USERGUIDE No. AU045 | August 2012

Differences in the systematic and random errors between the Eppendorf Reference[®] pipette and other manufacturer pipettes

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Abstract

In an independent practical study, noticeable differences were demonstrated in the characteristics of the systematic and random error during pipetting operations between the Eppendorf Reference pipette and three other manufacturers' pipettes. For this purpose, three pipettes from each manufacturer were each calibrated under standardized

Introduction

Errors in pipetting can be caused by the pipette itself or the user. Furthermore, they may also be the result of poor quality pipette tips, special physical properties of the pipetted liquid and a changing physical environment [1]. The influence of the pipette itself mainly relates to the systematic error. This may reflect, for example, manufacturing tolerances or production errors. In addition, inadequate pipette maintenance and cleaning can also lead to a systematic error outside the specifications to be complied with [1]. In contrast to the systematic error, the random error is almost exclusively influenced by the user. It may, for example, result from a lack of experience in using a pipette of a specific manufacturer. Possible options available to the manufacturer conditions by two independent users. The resulting errors were compared with each other and with the corresponding manufacturer's specifications and the specifications according to EN ISO 8655. Findings revealed that overall, among all the tested pipettes, the Eppendorf Reference pipette showed the lowest systematic and random error.

to minimize the random error include, among other things, clearly defined stops for the piston and pipette handle and control button designs according to modern ergonomic principles [1].

In this study, the effects of the pipette tip, liquid and the environment on the pipetting results were minimized by a series of standardized procedures. It was, therefore, possible to focus on the influencing factors that are solely attributable to the manufacturer and the user. Based on this, differences in the systematic and random error between the Eppendorf Reference pipette and three other manufacturer pipettes were determined in an independent practical study.

Materials and Methods

Preparation

The study used adjustable air-cushion pipettes, each with a nominal volume of 10, 100 and 1000 μ L. The pipettes were new and were obtained as such directly from the respective manufacturer [2].

To increase the validity of the study, three pipettes of a nominal volume were used in each case. For each of the 36 pipettes in total, three volume settings were each tested: 100%, 50% and 10% of the nominal volume. The use of the respective manufacturers' tips, as recommended by the manufacturer for their pipette, made it possible to test each pipette together with the tip as a total system. Before use, the pipettes and tips were equilibrated with the

test room conditions for at least 2 hours.

Materials

- > Eppendorf Reference pipettes with manufacturer's tips
- > Pipettes from manufacturer B with manufacturer's tips
- > Pipettes from manufacturer F with manufacturer's tips
- > Pipettes from manufacturer G with manufacturer's tips
- > Mettler[®] balance model AX205 with a moisture trap
- $\,>\,$ Mettler® balance model MT5 with a moisture trap

Calibration

For the calibration of the pipettes, the method according to ASTM E1154 was followed in all details. This is also compliant with ISO 8655-6.

- 1. A moisture trap was placed on the balance and equilibrated for 1 hour. A vessel with a small amount of deionized water was placed in the moisture trap.
- 2. Before each measurement, the pipette tip was pre-wetted with the test liquid for 5 times.
- 3. A moisture test was performed by taring the balance under simulation of a sample delivery.
 A stopwatch was started and the weight was recorded after 20 ±5 seconds.
- 4. A total of 10 data points were recorded per volume.
- 5. At the end, another moisture test was performed.

All calibrations were performed in the laboratory of Artel Inc. [2]. The methods used comply with ISO 17025.

The calibrations were performed by two users with each user calibrating each pipette.

The standard deviation and the average delivered volume were calculated for each pipette. This was used to determine the systematic and random error.

Results and Discussion

In the following figures, the first 3 data points within one dataset (6 data points) illustrate the error of the corresponding pipettes no. 1 to no. 3 being used by user 1. The last 3 data points of the same dataset show the error of the same three pipettes in the same order but being used by user 2.

Random error

It was defined that a pipette was faulty if one and the same pipette had an error that lied outside its manufacturer's specifications or ISO specifications when used by both users. According to the above definition, the random error of all pipettes was within ISO 8655-2 specifications. Based on Figure 1, the massive errors of different pipettes from manufacturer B under user 2 are particularly striking. Nevertheless, the corresponding errors under user 1 are significantly lower. It can, therefore, be assumed that the high errors under user 2 are influenced by the user himself. They could be an indication of a lack of experience with pipettes from manufacturer B. In contrast to this finding, the mostly high errors under both users with pipettes from manufacturer F at 1 μ L should be questioned more critically. Since in this case both users generate relatively high random errors at a dispensing volume of 1 μ L, it seems that there is not only a lack of experience with this type of pipette, but also a deficiency in terms of the use and handling of this dispensing volume.

eppendorf



Pipettes with a nominal volume of 10 µL

Figure 1: Random error of the pipettes with a nominal volume of 10 µL.

