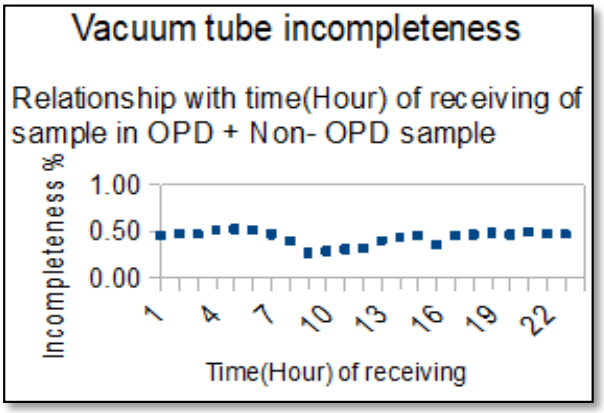
For Analysis Of Relationship Total Volume Required In A Patient And Volume Incompleteness:

Total Volume Required = Sum Of Designated Volume Of All Tubes

In which samples are collected from same patient at given time. It was calculated manually for each patient.

Results And Discussion: In month of April-2021 to October-2021, total 10663 sample were analyzed for volume incompleteness. Relationship of average incompleteness with time of receiving, place of collection, type of vacuum tube, method of collection and total volume required in a patient were done.

Figure 1: The Scatter Plot Of Average Incompleteness In Relation With Time Of Receiving Of Sample In OPD + Non-OPD Sample



Average incompleteness varies from 0.27 to 0.50. Maximum incompleteness is 0.52 and it is during early morning period between 4 am to 6pm. Minimum incompleteness is 0.27 and it is during 9 am to 12 am. As during morning hour between 9 am to 1 pm and evening hour 4 pm to 6 pm, there were samples from OPD as well as from wards. So, low incompleteness during morning hour may be due to better collection of OPD samples. To understand, data analysis of OPD and Non-OPD sample was done separately.

In Non-OPD samples, maximum incompleteness is 0.52 and is during early morning period 4 am to 6 am and minimum incompleteness is 0.36 and is during 10 am to 12 am. Thus lesser incompleteness is seen in OPD as well as non-OPD samples during morning hours, despite higher workload.

Thus, better completeness during morning half may be related to some other factors. On informal inquiry it was found that, early morning samples are collected by nursing staff while samples during 10-13 hours are collected by interns and residents. In OPD sample average incompleteness is good in-comparison with Non-OPD sample.

It varies from 0.17 to 0.37. Minimum incompleteness is seen in evening OPD during 4 pm to 5 pm. Maximum incompleteness is also seen in evening OPD during 5 pm to 6 pm. In morning OPD overall incompleteness is same and is around 0.25. Increased incompleteness during last hour of sample collection in OPD (i.e 5-6 PM) is notable and may be due to haste by Phlebotomist during end of duty. During all OPD hours, average incompleteness in OPD

Samples are better than Non-OPD samples. This may be due to sample collection by Phlebotomist in OPD as compared to sample collection in Wards by nursing staff, intern and resident doctors. Phlebotomist has lesser incompleteness as compared to ward staff probably due to daily practice and single task. Doctors and nursing staff have many distractions as they need to take care of patients other needs in addition to sample collection.

Table 1: Average Incompleteness And Number Of Sample In OPD And Non-OPD Sample

Place Of Collection	Average Incompleteness	Number Of Sample
OPD	0.26	2887
Non -OPD	0.42	7776

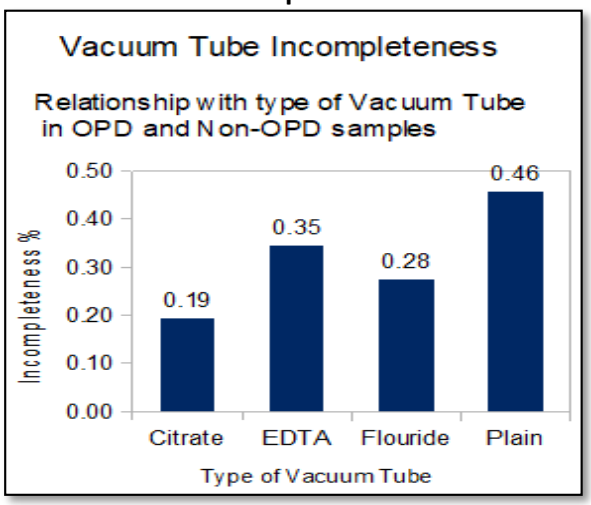
Hospital has different wards for different department. Average incompleteness vary in different ward from 0.26 to 0.74. Maximum incompleteness is 0.74 and is seen in samples from stem cell hospital. Minimum incompleteness is 0.26 and is seen in samples from OPD. Not only OPD sample collection is better, there is more or less uniform lack of completeness across all wards.

