OMCL Network of the Council of Europe
QUALITY ASSURANCE DOCUMENT

PA/PH/OMCL (09) 64 2R

QUALIFICATION OF EQUIPMENT
ANNEX 6: QUALIFICATION OF PISTON PIPETTES

Full document title and reference	Qualification of Equipment Annex 6: Qualification of piston pipettes PA/PH/OMCL (09) 64 2R
Document type	Guideline
Legislative basis	-
Date of first adoption	19th May 2010
Date of original entry into force	1st July 2010
Date of entry into force of revised document	-
Previous titles/other references	-
Custodian Organisation	The present document was elaborated by the OMCL Network/EDQM of the Council of Europe
Concerned Network	GEON

ANNEX 6 OF THE OMCL NETWORK GUIDELINE
"QUALIFICATION OF EQUIPMENT"

QUALIFICATION OF PISTON PIPETTES

INTRODUCTION

The present document is the 6th Annex of the core document "Qualification of Equipment", and it should be used in combination with it when planning, performing and documenting the qualification process of piston pipettes.

The core document contains the Introduction and general forms for Level I and II of qualification, which are common to all type of instruments.

The present annex contains instrument-related recommendations on parameters to be checked at Level III and IV of qualification and the corresponding typical acceptance limits, as well as practical examples on the methodology that can be used to carry out these checks.

AIM AND SCOPE OF THE GUIDELINE

This guideline describes the requirements for piston pipettes used in chemical and biological tests irrespective of the mode of operation (manual or electronical).

Requirements and test procedures are based on the EN ISO 8655.

The following types of pipettes have been considered in this guideline:

1. Fixed volume monochannel pipettes (air-displacement (type A) and positive displacement or direct displacement pipettes (type D))

2. Variable volume monochannel pipettes (air-displacement (type A) and positive displacement or direct displacement pipettes (type D))

3. Fixed volume multichannel pipettes (air-displacement (type A))

4. Variable volume multichannel pipettes (air-displacement (type A))

NOTE: Repetitive piston pipettes are also covered in the guideline as "variable volume monochannel pipettes (positive displacement or direct displacement pipettes)".

CONSIDERATIONS FOR LEVEL I AND II OF EQUIPMENT QUALIFICATION

It is recommended, at Level I of the qualification of pipettes (Selection of instruments and suppliers) to select a manufacturer of pipettes that can certify its compliance with the requirements of EN ISO 8655.

It is recommended, at Level II of the qualification of pipettes (Installation and release for use), to check if all requirements set during the selection of the instrument and supplier are met by the pipette supplier and all necessary aspects are covered in the provided certificate.

FREQUENCY OF CALIBRATION

The recommended minimum calibration frequency is as follows:

- At reception (unless already calibrated by the supplier).
- Once a year during the use of the pipette.
- After any repair or adjustment.

GLOSSARY

The following terms and definitions are extracted from the EN ISO 8655-Part 1, chapter 3: Terms and definitions.

Systematic error *(accuracy)*

(piston-operated volumetric apparatus) Difference between the dispensed volume and the nominal volume or selected volume of the piston-operated volumetric apparatus.

Random error *(repeatability)*

(piston-operated volumetric apparatus) Scatter of the dispensed volumes around the mean of the dispensed volumes.

Nominal volume

(piston-operated volumetric apparatus) Volume specified by the manufacturer and used for identification and for indication of the measuring range.

NOTE: for a variable-volume piston-operated volumetric apparatus, the nominal volume corresponds to the maximum volume that can be set by the user and that is specified by the manufacturer.

Selected volume

(variable-volume volumetric apparatus) Volume set by the user, in order to dispense a volume chosen from the useful volume range of a variable-volume piston-operated volumetric apparatus.

NOTE: for a fixed-volume piston-operated volumetric apparatus, the selected volume is equal to the nominal volume.

Useful volume range

That part of the nominal volume which allows dispensing under observance of the maximum permissible errors as specified in the applicable part of ISO 8655.

NOTE: The upper limit of the useful volume range is always the nominal volume. The lower limit is 10 % of the nominal volume if not otherwise specified by the supplier.