At the dispensing volume of 1 μ L, it is possible to achieve the lowest overall random errors using pipettes from manufacturer G. At the same time, these errors are relatively close to each other. In absolute terms, the lowest random errors are achieved with the Eppendorf Reference pipette under user 1. However, the corresponding errors under user 2 are significantly higher in comparison to user 1. This was also observed at the volume setting of 5 μ L and 10 μ L. One reason for this could be due to a lack of experience in pipetting with the Eppendorf Reference pipette. The pipettes from manufacturer G at this volume setting, thus, proved to be the most precise among the tested pipettes, followed by the Eppendorf Reference pipettes.

At 5 μ L, the Eppendorf Reference pipette demonstrates the lowest overall random errors, followed by pipettes from manufacturers F and G. Especially under user 1, the volumes

delivered by the Eppendorf Reference pipettes vary little from one to another. Pipettes from manufacturer B also show low random errors under user 1. However, the errors are massive under user 2 so that their overall performance is the most imprecise.

At 10 µL, only the Eppendorf Reference pipettes and pipettes from manufacturer G enable both users to achieve consistently low errors. As with the previous volume setting, a pipette from manufacturer B also stands out at this volume setting because of a high error. Pipettes from manufacturer F also show two increased errors at this setting.

In summary, it can be said that the Eppendorf Reference pipettes, together with those from manufacturer G, were found to be overall the most precise among the pipettes with a nominal volume of 10 μ L.



Pipettes with a nominal volume of 100 μ L

Figure 2: Random error of the pipettes with a nominal volume of 100 µL.

As similarly seen in Figure 1, a pipette from manufacturer B also stands out in Figure 2 because of a high random error at 10 μ L. The errors with the two other pipettes from manufacturer B, in contrast, are low in the case of both users and about as large as with the other pipette models.

At 50 μ L, the results with the Eppendorf Reference pipettes and pipettes from manufacturer B under user 1 are the most precise. With pipettes from manufacturer F, in contrast, higher errors were achieved by both users. The highest errors in total were achieved by user 2 with pipettes from manufacturer G. Eppendorf Reference pipettes and those from manufacturer B, therefore, overall were found to be the most precise at 50 $\mu L.$

At 100 μ L, user 1 achieved the most precise results using the Eppendorf Reference pipettes. With 2 of 3 Eppendorf Reference pipettes, user 2 achieved an average error rate in comparison with the other pipettes. With one pipette, he achieved an increased error.

All in all, none of the pipettes with a nominal volume of 100 μL was overall clearly more precise than any of the other pipettes.



Pipettes with a nominal volume of 1000 µL

Figure 3: Random error of the pipettes with a nominal volume of 1000 μ L.

Referring to Figure 3, when dispensing 100 µL, the most precise results overall are achieved with the Eppendorf Reference pipettes, whereas both users generated significantly increased errors especially with pipettes from manufacturer B. Furthermore, the high errors achieved by both users with pipette no. 2 from manufacturer B indicate that this pipette's behavior is consistently imprecise, which can be attributed to the pipette itself.

Even at 500 μ L, the Eppendorf Reference pipettes showed the lowest overall random errors. The errors of all the other manufacturer pipettes were comparably high with both users.

At 1000 μ L, both users achieved the most precise volumes with Eppendorf Reference pipette no. 1. The other Eppendorf Reference pipettes showed a high level of precision, similar to that of pipettes from manufacturer B. In contrast, both users achieved relatively high random errors with pipettes from manufacturer F. Pipettes from manufacturer G lie in between. Therefore, among the pipettes with a nominal volume of 1000 μ L, the Eppendorf Reference pipette had the lowest random error overall.

Thus, it can be concluded that among the pipettes with a nominal volume of 1000 μL , the lowest random error can be achieved with the Eppendorf Reference pipette.

Systematic error

It was defined that a manufacturing error exists if one and the same pipette had an error that lay outside of its manufacturer's specifications or ISO specifications when used by both users.



Figure 4: Systematic error of the pipettes with a nominal volume of 10 µL.

The manufacturer's specifications for the allowed systematic error are identical for all tested pipettes with a nominal volume of 10 μ L (Figure 4). Among all the manufacturers, only the Eppendorf Reference pipette complies with its own specifications. Pipette no. 1 from manufacturer F showed high errors when used by both users at all three volumes and thus, overall is not in the standard range. The Eppendorf Reference pipettes, together with pipettes from manufacturer B, delivered overall the lowest errors at volume setting 1 μ L.

At 5 μ L, the accuracy of the pipettes from all manufacturers is similar. The volumes that are pipetted with the Eppendorf Reference pipettes under user 1 are closest to the set volume but the pipetted volumes under user 2 are significantly less

accurate. Comparable results on average were achieved with pipettes from manufacturers B, F and G with the exception of pipette no. 1 from manufacturer F.

