HandyStep® electronic
HandyStep®

Testing Instructions (SOP)

May 2009

1. Introduction

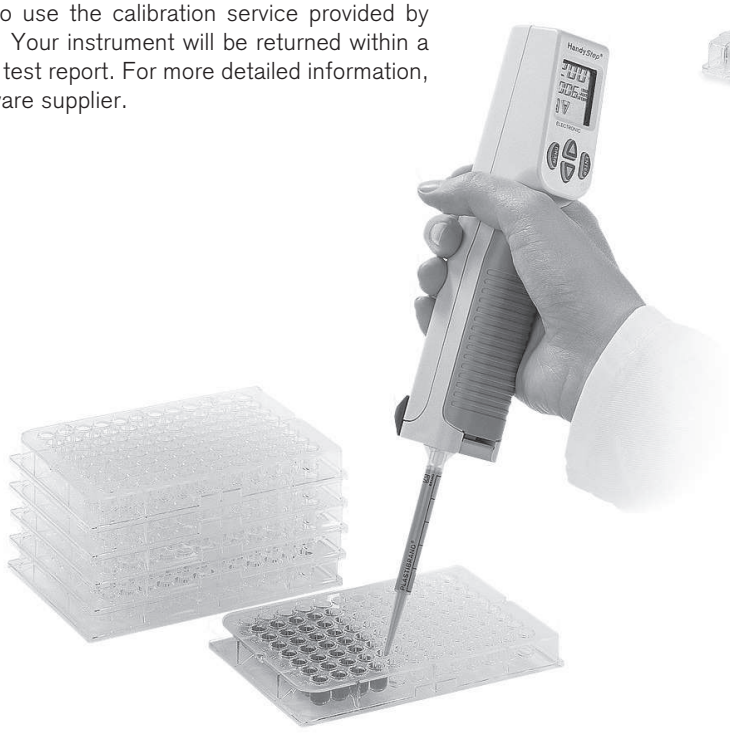
The standard DIN EN ISO 8655 describes both the design and the testing of the repetitive pipette HandyStep® electronic and HandyStep®. The following Testing Instructions describe how to apply the ISO standard in practice.

We recommend a testing of the repetitive pipette every 3-12 months. This interval may be adjusted to individual requirements. For example, when working very frequently or when using aggressive media, the instrument should be tested more frequently.

These Instructions may also be used as a basis for the monitoring of testing devices to DIN EN ISO 9001, DIN EN ISO 10012 and DIN EN ISO/IEC 17025.

Due to their PD-Tips, repetitive pipette allow the dispensing of long series in a stress-free and rapid way and with high precision. In combination with PLASTIBRAND® PD-Tips, the HandyStep® permits up to 49 pipetting steps, and the HandyStep® electronic up to 100 steps without refilling. The HandyStep® electronic features continuous volume adjustment and also allows the pipetting of liquids.

For the regular examinations required e.g. by DIN EN ISO 9001, DIN EN ISO 10012 and DIN EN ISO/IEC 17025 and the GLP Guidelines, you may also use the calibration service provided by BRAND (see chapter 7). Your instrument will be returned within a few days together with a test report. For more detailed information, please contact your labware supplier.



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2. Preparation for testing and visual examination

2.1 Type and serial number

- Read instrument type (embossed on the casing) ⇒ Enter number into Test Record see page 10 (1).
- Read PD-Tip size ⇒ Enter size into Test Record (1).
- Read customer's identification, if present ⇒ Enter identification into Test Record (1).

2.2 Minimal configuration of the HandyStep® electronic, resp. HandyStep®

- HandyStep® electronic, resp. HandyStep®
- PD-Tips ⇒ Use only appropriate dispenser tips. For best results, use original PLASTIBRAND® PD-Tips.

2.3 Cleaning

- Clean the casing adequately. ⇒ Wipe with a moist cloth (water or diluted soapy solution). Do not disassemble the instrument!
  ⇒ See Operating Manual for details.