Dead air volume

(piston-operated pipettes with air interface) Air volume between the lower part of the piston and the surface of the liquid.

Dead liquid volume

(positive displacement pipettes, burettes, dilutors and dispensers) Amount of liquid which does not belong to the dispensed volume and which is contained during operation in aspiration or expelling tubes, valves and within the cylinder.

Calibration

(piston-operated volumetric apparatus) Set of operations that establish the relationship between the dispensed volume and the corresponding nominal or selected volume of the apparatus.

NOTE 1: the result of a calibration permits the determination of correction values of the dispensed volume and its associated expanded uncertainty, e.g. following an adjustment or user adjustment.

NOTE 2: calibration requires no operation which permanently modifies the apparatus.

TABLE III

Level III. Periodic and motivated instrument checks

Examples of requirements for pipettes

1. FIXED AND VARIABLE VOLUME MONOCHANNEL PIPETTES

1a. Air-displacement pipettes (type A)

Parameter to be checked	Typical tolerance limits
action of the piston	smooth and positive
tip holder	no marks or distortion no liquid residues
leakage	no drop is formed at the tip
maximum permissible systematic error	see page 13
maximum permissible random error	see page 13

1b. Positive displacement or direct displacement pipettes (type D)

Parameter to be checked	Typical tolerance limits
action of the piston	smooth and positive
tip holder	no marks or distortion no liquid residues
leakage	no drop is formed at the tip
maximum permissible systematic error	see page 14
maximum permissible random error	see page 14

2. FIXED VOLUME AND VARIABLE VOLUME MULTICHANNEL PIPETTES

Air-displacement pipettes (type A)

Parameter to be checked	Typical tolerance limits
action of the piston	smooth and positive
tip holder	no marks or distortion no liquid residues
leakage	no drop is formed at the tip
maximum permissible systematic error	see page 15
maximum permissible random error	see page 15

TABLE IV

Level IV. In-use instrument checks

Examples of requirements for pipettes

1. ALL TYPES OF PIPETTES

Before every use, the pipette will undergo a short visual inspection (e.g. action of the piston, residue of liquids at the tip holder, generation of drops at the tip, etc.). If a malfunction is detected, the follow-up should be defined in an internal procedure. Appropriate measures would be checking and recalibration, repair or disposal.

The in-use check of the pipette should be appropriately documented (e.g. logbook, raw data etc.)

ANNEX I

Level III. Periodic and motivated instrument checks

This Annex contains practical examples of tests and their associated tolerance limits for several parameters related to the performance of pipettes.
These examples can be considered by the OMCLs as possible approaches to perform the Level III of the equipment qualification process: "Periodic and motivated instrument checks".

GENERAL CONSIDERATIONS

- The periodic calibration of pipettes should follow a pre-established calibration plan, in the frame of the Management System of the laboratory.

- Before setting the calibration plan, the laboratory should identify the pipettes for which calibration is needed, depending on the impact of their performance on the test results (e.g. volumetric pipettes).

- If calibration is carried out by an external company, the laboratory requesting calibration should verify and preferably choose a company that is accredited in accordance with ISO/IEC 17025 to perform calibrations in accordance with the specifications of EN ISO 8655.

- Range of calibration: taking into account factors such as the evaporation of water and the accuracy of the balance, the laboratory should make an evaluation and define the minimum nominal volume of pipettes from which the in-house calibration can provide reliable results. Lower volume pipettes should then be sent to an accredited external calibration company.

- If the calibration is performed by an external company there should be a calibration prior to service and recalibration. If the laboratory is working with infectious materials, the pipettes should be decontaminated before the calibration according to the instructions of the provider of the calibration service. In this case the risk that the fact of disassembling the pipettes can have an impact on their performance should be taken into account. A test before calibration would not be necessary as the results would not reflect the real working status of the pipettes before calibration.

- Tips used for the calibration of pipettes should be preferably the same as the tips used for the routine laboratory work.

- If applicable, the calibration of pipettes should be performed with the pipetting technique (direct mode or reverse mode) that is usually applied by the laboratory.