At 10 μ L, the Eppendorf Reference pipettes and pipettes from manufacturer G are overall closest to the set volume.

The Eppendorf Reference pipette delivered the lowest overall systematic errors at every volume setting. At the same time, none of the three Eppendorf Reference pipettes was outside of the manufacturer's specifications when used by both users. Therefore, among the pipettes with a nominal volume of 10 μ L, the lowest systematic error can be achieved with the Eppendorf Reference pipettes.

Table 1: Number (\sum) of pipettes with a nominal volume of 10 µL from manufacturers »Eppendorf«, »B«, »F« and »G«, whose systematic error lies outside of the manufacturer's specifications or the ISO specifications at 10%, 50% and 100% of the nominal volume.

Volume setting	ISO 8655 specifications	Manufacturer's specifications	Eppendorf ∑	вΣ	FΣ	GΣ
1 μL	± 12.0%	± 2.5%	0	1*	1*	1*
5 μL	± 2.4%	± 1.5%	0	0	1*	0
10 µL	± 1.2%	± 1.0%	0	0	1*	0

* The same pipette



Pipettes with a nominal volume of 100 µL

Figure 5: Systematic error of the pipettes with a nominal volume of 100 μ L.

At the dispensing volume of 10 μ L, the volumes pipetted with pipettes from manufacturers B and F are overall closer to the set volume than others (Figure 5). However, in the case of manufacturer F, two of three pipettes have errors outside of the manufacturer's specifications and ISO specifications at 100 μ L. Furthermore, the significantly high errors under both users with pipette no. 2 from manufacturer B at 50 μ L and 100 μ L indicate user-independent and consistently inaccurate results at the specified dispensing volumes, which could be attributed to the manufacture or construction of this pipette. At 50 μ L and 100 μ L, the Eppendorf Reference pipettes delivered the overall most accurate results.

It can be summarized that among all the tested pipettes with a nominal volume of 100 μ L, the Eppendorf Reference pipette produces the lowest systematic error.

Volume ISO 8655		Eppendorf		В		F		G	
setting	specifications	specifications	Σ	specifications	Σ	specifications	Σ	specifications	Σ
10 µL	± 8.0%	± 3.0%	0	± 2.0%	0	± 3.0%	0	± 3.5%	0
50 µL	± 1.6%	± 1.0%	0	± 1.0%	0	± 1.2%	0	± 0.8%	0
100 µL	± 0.8%	± 0.8%	0	± 0.8%	1*	± 0.8%	2*	± 0.8%	0

Table 2: Number (\sum) of pipettes with a nominal volume of 100 µL from manufacturers »Eppendorf«, »B«, »F« and »G«, whose systematic error lies outside of the manufacturer's specifications or the ISO specifications at 10%, 50% and 100% of the nominal volume.

* The same pipette



Pipettes with a nominal volume of 1000 μ L

Figure 6: Systematic error of the pipettes with a nominal volume of 1000 μ L.

At 100 μ L, the volumes pipetted with pipettes from manufacturer B are closer to the set volume than other manufacturer pipettes but they show the highest systematic error at 500 μ L and 1000 μ L together with pipettes from manufacturers F (figure 6). Therefore, the pipettes from manufacturers B and F are overall the least accurate among the tested pipettes. For all volume settings, the pipettes from manufacturer F are significantly less accurate than those from manufacturer B. In addition, pipette no. 3 from manufacturer B had errors outside of the manufacturer's specification at 1000 μ L.

It should be noted that all three pipettes from manufacturer F lay outside of the manufacturer's specifications for all three volume settings. All the three pipettes also lay outside of the

ISO specifications at 1000 μ L. Such behavior is abnormal such that they exhibit a high degree of inaccuracy, which can be attributed to the manufacture or construction of these pipettes.

Pipettes from manufacturer G are closest to the set volume of 1000 μ L but deliver the least accurate results of all pipettes at 100 μ L. At 500 μ L, the Eppendorf Reference pipettes and those from manufacturers G have the lowest errors.

Given the high variability of inaccuracy observed among the tested pipettes with a nominal volume of 1000 μ L, it can be seen that overall the Eppendorf Reference pipettes produce the lowest systematic error.

Table 3: Number (Σ) of pipettes with a nominal volume of 1000 µL from manufacturers »Eppendorf«, »B«, »F« and »G«, whose systematic error lies outside of the manufacturer's specifications or the ISO specifications at 10%, 50% and 100% of the nominal volume.