2.4 Visual examination for damage

- Casing ⇒ Mechanical damage?
- PD-Tip ⇒ Scratches? Deformations? Mechanical damages?

Possible faults and corrective actions:

Problem	Possible cause	Corrective action
PD-Tip dripping	■ PD-Tip leaking	⇒ Replace PD-Tip
Important parts damaged	■ Mechanical or chemical damage	⇒ Send instrument in for repair

2.5 Functional test

3.2.1 HandyStep® electronic

- Mount new PD-Tip
- Then PD_Tipis automatically recognized and with compatible dispenser tips simply enter the tip size.
- Adjustment of volume to be dispensed
- Filling the PD-Tip ⇒ Immerse PD-Tip into the testing liquid approx. 2 - 3 mm. Aspirate liquid at a steady rate.
- Hold the HandyStep® electronic vertically for approx. 10 seconds and observe if a drop forms at the end of the tip. ⇒ If a drop forms, see table below.
- Release the liquid step by step. ⇒ Liquid should emerge at a steady rate.
- Empty the PD-Tip completely and remove. ⇒ Enter findings into Test Record (3).

Possible faults and corrective actions:

Problem	Possible cause	Corrective action
PD-Tip dripping	■ PD-tip leaking	⇒ Mount new PD-Tip
PD-Tip not recognized	■ Type encoding missing or damaged, or tip not mounted correctly	⇒ Mount new PD-Tip, or mount again. Select volume range if required.

Note:
If the instrument displays an error message, follow the Operating Manual.

2.5.2 HandyStep®

- Mount new PD-Tip
- Does the piston properly lock into place? ⇒ Operating key must move easily and smoothly.
- Does the volume adjustment work?
- Filling the PD-Tip ⇒ Immerse PD-Tip into the testing liquid and fill the tip. Operating key must move easily and smoothly.
- Hold the HandyStep® vertically for approx. 10 seconds and observe if a drop forms at the end of the tip. ⇒ If a drop forms, see table at the end of this section.
- Release the liquid step by step. ⇒ Liquid should emerge at a steady rate.
- Empty the PD-Tip completely and remove. ⇒ Enter findings into Test Record (3).

Possible faults and corrective actions:

Problem	Possible cause	Corrective action
PD-Tip cannot be inserted	■ Filling/locking key not in bottom position and not tilted upward. ■ Piston of the PD-Tip not inserted completely.	⇒ Push filling/locking key all the way to the bottom, and tilt upward. ⇒ Push piston of the PD-Tip into the cylinder completely.
Filling/locking key cannot be pushed upward	■ Filling/locking key not completely pushed in (closed).	⇒ Push piston of the PD-Tip into the dispenser completely. Close the filling/locking key.
PD-Tip dripping	■ PD-tip leaking	⇒ Replace the PD-Tip.

Note:
For additional examinations and adjustments, see the Operating Manual for HandyStep® and HandyStep® electronic.

3. Equipment required for testing

3.1 For HandyStep® electronic, resp. HandyStep®

- **Recipient vessel** filled with deionized water (e. g. Erlenmeyer flask) ⇒ Match temperature of room, water and instrument
- **Weighing vessel** filled with small amount of water (e. g. Erlenmeyer flask) ⇒ Bottom must at least be covered. In case of testing volumes < 100 µl, protect against evaporation.
- Required **accuracy of balance**: ⇒ Approx. 1/10th of the error limit of the instrument.