- If in-house customized Excel sheets are used for the calculation of the calibration results, please refer to Annex 1 of the OMCL Guideline on Validation of Computerized Systems: "Validation of computerised calculation systems. Example of validation of in-house software document" (PA/PH/OMCL (08) 87).

PERIODIC AND MOTIVATED INSTRUMENT CHECKS FOR ALL TYPES OF PIPETTES

1.1. VISUAL INSPECTION

Materials:

- Purified water (at least, distilled or deionised water)
- Tips appropriate for the tested pipette

Method:

- The piston is pressed up and down several times. The action of the piston must be smooth and positive.
- The tip holder is inspected carefully for marks or distortion and residues of liquids.
- The tip is prewetted with water. The nominal volume is drawn.

Limits: After 10 s no drop is formed at the tip.

1.2. GRAVIMETRIC TEST

Settings:

- The area in which the calibration is performed should be draught free, with a relative humidity above 50 %. This can be achieved with an evaporation trap or an open vessel containing water.
- Temperature of the purified water should be stable within ± 0.5 °C during calibration and maintained between 15° and 30°C. The influence of the temperature is compensated by the Z-factor.
- All the material needed for the test must be equilibrated for at least 2 hours in the room where the test is performed.

Materials:

- Appropriate analytical balance (for minimum requirements see table below)

selected volume of the pipette	**Display resolution**
1 µl to 10 µl	0.001 mg
> 10 µl to 100 µl	0.01 mg
>100 µl	0.1 mg

- Thermometer with a resolution of 0.1 °C or better.
- Barometer with a resolution of 0.5 kPa or better. Alternatively, call the local meteorological office and ask for the current air pressure or check the appropriate website for your region (Note: it is important to take into account that the air pressure in an air-conditioned laboratory might be appreciably different to the ambient pressure outside. A local barometric reading from a local weather station may therefore be misleading. Consequently, this alternative method for measuring the air pressure is not applicable in the case of an air-conditioned laboratory).
- Weighing vessel of suitable size filled with purified water (see water requirements in chapter “Visual Inspection”) to a depth of at least 3 mm.

- A reservoir vessel with purified water to an excess that allows performing all tests.
- Tips appropriate for the tested pipette.

Method:

- Variable volume pipettes are tested at 3 different volumes, each with 10 measurements:
    - the nominal volume
    - 50 % of the nominal volume
    - the lower limit of the useful volume range or 10 % of the nominal volume (whichever is greater)

  If a pipette is dedicated to a special task the laboratory may select 3 different volumes in the range of the pipette that cover the requirements.

- Each channel of multi-channel pipettes is checked separately with 3 different volumes, each with 10 measurements:
    - the nominal volume
    - 50 % of the nominal volume
    - 10 % of the nominal volume

  If a pipette is dedicated to a special task the laboratory can select 3 different volumes in the range of the pipette that cover the requirements.

- For repetitive piston pipettes the used tip defines the nominal volume. Thus, these pipettes have to be checked together with all types of tips used to dose the required volumes. The pipettes are tested at 2 different volumes, each with 10 measurements, with every type of tip used:
    - the largest volume, that can be dispensed with the tip
    - the smallest volume, that can be dispensed with the tip

  Take into account that the first and the last volume of a tip must be discarded.
  If defined volumes are used the laboratory can select different volumes in the range of the tip.

- Measure the temperature of the test liquid.
- Place the appropriate tip on the pipette.
- For variable volume pipettes choose the volume to be tested.
- For air-displacement pipettes fill the pipette at least 3 times with the test liquid and discard the fillings (pre-wetting), in order to create a moisture balance in the dead air volume.
- Dip the pipette into the test liquid.
- Draw the volume to be tested slowly and evenly. Observe the waiting time recommended by the manufacturer.
- Pull the pipette tip slowly out of the liquid, wiping it on the vessel wall.
- Determine the tare mass (reset balance).
- Deliver the test liquid slowly to the weighing vessel.
- Record the mass.
- Repeat drawing and dispensing of the test liquid further 9 times.
- Measure the temperature of the test liquid again.

