Understanding Measurement: A Guide to Error, Contamination and Carryover in Volumetric Labware, Syringes and Pipettes

Volumetric measurement is a common repeated daily activity in most analytical laboratories. Many processes in the laboratory from sample preparation to standards calculation depend on accurate and contamination free volumetric measurements. Unfortunately, laboratory volumetric labware, syringes and pipettes are one of the most common sources of contamination, carryover and error in the laboratory.

The root of these errors is based on the four 'I' errors of volumetrics: 1. Improper Use; 2. Incorrect Choice; 3. Inadequate Cleaning; 4. Infrequent Calibration. These four I's can lead to error and contamination which negate all intent of careful measurement processes.

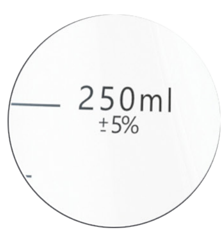
The first two I's stand for improper use, meaning that the volumetric is not used correctly, and incorrect choice. Many errors can be avoided by understanding the markings displayed on the volumetrics and choosing the proper tool for the job. There

a lot of information displayed on volumetric labware. Most labware, especially glassware, is designated as either Class A or Class B labware. Class A glassware is a higher quality analytical class of glassware, whereas Class B glassware is a lower quality glassware with a larger uncertainty and tolerance. If a critical measurement process is needed, then only Class A glassware should be used for measurement.

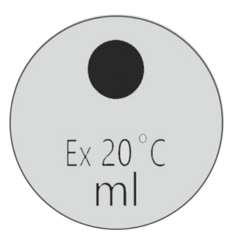
Other information which can be found on labware is the name of the manufacturer, country of origin, tolerance or uncertainty of the measurement of the labware, and a series of descriptors as to how the glassware should be used. Labware can be marked with letters which designate the purpose of the container. If a volumetric is designed to contain liquid, it will be marked by either the letters TC or IN. Labware which is designated to deliver liquid will be marked by either the letters TD or EX. Sometimes there are additional designations such as wait time or delivery time inscribed on the labware. The delivery time refers to period of time required for the meniscus to flow from the upper volume mark to reach the lower volume mark. The wait time refers to the time needed for the meniscus to come to rest after the residual liquid has finished flowing down from the wall of the pipette or vessel.

Manufacturer, Country, Certifications

Nominal Value

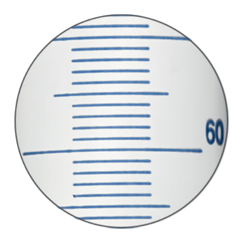


Tolerance



EX/IN To Deliver & To Contain

Precision Class
• Class A - Highest Quality
• Class B - Qualitative



Etchings, Graduations

A second type of improper use and incorrect choice can be seen in the selection of pipettes and syringes for analytical measurements. Many syringe manufacturers recommend a minimum dispensing volume of approximately 10% of the total volume of the syringe or pipette. A study by SPEX CertiPrep showed that dispensing such a small percentage of the syringe's total volume created a large amount of error. In this study, four syringes 10 µL, 25 µL, 100 µL, and 1,000 µL were used to dispense between 8-100% of the syringe's total volume of water. Each volume was weighed and replicated ten times by several analysts and the results were averaged together to calculate average error.

The largest rates of error were seen in the smaller syringes of 10 µL and 25 µL. Dispensing 20% of the 10 µL syringe created 23% error. Error only dropped down to below 5% as the volume dispensed approached 100%. In the larger syringes, measurements over 25% were able to see error in and around 1%. The larger syringes were able to get closer to the 10% manufacturer's dispensing minimum without a large amount of error, but the error did drop as the dispensed volume approached 100% (see Table 1).

Syringe Size	µL Dispensed	% of Syringe Volume	% Error
10	2	20%	23.15%
10	5	50%	8.16%
10	10	100%	2.72%
25	2	8%	8.82%
25	5	20%	5.47%
25	10	40%	2.37%
25	20	80%	1.05%
25	25	100%	1.25%
100	10	10%	6.09%
100	25	25%	1.67%
100	50	50%	0.64%
100	100	100%	0.61%
1,000	250	25%	1.05%
1,000	500	50%	1.14%
1,000	1,000	100%	0.47%

The third 'I' of volumetric error is inadequate cleaning. Many volumetrics can be subject to memory effects and carryover. In critical laboratory experiments labware sometimes needs to be separated by purpose and use. Labware subject to high levels of organic compounds or persistent inorganic compounds can develop chemical interactions and memory effects. It is also sometimes difficult to eliminate carryover from labware and syringes even when using a manufacturer's stated instructions. For example, many syringes are cleaned by several repeated solvent rinses prior to use. A study of syringe carryover by SPEX CertiPrep showed that some syringes are subject to high levels of chemical carryover despite repeated rinses.

In this study, several syringes, ranging in volume from 10 µL to 1,000 µL, were used to dispense a 2,000 µg/mL internal standard mix of deuterated compounds. The subsequent washes were collected and tested by GC/MS to determine the amount of carryover in each wash (see Table 2).

Table 2. Syringe Carryover Study (ppm of carryover observed)

Injection Wash #	1,000 µL	10 µL
1	24.4	360.51
2	0.69	46.96
3	0.06	8.12
4	0.03	4.1
5	0.04	2.02
7	0.01	1.37
10	0.03	1.37
15	0.02	1.13
20	0.01	0.43

The larger syringes needed less rinses to reduce carryover than the smaller 10 µL syringes. The smaller syringes had more than 1 ppm carryover through over 15 rinses. The typical amount of rinses usually employed to rinse syringes is between 3 and 5 rinses in which case, the smaller syringe would not be adequate to clear all the carryover from the syringe.

The final source of error is infrequent calibration. Many laboratories have schedules of maintenance for equipment such as balances and automatic pipettes, but often overlook calibration of reusable burettes, pipettes, syringes, and labware. Under most normal use, labware often does not need frequent calibration but there are some instances where a schedule of recalibration should be employed. Any glassware or labware in continuous use for years should be checked for calibration. Glass manufacturers suggest that any glassware used or cleaned at high temperatures, used for corrosive chemicals or autoclaved should be recalibrated more frequently. It is also suggested that under normal conditions soda-lime glass be checked or recalibrated every five years, and borosilicate glass be checked or recalibrated after it has been in use for ten years. The error associated with the use of volumetrics can be greatly reduced by choosing the correct volumetric for the task, using the tool properly, and by making sure the volumetrics are properly cleaned and calibrated before use.