Testing volume	Balance display	Reproducibility and linearity
0.1 to 10µl	6-digit	± 0.002 mg
> 10 to 100µl	5-digit	± 0.02 mg
> 100 to 1000µl	4-digit	± 0.2 mg
> 1 to 10ml	4-digit	± 0.2 mg
> 10 to 50ml	3-digit	± 2 mg

- **Thermometer** with 0.2 °C accuracy
- Place the HandyStep® electronic, resp. the HandyStep®, including appropriate tips, into the testing room for at least 1 hour (unpacked). ⇒ Allow instrument to adjust to room temperature

Traceability of test results to national standards
Through the use of calibrated testing devices (balance and thermometer), the requirement of DIN EN ISO 9001, DIN EN ISO 10012 and DIN EN ISO/IEC 17025 to refer the test to the national standard is fulfilled. The calibration of the balance e.g. can be carried out either by DKD calibration or official certification of the balance, or by calibrating the balance with appropriate weights that are traced to the national standard. The calibration of the thermometer can also be carried out by DKD calibration or official certification, or by a comparison with thermometers that are traced to the national standard (under defined conditions).

4. Gravimetric test

4.1 HandyStep® electronic

This test can be carried out with PD-Tips of any size. However, size 5 ml is most commonly used for this purpose.

1. Set instrument to nominal volume.
2. Determine temperature of the deionized water.
   ⇒ Enter temperature into Test Record.
3. Filling the PD-Tip
   ⇒ Immerse PD-Tip vertically into the testing liquid by 2 - 3 mm, and press the step key to take up liquid. After taking in liquid, the HandyStep® electronic will automatically compensate for play to minimize surface tension inside the tip. A small amount of liquid is thereby discharged.
4. Place the weighing vessel (containing a small amount of deionized water) on the balance and tare.
5. Remove weighing vessel from the balance.
6. Dispense the first step into the weighing vessel.
   ⇒ Up to a volume of 5 ml, lean the PD-Tip against the wall of the vessel at an angle of about 45°, then wipe it off against the wall (approx. 10 mm). Volumes over 5 ml may be dispensed in a free jet.
7. Place the weighing vessel on the balance.
   ⇒ Enter weighed value into Test Record ((6.) V1)
8. Re-tare the balance.
9. Repeat steps 2 to 7 ten times.
   ⇒ Enter weighed values into Test Record ((6.) V_1)
10. Repeat the same testing procedure at 50 % and 10 % of nominal volume.
    ⇒ For weighings at 50 % (V_2) and 10 % (V_3) of nominal volume, there is no need to refill the HandyStep® electronic after each measurement, since volumes are dispensed in steps.
    ⇒ Enter all weighed values into the Test Record; a total of 30 values.

4.2 HandyStep®

This test can be carried out with PD-Tips of any size. However, size 5 ml is most commonly used for this purpose.

1. Set the HandyStep® to Step 5.
2. Determine temperature of the deionized water for testing.
   ⇒ Enter temperature into Test Record.
3. Filling the PD-Tip
   ⇒ Immerse PD-Tip vertically into the testing liquid by 2 - 3 mm.
4. Reject the first step; it only serves to compensate for play to minimize surface tension inside the tip.
5. Place the weighing vessel (containing a small amount of deionized water) on the balance and tare.
6. Remove weighing vessel from the balance.
7. Dispense the second step into the weighing vessel.
   ⇒ Up to a volume of 5 ml, lean the PD-Tip against the wall of the vessel at an angle of about 45°. Push the dispensing lever at a steady rate to the first stop, hold it there, and wipe the tip off against the wall (approx. 10 mm). Volumes over 5 ml may be dispensed in a free jet.
8. Place the weighing vessel on the balance.
   ⇒ Enter weighed value into Test Record ((6.) V_1)
9. Re-tare the balance.
10. Repeat steps 6 to 9 ten times.
    ⇒ Enter weighed values into Test Record ((6.) V_1); a total of 30 values.

    Note:
    At the Step 5 setting, the PD-Tip has to be filled again for dispensing the tenth step.

11. Repeat the same testing procedure at the settings Step 3 (V_2) and Step 1 (V_3).

5. Evaluation of gravimetric test results

The values obtained by weighing during the gravimetric test are only the mass values of dispensed volume. In order to obtain the actual volume, an adjustment calculation must be carried out. To facilitate your calculations and evaluations, we recommend the use of the Windows-compatible calibration software EASYCAL™ from BRAND.