Average incompleteness is better in OPD samples as compared to all wards. In a busy hospital there is always rush of patient as compared to Phlebotomist. Dedicated phlebotomist collected blood is more complete, probably due to single task and repeated training and evaluation.

Provision of additional dedicated phlebotomist in ward can improve completeness of blood sample volume collection. However, they require additional human resources, which is limitation of a public funded government hospital. There is relative deficiency of technicians in OPD collection center due to heavy OPD in public free-

of-charge hospital, pressurizing technicians to complete all sample collections in fixed limited period. Increasing OPD sample collection staff can further improve OPD sample collection completeness.

Figure 2: Bar Diagram Of Average Incompleteness With Type Of Vacuum Tube In OPD And Non-OPD Samples

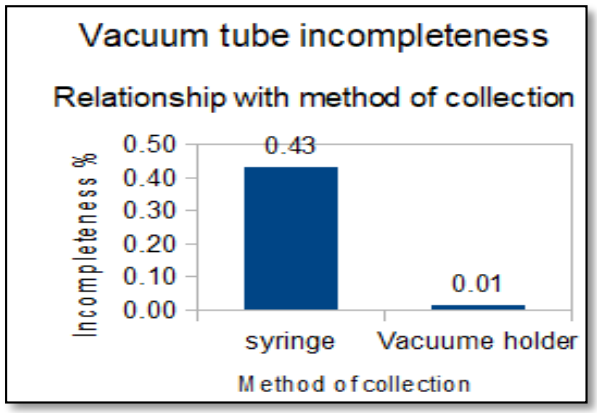


Average incompleteness in different vacuum tube varies from 0.19 to 0.46. Maximum incompleteness is 0.46 and is seen in plain vacuum tube. Minimum incompleteness is 0.19 and is seen in citrate vacuum tube, followed by fluoride tube. In OPD samples average incompleteness is minimum in fluoride vacuum tube and maximum in EDTA vacuum tube.

In Non-OPD samples, average incompleteness in minimum in citrate vacuum tube and is 0.19 and maximum incompleteness is in plain vacuum tube and is 0.49. Incompleteness with relation to vacuum tube type is in general Plain > (EDTA/Fluoride) > Citrate. Citrate tube requires perfect collection in order to give reliable results of coagulation related examinations. Due to these strict requirements, all samples collected are likely to be collected more carefully as compared to other tubes. Moreover, amount of volume required in EDTA(3 ml) and fluoride(2 ml) is less than that required in Plain tube(4 ml).

Rationing of sample collected in syringe may be responsible for observed pattern of incompleteness. Better completeness of citrate tube shows that, awareness of need to complete filling of blood sample collection tube improves compliance of phlebotomist. Training phlebotomist to explain that even in EDTA, Plain and Fluoride tube, incompleteness is associated with wrong results can improve their compliance to complete tube filling.

Figure 3: Bar Diagram Of Average Incompleteness In Different Method Of Blood Collection In OPD And Non-OPD Sample

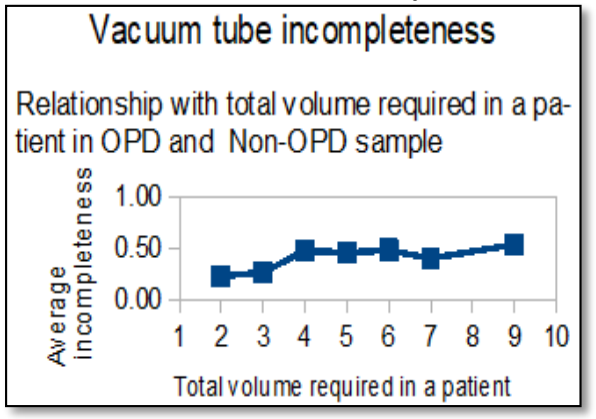


Average incompleteness better in sample collected with vacuum tube needle with holder and is 0.01. In OPD sample also average incompleteness is better in sample collected with vacuum tube needle with holder than syringe.

