

SOP for Pipette Quick Checks on Laboratory Balances

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Scope

The scope of this Standard Operating Procedure (SOP) is to describe how to verify and check pipettes to ensure that dispensing is carried out with the intended accuracy. This means determining the difference between the dispensed volume and the selected volume.

Material

To perform a quick check, the following materials are required: Pipette, pipette tips (recommended by the manufacturer), appropriate and calibrated laboratory balance, evaporation trap (or suitable weighing container), test liquid (deionized water), thermometer, barometer, gloves, work sheet or software.

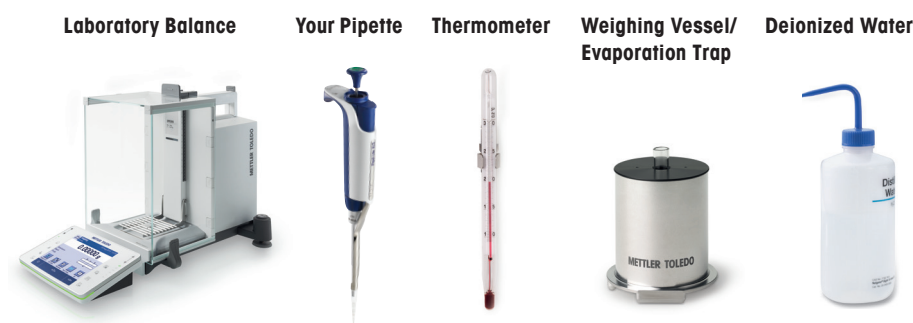


Fig. 1: A quick check requires minimal equipment

The ISO 8655 standard is stringent when it comes to the minimum requirements for the weighing equipment to be used, which, for practical purposes, depends on the nominal volume of the pipette being tested.

Pipette Dispensing Volume	Balance Resolution (mg)	Repeatability and Linearity (mg) ¹⁾	Standard Uncertainty Measurement (mg)	Evaporation Trap ²⁾
1 µL–10 µL	0.001	0.002	0.002	recommended
10 µL–100 µL	0.01	0.02	0.02	recommended
100 µL–1,000 µL	0.1	0.2	0.2	optional
1 mL–10 mL	0.1	0.2	0.2	optional
10 mL–200 mL	1	2	2	optional

¹⁾ If the standard uncertainty measurement of a balance is known (e.g. from the balance calibration certificate), it may be used instead of repeatability and linearity. The standard uncertainty measurement shouldn't be more than three times the resolution.

²⁾ METTLER TOLEDO recommendation

Tab. 1: Minimum balance requirements for pipette performance testing (ISO8655-2:2002)

Frequency of Quick Checks, Test Volumes and Repetitions

The higher the risk of failure, the more frequently pipettes should undergo a quick check.

	Pipettes Used Daily	Pipettes Used Infrequently
Routine lab work	At least monthly	Every three months should suffice
Regulated industries	Check appropriate regulatory guidance to ensure results quality (ISO, FDA, ASTM, etc.)	Check appropriate regulatory guidance to ensure results quality (ISO, FDA, ASTM, etc.)

Tab. 2: Frequency of pipette quick checks

A pipette should be checked at least at 100% (nominal volume). Depending on the guideline followed, more volumes (10%, 50%) have to be tested.

Guideline	Repetition	Volumes
ISO	10×	100%, 50%, 10%
SOP	4×	100%, 10%
n/a	4×	100%

Standard Pipette Quick Check Procedure

Quick checks are generally performed in the following 10 steps:

1. With a prepared vessel on a tared balance, adjust the pipette to 100% of its nominal volume and load the manufacturer's recommended tip.
2. Pre-rinse the tip by aspirating and dispensing the set volume of deionized water three times.
3. Holding the pipette vertically, immerse the tip to the appropriate depth and aspirate the deionized water.
4. Carefully move the pipette over the vessel on the balance and dispense the water. Slide the tip up the side wall to remove any excess liquid.
5. To avoid the effects of evaporation, as soon as the balance stabilizes, record the mass of the water in milligrams.
6. Repeat the process 3 more times using the same tip, recording the mass each time.
7. Eject the tip after the final dispense.
8. Adjust the pipette down to 10% of its nominal volume. Pre-rinse a new tip three times, and repeat steps 3–7.
9. With the collected data, calculate the pipette's mean volume, accuracy (mean error) and precision (standard deviation); formulas, equations and Z-factors (Tab. 2), see below.
10. Compare results vs. specifications (Tab. 3). If the results don't match, the pipette might require servicing, or the operator may require further training in technique.