The following calculations must be carried out:

1. Mean weight:
(Example for 10 weighing values)

$$\bar{x} = \frac{x_1 + x_2 + x_3 + x_4 + x_5 + x_6 + x_7 + x_8 + x_9 + x_{10}}{10}$$

2. Mean volume:

$$\bar{V} = \bar{x} \cdot Z$$

⇒ For factor Z, see Table 1.
⇒ Enter value into Test Record (7a).

3. Standard deviation:

$$s = Z \cdot \sqrt{\frac{(x_1 - \bar{x})^2 + (x_2 - \bar{x})^2 + (x_3 - \bar{x})^2 + (x_4 - \bar{x})^2 + \ldots + (x_{10} - \bar{x})^2}{9}}$$

⇒ For factor Z, see Table 1.
⇒ Enter value into Test Record (6b).

4. Accuracy:

$$A\,[\%] = \frac{\bar{V} - V_{\text{nominal value}}}{V_{\text{nominal value}}} \cdot 100$$

⇒ Enter value into Test Record (6c).

5. Coefficient of variation:

$$CV\,[\%] = \frac{s \cdot 100}{\bar{V}}$$

⇒ Enter value into Test Record (6d).

Comparison actual/nominal values:

- See tables of error limits and accuracy for each instrument on the following pages, or define your own error limits.

⇒ Enter value into Test Record (6e, f).

Result:

⇒ Enter value into Test Record (6g).

If the calculated values A [%] and CV [%] are smaller than or equal to the error limits, the instrument is in good working order.

If the calculated values **exceed** the error limits:

- Verify if all instructions in this Manual have been followed carefully.
- Observe the "Troubleshooting" information in the Operating Manual.

If these measures are not successful, we offer you the possibility to have your instruments calibrated by the BRAND Calibration Service (see page 12).

Table 1:

Excerpt from DIN EN ISO 8655, part 6.
Table refers to 1013 hPa
Validity range 980 hPa to 1040 hPa

Temperature °C	Factor Z ml/g	Temperature °C	Factor Z ml/g
15	1.0020	23	1.0035
15.5	1.0020	23.5	1.0036
16	1.0021	24	1.0038
16.5	1.0022	24.5	1.0039
17	1.0023	25	1.0040
17.5	1.0024	25.5	1.0041
18	1.0025	26	1.0043
18.5	1.0026	26.5	1.0044
19	1.0027	27	1.0045
19.5	1.0028	27.5	1.0047
20	1.0029	28	1.0048
20.5	1.0030	28.5	1.0050
21	1.0031	29	1.0051
21.5	1.0032	29.5	1.0052
22	1.0033	30	1.0054
22.5	1.0034		

Table 2:

Excerpt from DIN EN ISO 8655, part 5.

Nominal volume µl	1	2	3	10	20	50	100	200	500
A ±%	5	5	2.5	2	1.5	1.0	1	1	1
CV %	5	5	3.5	2.5	2.0	1.5	1.0	1.0	0.6
Nominal volume ml	1.0	2.0	5.0	10	25	50			
A ±%	1	0.8	0.6	0.5	0.5	0.5			
CV %	0.4	0.4	0.3	0.3	0.3	0.25			

Table 3:

Volumetric error limits for repetitive pipette:

The stated error limits refer to new instruments under optimized testing conditions (qualified operators and standardized ambience conditions).