Although, vacuum tube needle with holder is not usually used for sample collection by most staff of the hospital, in the study, about 1000 samples were collected by vacuum tube needle with holder. Drastic improvement in incompleteness show that faulty practice of using syringes (instead of vacuum tube needle with holder) is responsible for incomplete filling of tubes. Use of vacuum tube needle with holder (instead of syringe) improves blood collection tube filling.

Improving availability of vacuum tube needle with holders and specialized needles and training blood collection staff in its technique will require further financial resources to the hospital.

Figure 4: Scatter Plot Of Average Incompleteness In Relation With Total Volume Required In A Patient In OPD And Non-OPD Sample



Average incompleteness increases as total volume required in a patient increases. Average incompleteness is minimum if total volume required is 2 ml and 3 ml and is 0.23 and 0.27 respectively. Average incompleteness is maximum when total volume required in a patient is 9 ml and is 0.54. Characteristically there is better collection of sample when required volume is 7 ml instead of 4-6ml. For sample collection up to 6 ml, 5 ml syringe is used. When sample requirement jumps to 7, 10 ml syringe is used. Thus, tube filling is better at 7 ml requirement as compared to 6 ml or 9 ml requirement.

Figure 5: Fish Bone Diagrams for Various Factors Responsible For Blood Sample Volume Incompleteness In This Study

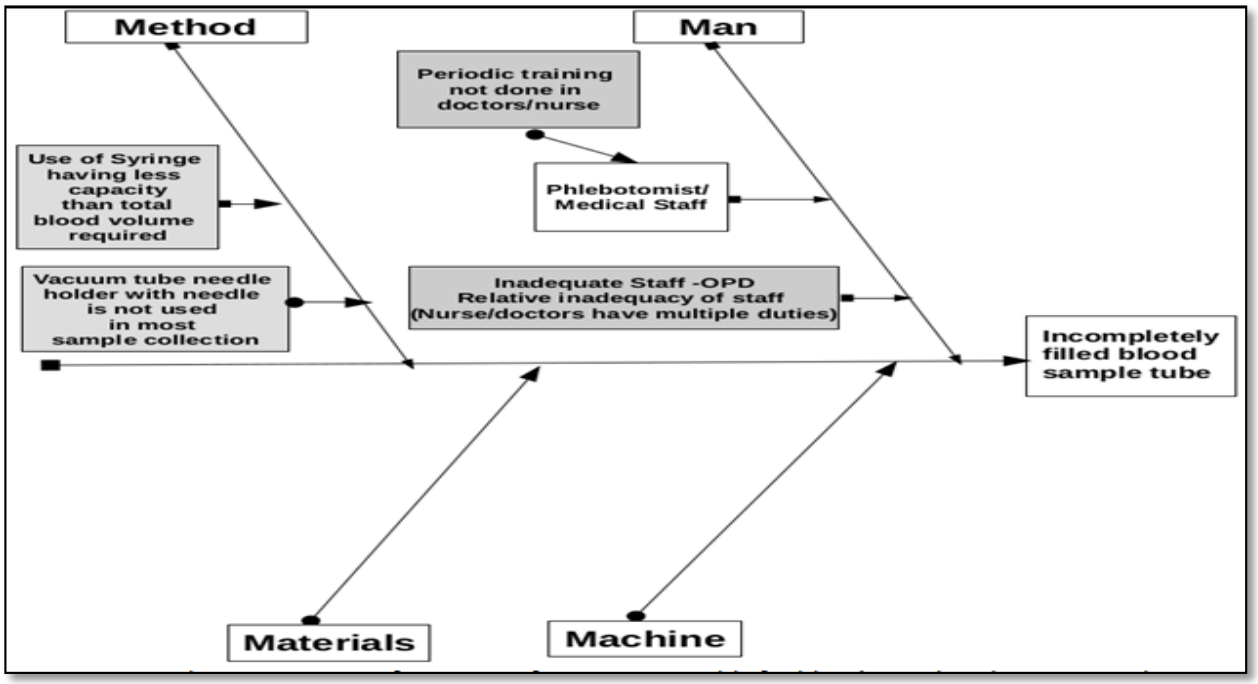


Figure 5 shows Fish Bone Diagrams for various factors responsible for blood sample volume incompleteness in this study. This diagram can be starting point for implementing corrective actions to improve vacuum tube completeness. Based on Figure 5, Training staff (Man), increasing human resources (Man) and changing blood collection system from syringe to vacuum tube with needle (Method) can improve blood sample collection completeness.

Conclusion: Dedicated phlebotomist and use of vacuum tube needle with holder is required to bring overall improvement in completeness of blood collection.

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