

## Impact of Calibration of pipette on Quality Control Results

Ketan K Mangukiya<sup>1,\*</sup>, Mittal Panchal<sup>2</sup>

<sup>1,2\*</sup>Assistant Professor, Department of Biochemistry,  
Parul Institute of Medical Science and Research(PIMSR), Vadodara, Gujarat

**\*Correspondence Author:**

E-mail: dr.ketan.mangukiya@gmail.com

### Abstract

**Background:** Majority of biochemical investigations requires pipette for delivery of reagents and samples in clinical laboratory so that pipette that is used for the same must be properly calibrated for accurate results.

**Aims:** The aim of this study to investigate that the impact of automatic pipette calibration on quality control results.

**Methodology:** Total 6 pipettes are used in our study. Among them 3 pipettes are with volume 1.0 ml and another 3 are with volume 0.1 ml, all pipettes were examined for work calibration. Distilled water weighing method is used for calculation of accuracy and precision of pipettes. Control sera of glucose and uric acid is used for determining accuracy and Precision of pipettes.

**Result & Conclusion:** Uncalibrated pipette had high effect on accuracy of control data as well as patients results therefore it is recommended that all pipette that is used in clinical laboratory must be periodically checked for calibration.

**Keywords:** Accuracy, Calibration, Gujarat, Pipette, Precision.

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### Introduction

Pipettes are necessary equipment often used in chemical laboratories to measure and transfer specific Volume of liquid. Pipettes are essentially narrow tube like equipment with a rubber bulb at the top. The tube is graduated from the top to the bottom mostly in increments of ten millimeters.[1] Prior to discussing pipette calibration it is important to understand accuracy and precision. Inaccuracy can be expressed as the deviation of the mean of a number of sample replicated from a set point volume and is expressed in either absolute units such as microliters, or relative units such as percent. Precision is expressed as the standard deviation (STD) of the number sample replicates and is expressed as the coefficient of variation (CV) of samples volume replicates. ISO standards recommend 10 readings.[2]

Under a constant temperature and atmospheric pressure, the density of distilled water is constant. The volume of water can be determined by weighing dispensed water. The calibration of pipette is carried out by gravimetric method. When determining the volume of water, the accuracy of measurements is effected by ambient temperature, atmospheric pressure and relative humidity. These factors are usually combined to give the Z factor, used in calculation of

volume of water. Then the calculated volume of water is compared with the theoretical volume to determine the accuracy and precision of the pipette[3].

### Methodology

This Study is conducted at biochemistry department of parul institute of medical science and research and attached parul sevashram hospital, Vadodara, Gujarat, India. 3 pipette of volume 1 ml and 3 pipette of volume 0.1 ml is and Randox normal control sera was exercised in this experiment and glucose and serum uric acid were estimated in by using that pipette in semi-automated microlab 300 biochemistry analyser.

Glucose is estimated by using GOD-POD method and serum uric acid is estimated by using uricase endpoint method.

**Method of pipette calibration:** Distilled water weighing method is used for pipette calibration. Procedure to check calibration: The pipette is checked with the maximum volume (nominal volume), the minimum volume or 10% of the maximum volume, whichever is higher. For example, pipette 0.5-10 $\mu$ l is tested at 10 $\mu$ l and 1 $\mu$ l.

Pipetteing of distilled water is done and weighed in single pan balance and the result is noted. This procedure is done consecutively for ten times with each volume and results obtained are noted. Mean, SD and CV is calculated by using Microsoft office excel by using prizm software. If the calculated results are within the selected limits, the calibration of the pipette is correct. The precision and accuracy was estimated for all automatic pipettes before and after calibration. The

precession and accuracy for glucose and uric acid were estimated in normal control sera by using Clinical and Laboratory Standards Institute (CLSI) document.

## Results

Results of Accuracy and precision of pipettes that is before and after calibration is mentioned below tables.(Table 1,2,3)

**Table 1: precision and accuracy for automatic pipette with volume (1 ml and 0.1 ml) before and after calibration**

Pipette Number	1000 microliter Accuracy		1000 microliter Precision (SD)		100 microliter Accuracy		100 microliter Precision (SD)	
	Before	After	Before	After	Before	After	Before	After
1	3.6	8.2*	18.2*	3.2	0.5	0.56*	1.87*	0.20
2	4.1	6.2*	2.3*	2.1	0.5	0.66*	0.85*	0.30
3	4.9	5.6*	4.1*	2.3	0.5	0.74*	1.4*	0.19

\* Significant difference between and calibration

Before - Before calibration

After - After calibration

Precession and accuracy of uric acid and glucose before and after calibration in normal control sera compared with total allowable error (TAE) for each test in tables below.

TAE for uric acid:  $\pm 17\%$

TAE for Glucose:  $\pm 5\%$

**Table 2: showing accuracy and precision status of blood glucose before and after calibration**

Pipette Number	Accuracy		Precision (SD)	
	Before	After	Before	After
1	1.5	37.2	0.55	0.42
2	1.6	24.6	0.58	0.19
3	1.5	18.6	0.45	0.33

Pipette Number	Accuracy		Precision (SD)	
	Before	After	Before	After
1	10.1	21.3	4.87	1.23
2	5.6	19.5	2.85	0.89
3	4.5	18.6	4.69	1.36

## Discussion

Calibration of pipette is very important thing for accurate result of control data as well as patients because same pipette is used for both Quality control data as well as patients sample measurement. Inaccuracy can be expressed as the deviation from the mean of a number of sample replicates from a set point volume and is expressed in either absolute units such as microliters, or relative units such as percent. Imprecision is expressed as the standard deviation (STD) of the number sample replicates and is expressed as the coefficient of variation (CV) of samples volume replicates.[4,5]

Poor precision and accuracy affect the quality control results. Precision and accuracy were calculated for each pipette, the accuracy, standard deviation and CV failed pre calibration and became less than standard

value that shown above. After that serum uric acid, and glucose were estimated in normal control sera 20 times before and after calibration for precision and accuracy then compared with standard precision and total allowable error for each test and found that results were affected after calibration. And before calibration, the results were not reliable and silent failure occurred.[6]

## Conclusion and Recommendation

In this study had been found that all pipettes performed outside established specifications for precision and accuracy, yet the operators were unaware that silent failures were occurred and had not taken these mal functioning pipettes out of service. The study shown that Inaccuracy and imprecision of automatic pipette had high effect on quality control results. Since pipettes are subject to silent and random failures and

have a higher rate of failure than many other laboratory instruments, the most important aspect of pipette quality control is a calibration frequency that ensures sufficiently high reliability. And the calibration must be done every three months and also verification for new automatic pipettes.

**Source of Support: Nil**

**Conflict of Interest: None**

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