NOTE 1: When performing the calibration tests, the pipette tips can be changed as frequently as considered necessary. It should be kept in mind to pre-wet every new tip.

NOTE 2: If it is not possible to ensure that the temperature of the water will remain constant during the whole period of the calibration test, the water temperature should be measured in between e.g. after each weighing.

NOTE 3: This procedure must always take into account the manufacturer's instructions and technical specifications for different types of pipettes and tips.

Calculations:

- Calculate the mean temperature *t* of the test liquid (rounded to the nearest 0.5 °C)

$$t = \frac{t_1 + t_2}{2}$$

t mean temperature
t_1 temperature before the first weighing
t_2 temperature after the last weighing

- Use the barometric pressure B and mean temperature *t* to find the corresponding Z-factor from the table.

t (°C) \ *B* (kPa)	80	85	90	95	100	101.3	105
				Z (µL/mg)			
15.0	1.0017	1.0018	1.0019	1.0019	1.0020	1.0020	1.0020
15.5	1.0018	1.0019	1.0019	1.0020	1.0020	1.0021	1.0021
16.0	1.0019	1.0020	1.0020	1.0021	1.0021	1.0021	1.0022
16.5	1.0020	1.0020	1.0021	1.0021	1.0022	1.0022	1.0022
17.0	1.0021	1.0021	1.0022	1.0022	1.0023	1.0023	1.0023
17.5	1.0022	1.0022	1.0023	1.0023	1.0024	1.0024	1.0024
18.0	1.0022	1.0023	1.0023	1.0024	1.0025	1.0025	1.0025
18.5	1.0023	1.0024	1.0024	1.0025	1.0025	1.0026	1.0026
19.0	1.0024	1.0025	1.0025	1.0026	1.0026	1.0027	1.0027
19.5	1.0025	1.0026	1.0026	1.0027	1.0027	1.0028	1.0028
20.0	1.0026	1.0027	1.0027	1.0028	1.0028	1.0029	1.0029
20.5	1.0027	1.0028	1.0028	1.0029	1.0029	1.0030	1.0030
21.0	1.0028	1.0029	1.0029	1.0030	1.0031	1.0031	1.0031
21.5	1.0030	1.0030	1.0031	1.0031	1.0032	1.0032	1.0032
22.0	1.0031	1.0031	1.0032	1.0032	1.0033	1.0033	1.0033
22.5	1.0032	1.0032	1.0033	1.0033	1.0034	1.0034	1.0034
23.0	1.0033	1.0033	1.0034	1.0034	1.0035	1.0035	1.0036
23.5	1.0034	1.0035	1.0035	1.0036	1.0036	1.0036	1.0037
24.0	1.0035	1.0036	1.0036	1.0037	1.0037	1.0038	1.0038
24.5	1.0037	1.0037	1.0038	1.0038	1.0039	1.0039	1.0039
25.0	1.0038	1.0038	1.0039	1.0039	1.0040	1.0040	1.0040
25.5	1.0039	1.0040	1.0040	1.0041	1.0041	1.0041	1.0042
26.0	1.0040	1.0041	1.0041	1.0042	1.0042	1.0043	1.0043
26.5	1.0042	1.0042	1.0043	1.0043	1.0044	1.0044	1.0044
27.0	1.0043	1.0044	1.0044	1.0045	1.0045	1.0045	1.0046
27.5	1.0045	1.0045	1.0046	1.0046	1.0047	1.0047	1.0047
28.0	1.0046	1.0046	1.0047	1.0047	1.0048	1.0048	1.0048
28.5	1.0047	1.0048	1.0048	1.0049	1.0049	1.0050	1.0050
29.0	1.0049	1.0049	1.0050	1.0050	1.0051	1.0051	1.0051
29.5	1.0050	1.0051	1.0051	1.0052	1.0052	1.0052	1.0053
30.0	1.0052	1.0052	1.0053	1.0053	1.0054	1.0054	1.0054

- Calculate the volume V_i in µl from the individual masses m_i.