Formula, Z-Factor and Calculation

Formula to calculate the pipette's mean volume, accuracy (mean error) and precision (standard deviation):

Volume

Expressed in mL.

$$v_i = (w_i)Z$$

v_i = individual volumes
 w_i = individual weighings
 Z = Z-factor

Mean Volume

The mean weight result with correction for Z-factor. Expressed in μL .

$$\bar{v} = \frac{\sum_{i=1}^n v_i}{n}$$

$\bar{v}$ = mean volume
 n = number of weighings
 v_i = individual volumes

Temp °C	Actual Z-Factor	Temp °C	Actual Z-Factor	Temp °C	Actual Z-Factor	Temp °C	Actual Z-Factor
15.0	1.0020	19.0	1.0027	23.0	1.0035	27.0	1.0045
15.5	1.0020	19.5	1.0028	23.5	1.0036	27.5	1.0047
16.0	1.0021	20.0	1.0029	24.0	1.0038	28.0	1.0048
16.5	1.0022	20.5	1.0030	24.5	1.0039	28.5	1.0050
17.0	1.0023	21.0	1.0031	25.0	1.0040	29.0	1.0051
17.5	1.0024	21.5	1.0032	25.5	1.0041	29.5	1.0052
18.0	1.0025	22.0	1.0033	26.0	1.0043	30.0	1.0054
18.5	1.0026	22.5	1.0034	26.5	1.0044		

Tab. 3: Z-factor values ($\mu\text{L}/\text{mg}$) as a function of temperature, for distilled water at 1 atm

Accuracy

Mean error is the difference between the mean volume of actual measurements and the true value as specified by the volume setting of the pipette. Expressed in μL .

$$E = \bar{v} - v_s$$

E = mean error
 $\bar{v}$ = mean volume
 v_s = volume setting

Precision

Standard deviation quantifies the magnitude of scatter due to random error.

$$s = \sqrt{\frac{\sum_{i=1}^n (v_i - \bar{v})^2}{n-1}}$$

s = standard deviation
 n = number of weighings
 v_i = individual volumes
 $\bar{v}$ = mean volume

Tolerances, Maximum Permissible Errors (ISO 8655)

Maximum Error Limits for piston-operated pipettes are clearly defined in ISO 8655. The standard characterizes both the maximum permissible systematic error, as well as the maximum permissible random error limits for a device at specific volumes ranging from 1–10,000 μL . These errors are doubled for multichannel pipettes. Most pipette manufacturer's tolerance specifications fall well below these limits, but at low volume measurements, meeting ISO 8655 tolerances can be a challenge for many service providers.

Nominal volume μL	Maximum permissible systematic error $\pm \%$	$\pm \mu\text{L}^a$	Maximum permissible random error $\pm \%^b$	$\pm \mu\text{L}^c$
1	5.0	0.05	5.0	0.05
2	4.0	0.08	2.0	0.04
5	2.5	0.125	1.5	0.075
10	1.2	0.12	0.8	0.08
20	1.0	0.2	0.5	0.1
50	1.0	0.5	0.4	0.2
100	0.8	0.8	0.3 ^d	0.3 ^d
200	0.8	1.6	0.3 ^d	0.6 ^d
500	0.8	4.0	0.3	1.5
1,000	0.8	8.0	0.3	3.0
2,000	0.8	16	0.3	6.0
5,000	0.8	40	0.3	15.0
10,000	0.6	60	0.3	30.0

^a Expressed as the deviation of the mean of a tenfold measurement from the nominal or selected volume (see ISO 8655-6:2002, 8.4)

^b Expressed as the coefficient of variation of a tenfold measurement (see ISO 8655-6:2002, 8.5)

^c Expressed as the repeatability standard deviation of a tenfold measurement (see ISO 8655-6:2002, 8.5)

^d For piston pipettes of type D1 the maximum permissible error may be $\pm 0.4\%$

Tab. 4: Maximum permissible errors for Air Displacement and Certain Positive Displacement pipettes [ISO 8655-2:2002, p. 6]

If your results don't match the pipette's specification, your pipette needs service or calibration.