Volume	ISO 8655	Eppendorf		В		F		G	
setting	specifications	specifications	Σ	specifications	Σ	specifications	Σ	specifications	Σ
100 µL	± 8.0%	± 3.0%	0	± 1.0%	0	± 1.0%	3*	± 3.0%	0
500 µL	± 1.6%	± 1.0%	0	± 0.6%	0	± 0.8%	3*	± 0.8%	0
1000 µL	± 0.8%	± 0.6%	0	± 0.6%	1*	± 0.6%	3*	± 0.8%	0

* The same pipette



Conclusion

In comparison with the tested pipettes from other manufacturers, the Eppendorf Reference pipette is the only one that complies with both its own specifications and the ISO specifications without exception. It also has the lowest systematic error. While no great differences were found among the pipettes with a nominal volume of 100 μ L, the Eppendorf Reference pipette delivered the lowest random error among the 1000 μ L pipettes. The Eppendorf Reference pipette, together with the pipette from manufacturer G, also offer the lowest random error among the pipettes with a nominal volume of 10 μ L.

References

[1] Ewald, K. Liquid Handling: Laboratory Practice. Munich: Verlag Moderne Industrie; 2005.
 [2] Report compiled by ARTEL (Corporate Headquarters) for Eppendorf AG, Hamburg, Germany.

Ordering information	ı			
Eppendorf Reference	e® (adjustable)			
Volume range	Volume	Systematic error	Random error	Order no.
Dark gray dispensing	g button for epT.I.P.S.® 10	uL pipette tips		
	0.25 μL	±12.0%	≤6.0%	
0.1–2.5 μL	1.25 μL	±2.5%	≤1.5%	4910 000.085
	2.5 μL	±1.4%	≤0.7%	
Gray dispensing but	ton for epT.I.P.S.® 20 µL pi	pette tips		
	1 µL	±2.5%	≤1.8%	
0.5–10 μL	5 µL	±1.5%	≤0.8%	4910 000.018
	10 µL	±1.0%	≤0.4%	
	2 µL	±3.0%	≤2.0%	
2–20 µL	10 µL	±1.0%	≤0.5%	4910 000.026
	20 µL	±0.8%	≤0.3%	
Yellow dispensing bu	utton for epT.I.P.S.® 200 μL	and 300 µL pipette tips		
	2 µL	±5.0%	≤1.5%	
2–20 µL	10 µL	±1.2%	≤0.6%	4910 000.034
	20 µL	±1.0%	≤0.3%	
	10 µL	±3.0%	≤0.7%	
10–100 µL	50 µL	±1.0%	≤0.3%	4910 000.042
	100 µL	±0.8%	≤0.15%	
	50 µL	±1.0%	≤0.3%	
50–200 μL	100 µL	±0.9%	≤0.3%	4910 000.093
	200 µL	±0.6%	≤0.2%	
Blue dispensing butto	on for epT.I.P.S.® 1,000 µL p	ipette tips		
	100 µL	±3.0%	≤0.3%	
100–1,000 μL	500 μL	±1.0%	≤0.2%	4910 000.069
	1,000 μL	±0.6%	≤0.2%	
Red dispensing butto	n for epT.I.P.S.® 2,500 µL p	pette tips		
	500 μL	±1.5%	≤0.3%	
500–2,500 μL	1,000 μL	±0.8%	≤0.2%	4910 000.077
	2,500 μL	±0.6%	≤0.2%	

The data for systematic and random errors only applies when using Eppendorf epT.I.P.S. pipette tips.

Ordering information

Eppendorf Reference® (fixed volume)						
Volume	Systematic error	Random error	Order no.			
Gray dispensing butto	on for epT.I.P.S.® 20 µL pipette tips					
1 µL	±2.5%	≤1.8%	4900 000.010			
2 µL	±2.0%	≤1.2%	4900 000.028			
5 µL	±1.5%	≤0.8%	4900 000.036			
10 µL	±1.0%	≤0.5%	4900 000.044			
Yellow dispensing but	ton for epT.I.P.S. [®] 200 μL pipette tips					
10 µL	±1.0%	≤0.5%	4900 000.109			
20 µL	±0.8%	≤0.3%	4900 000.117			
25 µL	±0.8%	≤0.3%	4900 000.150			
50 µL	±0.7%	≤0.3%	4900 000.125			
100 µL	±0.6%	≤0.2%	4900 000.133			
Blue dispensing butto	n for epT.I.P.S.® 1,000 µL pipette tips					
200 µL	±0.6%	≤0.2%	4900 000.508			
250 μL	±0.6%	≤0.2%	4900 000.540			
500 μL	±0.6%	≤0.2%	4900 000.516			
1,000 µL	±0.6%	≤0.2%	4900 000.524			
Red dispensing butto	n for epT.I.P.S. [®] 2,500 μL pipette tips					
1,500 µL	±0.6%	≤0.2%	4900 000.923			
2,000 µL	±0.6%	≤0.2%	4900 000.907			
2,500 µL	±0.6%	≤0.2%	4900 000.915			

The data for systematic and random errors only applies when using Eppendorf epT.I.P.S. pipette tips.

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