$$V_i = Z \times m_i$$

V_i individual volume (calculated)
m_i weight of the individual volume

- Compute the mean volume $\bar{V}$ from the series of volumes.

$$\bar{V} = \frac{\sum V_i}{n}$$

$\bar{V}$ mean of the individual volumes from the series
V_i individual volume (calculated)
n number of weighings in the series

- Calculate the accuracy *e*, which is the difference between the mean volume of actual measurements and the true value as specified by the selected volume V_s. Accuracy is expressed in µl.

$$e = \bar{V} - V_s$$

e accuracy
$\bar{V}$ mean of the individual volumes from the series
V_s selected volume

- Calculate the repeatability *s*, which quantifies the scattering of individual weighings. It is expressed as variation coefficient (CV) in %.

$$s = \sqrt{\frac{\sum (V_i - \bar{V})^2}{n-1}}$$

$$CV = \frac{100 \times s}{\bar{V}}$$

s repeatability
Vi individual volume (calculated)
$\bar{V}$ mean of the individual volumes from the series
n number of weighings in the series
CV variation coefficient

Limits:
The following limits for the maximum permissible systematic error and the maximum permissible random error are applicable to pipettes used for quantitative determinations.
The laboratory may specify limits notwithstanding the following limits in order to meet the laboratory's requirements. For the definition of these differing limits the uncertainty of measurement should be taken into consideration.

Type A: Fixed and variable volume monochannel pipettes (air-displacement)

nominal volume	**maximum permissible systematic error**		**maximum permissible random error**	
[µl]	[%]	[µl]	[%]	[µl]
1	± 5	± 0.05	± 5	± 0.05
2	± 4	± 0.08	± 2	± 0.04
5	± 2,5	± 0.125	± 1.5	± 0.075
10	± 1.2	± 0.12	± 0.8	± 0.08
20	± 1	± 0.2	± 0.5	± 0.1
50	± 1	± 0.5	± 0.4	± 0.2
100	± 0.8	± 0.8	± 0.3	± 0.3
200	± 0.8	± 1.6	± 0.3	± 0.6
500	± 0.8	± 4.0	± 0.3	± 1.5
1000	± 0.8	± 8.0	± 0.3	± 3.0
2000	± 0.8	± 16.0	± 0.3	± 6.0
5000	± 0.8	± 40.0	± 0.3	± 15.0
10000	± 0.6	± 60.0	± 0.3	± 30.0

NOTE 1: For variable volume pipettes the limits of the nominal volume are valid for the whole range.

NOTE 2: For pipettes having a nominal volume not mentioned in the table above the limits of the nearest higher volume are valid.

Type D: Fixed and variable volume monochannel pipettes (positive displacement or direct displacement)

nominal volume	**maximum permissible systematic error**		**maximum permissible random error**	
[µl]	[%]	[µl]	[%]	[µl]
5	± 2.5	± 0.13	± 1.5	± 0.08
10	± 2	± 0.2	± 1.0	± 0.1
20	± 2	± 0.4	± 0.8	± 0.6
50	± 1.4	± 0.7	± 0.6	± 0.3
100	± 1.5	± 1.5	± 0.6	± 0.6
200	± 1.5	± 3.0	± 0.4	± 0.8
500	± 1.2	± 6.0	± 0.4	± 2.0
1000	± 1.2	± 12.0	± 0.4	± 4.0

NOTE 1: For variable volume pipettes the limits of the nominal volume are valid for the whole range.

NOTE 2: For pipettes having a nominal volume not mentioned in the table above the limits of the nearest higher volume are valid.