Accuracy table HandyStep® electronic with PLASTIBRAND® PD-Tips

HandyStep® electronic with PD-Tip	Nominal volume 100%	50%	(A* ≤ ± %) 10%	Nominal volume 100%	50%	(CV* ≤ %) 10%
0.1 ml	1.0	1.2	1.6	0.5	1.0	2.0
0.5 ml	0.9	0.9	0.9	0.25	0.5	1
1.25 ml	0.6	0.6	0.9	0.15	0.3	0.6
2.5 ml	0.5	0.5	0.8	0.1	0.2	0.4
5.0 ml	0.5	0.5	0.8	0.08	0.15	0.3
12.5 ml	0.4	0.4	0.5	0.08	0.15	0.25
25.0 ml	0.3	0.3	0.3	0.08	0.15	0.25
50.0 ml	0.3	0.3	0.3	0.08	0.15	0.25

A* = Accuracy, CV* = Coefficient of variation

Measurements were performed with original PD-Tips by BRAND. According to ISO 8655, the volume which can be selected for each PD-Tip is relevant. The testing liquid was aqua dest.

Accuracy table HandyStep® with PLASTIBRAND® PD-Tips

HandyStep® with PD-Tip	Nominal volume 10% (Step 5)	6% (Step 3)	(A* ≤ ± %) 2% (Step 1)	Nominal volume 10% (Step 5)	6% (Step 3)	(CV* ≤ %) 2% (Step 1)
0.1 ml	1.6	2.7	8.0	2.0	3.0	5,0
0.5 ml	0.8	1.33	4.0	0.6	0.73	1.4
1.25 ml	0.8	1.33	4.0	0.3	0.38	0.8
2.5 ml	0.7	1.17	3.5	0.2	0.30	0.8
5.0 ml	0.5	0.83	2.5	0.2	0.27	0.6
12.5 ml	0.3	0.50	1.5	0.2	0.23	0.4
25.0 ml	0.3	0.50	1.5	0.2	0.23	0.4
50.0 ml	0.3	0.50	1.5	0.15	0.19	0.4

A* = Accuracy, CV* = Coefficient of variation

Measurements were performed with original PD-Tips by BRAND. According to ISO 8655, the volume which can be selected for each PD-Tip is relevant. The testing liquid was aqua dest.

For calibration, the error limits to be observed by the operator must be individually defined by the user. For this purpose, the following methods can be applied:

- If required by the application and if the optimized conditions for measuring are present, the stated error limits can also be expected in the case of used volumetric instruments in good working order.
- In analogy to the German regulations for official testing, it is also admissible to apply the limits which are typical for practice. These practice limits correspond to double the limits for official testing. In this case, the values found in Table 3 should be **doubled**.
- The user may also define his own individual tolerance limits corresponding to his particular application, and apply these error limits for the calibration of his instrument.

The above procedures fulfil the requirements of DIN EN ISO 9001, DIN EN ISO 10012 and DIN EN ISO/IEC 17025.

Test Record for Volumetric Instruments (EX)

1. Instrument:	❑ HandyStep® electronic	❑ HandyStep®	❑ _____
PD-Tip-size:	❑ 0.5 ml	❑ 1.25 ml	❑ 2.5 ml
	❑ 5.0 ml	❑ 12.5 ml	❑ 25.0 ml
	❑ 50.0 ml	❑ Other:	

Serial number: _____

Customer's identification: _____

2. Damage:

❑ None
❑ Type of damage: _____
❑ Damage repaired

3. Functional defects:

❑ None
❑ Type of functional defect: _____
❑ Functional defect repaired

4. Water temperature: _____ °C

Balance: _____

Thermometer: _____

Corrective factor Z: _____

Relative atmospheric humidity: 50 % ± 30 % (default value only)

5. Weighing values from gravimetric test

Weighing No.	**V_1 =**	**V_2 =**	**V_3 =**
x_1			
x_2			
x_3			
x_4			
x_5			
x_6			
x_7			
x_8			
x_9			
x_{10}			

6. Evaluation of gravimetric test

Procedure		**V_1 =**	**V_2 =**	**V_3 =**
a	$\bar{V}$			
b	s			
c	A [%] found			
d	CV [%] found			
e	A [%] nominal			
f	CV [%] nominal			
g	Result			

The testing was carried out according to DIN EN ISO 8655.

_____ _____

Date: **Signature:**

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