YALE-NEW HAVEN HOSPITAL	TITLE: PIPETTE CALIBRATION PROCEDURE		DEPT OF LAB MEDICINE Immunology, Flow Cytometry, and Molecular Diagnostics Laboratories Policy and Procedure Manual
			DOCUMENT # IMM 196
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WRITTEN BY: Voula Golfis	**EFFECTIVE DATE:** October 23, 2012	**REVISION:** NEW	**SUPERCEDES:**

I. PURPOSE:

The purpose of this procedure is to establish a consistent method for determining the accuracy and precision of mechanical pipettes used in the laboratory. This procedure will define the frequency of calibration, the method used to check calibration and the criteria for interpreting calibration results.

II. RESPONSIBILITY:

It is the responsibility for each laboratory section to monitor calibration of all pipettes used in their section. It is the responsibility of all technologists and technicians to know the frequency of pipette calibration and periodically check log books in their assigned areas.

III. FREQUENCY:

Pipettes should be calibrated when:
New pipette prior to placing in service, after maintenance or repair and/or routinely on a periodic basis as part of quality control:

A. Mechanical pipettes (Pipetman, MLA, SMI, Multi-channel, Eppendorf, etc.):

1. Pipettes used for volumetric measurement shall be calibrated for precision and accuracy at least every 6 months.

2. Pipettes used for non-volumetric sample transfer will not be calibrated and labeled "For Transfer Only, Not Calibrated".

IV. PROCEDURE:

Maintenance and calibration is performed for all mechanical pipettes by ACCU CAL Pipet Calibration & Document Services, Ashland, MA. See attached ACCU CAL procedure for complete procedure (Appendix A).

A. Check the room temperature is within 19-24°C

B. Each pipette is disassembled and cleaned prior to calibration. Worn out parts are replaced as needed.

C. Set the desired volume on the digital indicator. Begin with a volume near the low end of the pipette range. (See worksheet for exact settings)

D. Pre rinse the fresh pipette tip with Clinical Laboratory Reagent Water (CLRW).

E. Draw up CLRW and dispense into a small plastic weigh dish. Note the weight from the analytical balance.

F. Repeat this step 4 more times at the same volume. Check these values against the acceptable mean error criteria of 1% the set volume.

G. If any of the measurements are not within the specified limits of error, loosen the set screw and/or turn the calibration screw; clockwise to decrease and counter clockwise to increase the volume. Laboratory is notified if a specific pipette can not be calibrated within standards and the pipette is removed from service.

H. Re-tighten the set screws and measure the volume again.

I. Repeat steps F - G until measurements fall within range

J. Repeat steps B – G for the various pipette volumes

V. CALCULATION AND INTERPRETATION OF RESULTS:

ACCU Cal provides a Calibration Report / Certificate of Compliance for each pipette which includes the statistics used to verify calibration

A. Calculations:

1. Accuracy:

Mean = x = sum of X_i/N = _____ mg

Mean Volume = V = x = _____ µL

* Accuracy (Acc) = $\frac{\text{Mean Volume - Ideal Volume}}{\text{Ideal Volume}} \times 100$ = _____%

* Accuracy as used here is actually the percentage of inaccuracy relative to the "ideal" or expected volume.

2. Precision:

a. Standard deviation (SD):

$$S.D. = \frac{\text{sum of } (x_i - x)^2}{N - 1} = \_\_\_\_\_ \mu L$$

b. Coefficient of variation (C.V.):

$$C.V. = \frac{S.D.}{x} \times 100 = \_\_\%$$

B. Guidelines for Interpretation of Results:

Pipette Volume	Precision (CV)	Accuracy (Acc)
1000 µL	± 1.0%	± 1.0%
500 µL	± 1.0%	± 1.0%
200 µL	± 1.0%	± 1.0%
100 µL	± 1.0%	± 1.0%
50 µL	± 1.0%	± 1.0%
20 µL	± 1.0%	± 1.0%

1. Precision:

   Precision indicates how well the results can be reproduced. This is indicated by the C.V. value.

2. Accuracy:

   Accuracy indicates how close the results are to the stated volume of the pipette. The mean volume indicates exactly the volume being delivered by the pipette.

3. Mean Error %: Allowable Mean Error is +/- 1%.

4. All calibrated pipettes are then tagged with a sticker which indicates calibration date and date next calibration is due

APPENDIX A

ACCUCAL
Pipet Calibration & Documentation Services
207 Captain Eames Circle, Ashland, MA 01721
Rodney Lewis
rodniel@aol.com.
1-800-888-7754

STANDARD OPERATING PROCEDURE

1.0 CALIBRATION PROCEDURE:

1.1 Check the room temperature is within 19-24°C.
1.2 Set the desired volume on the digital indicator. Begin with a volume near the low end of the pipette range. (See worksheet for exact settings)
1.3 Pre-rinse the fresh pipette tip with distilled water.
1.4 Draw up distilled water and dispense into a small plastic weigh dish. Note the weight from the analytical balance.
1.5 Repeat this step 4 more times at the same volume. Check these values against the acceptable mean error criteria on the Calibration Worksheet.
1.6 If any of the measurements are both within the specified limits of error, loosen the set screws and/or turn the calibration screw-clockwise to decrease and counter clockwise to increase the volume.
1.7 Re-tighten the set screws and measure the volume again.
1.8 Repeat steps 1.6 – 1.7 until measurements fall within range.
1.9 Repeat steps 1.2 – 1.8 for the volume given in the Worksheet.

2.0 MAINTENANCE PROCEDURE:

2.1 As a matter of routine precaution this pipette was checked for background radiation.
2.2 Disassemble the Pipette.
2.3 Clean the shaft, Pison and all internal parts with Acetone. Replace excessively worn out parts.
2.4 Replace the Teflon seal and O-ring.
2.5 Reassemble the unit.
2.6 Polish.
2.7 Calibrate the instrument following steps 1.1 – 1.9.

3.0 TAGGING PROCEDURE:

3.1 Following calibration. Place the sticker marked with due date for the next calibration (6 months from the Calibration date).

3.2 Each pipette we repair or certify carried with it our "No Questions Asked" six month warranty on repair and recalibration and replacement of parts.

3.3 Work performed by Rodney Lewis, certified technician with 18 years' experience. Pipettes are certified to perform within the tolerances for a period of 6 months.

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Signature Approval for Annual Review
Name: Pipette Calibration

Document #: IMM 196

Name (Print)	Title	Signature	Date of Review	Revision Page and Section # (Use Procedure Review Log to document staff review)	Issue Date for Training if Applicable	Effective Date for Use
TEODORICO LEE	LAB MANAGER	Teodorico Lee	11/7/12	NEW		10/23/12
BRIAN SMITH	LAB DIRECTOR	[illegible]	11/4/12	NEW		10/23/12