Type A: Fixed and variable volume multichannel pipettes (air-displacement)

nominal volume	**maximum permissible systematic error**		**maximum permissible random error**	
[µl]	[%]	[µl]	[%]	[µl]
1	± 10	± 0.10	± 10.0	± 0.1
2	± 8	± 0.16	± 4.0	± 0.08
5	± 5	± 0.25	± 3.0	± 0.150
10	± 2.4	± 0.24	± 1.6	± 0.16
20	± 2	± 0.40	± 1.0	± 0.2
50	± 2	± 1.0	± 0.8	± 0.4
100	± 1.6	± 1.6	± 0.6	± 0.6
200	± 1.6	± 3.2	± 0.6	± 1.2
500	± 1.6	± 8.0	± 0.6	± 3.0
1000	± 1.6	± 16.0	± 0.6	± 6.0
2000	± 1.6	± 32.0	± 0.6	± 12.0
5000	± 1.6	± 80.0	± 0.6	± 30.0
10000	± 1.2	± 120.0	± 0.6	± 60.0

NOTE 1: For variable volume pipettes the limits of the nominal volume are valid for the whole range.

NOTE 2: For Pipettes having a nominal volume not mentioned in the table above the limits of the nearest higher volume are valid.

NON-CONFORMITY OF PIPETTES

When a pipette has been shown to be outside specified limits after a calibration appropriate measures would be taking it out of service until it has been repaired and shown by another calibration to perform correctly.

In case of results that do not comply with the specification the laboratory should have a policy defining:

- the number of repeats
- the conditions for the repeats and
- the calculation of results.

3. INTERNAL CALIBRATION REPORT/CERTIFICATE

After the calibration, the laboratory should appropriately document the obtained results.
If an internal calibration report or calibration certificate is issued, the following minimum information should be included:

- Title of the report/certificate.
- Identification of the report/certificate.
- Entity having performed the calibration.
- Page numbering.
- Date of calibration.
- Reference to the gravimetric method used.
- Conditions of measurement (temperature, pressure).
- Z-factor.
- Unique identification of the material used to perform the calibration (balance, hygrometer, thermometer, etc) unless specified in another quality document.
- Unique identification of the pipette.
- Nominal volume of the pipette.
- Volumes selected for the calibration.
- Results of visual inspection or in-house maintenance before calibration.
- Calibration results (where applicable before and after maintenance).
- Uncertainty of measurement (if appropriate).
- Calculation of systematic error (accuracy) and random error (repeatability).
- Acceptance criteria (if appropriate).
- Conclusion (e.g. PASS/NOT PASS).
- Name and signature of the operator who performed the calibration.
- Name and signature of the supervisor responsible for the release for use of the pipette (preferably a different person from the operator who performed the calibration).

Note: If the laboratory requests calibration to an external company, it should be ensured that this minimal information is contained in the external calibration report/certificate. The responsible person in the laboratory should evaluate and approve this report/certificate as a release for use of the pipette.

4. INTERMEDIATE VERIFICATION OF PIPETTES

Intermediate in-house verifications between two periodic calibrations can be done either with the gravimetric method or alternatively with photometric or titrimetric methods according to ISO 8655-7. These methods have to be appropriately described in an internal document and must ensure the limits of section 1.2 with respect to the justified limits of the laboratory.

The gravimetric method is the reference method for pipette calibration.

NOTE: The results of intermediate verifications and the conditions under which these verifications were performed should be appropriately documented.

4.1. PHOTOMETRIC METHOD

The following method is proposed as an example of alternative method to the gravimetric method, for the intermediate in-house verification of pipettes between two periodic calibrations. It is applicable both to monochannel and multichannel pipettes.

NOTE: The described example is applicable to 200 µl pipettes. For pipettes of other volumes, the test has to be accordingly adapted.

4.1.1. PRECISION

Materials:

- Prepare a coloured solution with 0.46g of ferric chloride in 9.75ml of water and 250µl of hydrochloric acid. Protect the solution from light and use it within 20 min max.
- NOTE: Any suitable coloured solution read at the proper wavelength can be used.

Method:

Monochannel pipettes:

- Transfer 160µl of coloured solution to 6 wells of a microtiter plate (160µl x 6).
- Read the absorbance of the solution in each well at 450/620 nm.
- Calculate the mean, standard deviation and % CV of the 6 values.

Multichannel pipettes (example for 8 channels):

- Transfer 160µl of coloured solution to 8 wells of a microtiter plate (160µl x 8).
- Perform 6 measurements for each channel (48 measurements in total).
- Read the absorbance of the solution in each well at 450/620 nm.
- Calculate the mean, standard deviation and % CV for each channel and also for the 48 values.

Note: apply the same principle to pipettes with 12 and higher number of channels.

Limits: CV $\leq$ 2 %

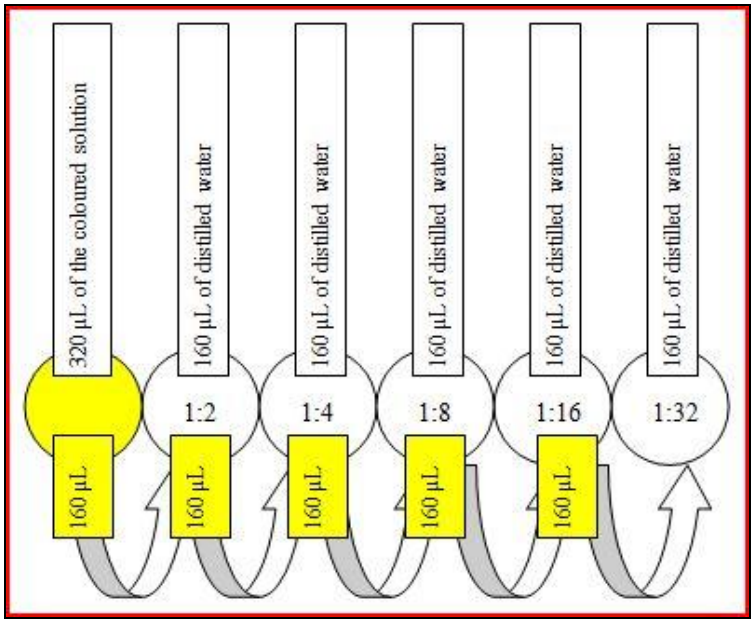
4.1.2. LINEARITY

Materials:

- Coloured solution (see 4.1.1).

Method:

- Prepare dilutions at 1:2, 1:4, 1:8, 1:16 and 1:32 as follows:



- Read the absorbance at 450/620 nm.
- Calculate the linear regression (absorbance/dilutions).

Limits:

- $R^2 \geq 0.98$
- Standard Error ≤ 0.02

NOTE 1: It is possible to use the "yellow solution" stated in the European Pharmacopoeia (see chapter 2.2. Physical and Physicochemical Methods – 2.2.2. Degree of Coloration of liquids, Reagents).

NOTE 2: The absorbance (O.D) of the non-diluted solution must be c.a. 0.7 in order to have a good linearity at a wavelength of 450 nm.

ANNEX II

Level IV. In-use instrument checks

This Annex contains practical examples of tests and their associated tolerance limits for several parameters related to the performance of pipettes.
These examples can be considered by the OMCLs as possible approaches to perform the Level IV of the equipment qualification process: "In-use instrument checks".

1. IN-USE INSTRUMENT CHECKS FOR ALL TYPES OF PIPETTES

Before every use a visual check for leaks, broken parts, air bubbles and contamination should be carried out.

To ensure a proper function of the pipette the following items should be considered:

- the type of pipette and the pipetting technique must be adequate for the dispensed liquid (viscosity, vapour pressure)
- the tip must be appropriate for the pipette
- the tips should be properly pre-wetted
- the temperature gradient between the pipette, the liquid and the air should be low

REFERENCES

(For all references, if the version is not mentioned, the latest version applies)

1) European Pharmacopoeia.
2) EN ISO 8655: Piston-operated volumetric apparatus.
3) FX07-011: Métrologie - Essais - Métrologie dans l'entreprise - Constat de vérification des moyens de mesure.
4) Measurement Good Practice Guide No. 69. The Calibration and Use of Piston Pipettes. ISSN 1368-6550. July 2004. National Physical Laboratory, United Kingdom. www.npl.co.uk.
5) Annex 1 of the OMCL Guideline on Validation of Computerized Systems: "Validation of computerised calculation systems. Example of validation of in-house software document" (PA/PH/OMCL (08) 87).