



**US Environmental Protection Agency  
Office of Pesticide Programs**

**Office of Pesticide Programs  
Microbiology Laboratory  
Environmental Science Center, Ft. Meade, MD**

**Standard Operating Procedure for  
Calibration of Kimble Class A Burets**

**SOP Number: EQ-06-04**

**Date Revised: 06-27-08**

EPA/OPP MICROBIOLOGY LABORATORY  
ESC, Ft. Meade, MD

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Initiated By: \_\_\_\_\_ Date: \_\_\_/\_\_\_/\_\_\_

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1.0 SCOPE AND APPLICATION:

1.1 This protocol describes the calibration method for 10 mL and 50 mL Kimble Class A burets. The calibration is done on an annual basis.

2.0 DEFINITIONS:

2.1 Calibration = The determination of the difference between the volume dispensed and the expected volume.

2.2 Adjustment = The correction of the difference between the measured value and the expected volume of the liquid displaced.

2.3 NIST= National Institute of Standards and Technology.

3.0 HEALTH AND SAFETY: Not applicable

4.0 CAUTIONS:

4.1 The water used for calibration should be weighed immediately after being dispensed to avoid evaporation.

4.2 Burets should be inspected for chips and cracks prior to use and prior to calibration.

5.0 INTERFERENCES:

5.1 It is important that the volumes of water are dispensed precisely, otherwise the entire calibration process can be impacted.

6.0 PERSONNEL QUALIFICATIONS:

6.1 Personnel are required to be knowledgeable of the procedures in this SOP.

7.0 SPECIAL APPARATUS AND MATERIALS:

7.1 Kimble Class A buret (10 mL) with 0.1 mL increments.

7.2 Kimble Class A buret (50 mL) with 1.0 mL increments.

7.3 Sartorius Basic Plus Model BP 211D (Serial Number 80904707): Weighs 0 to 40 / 80 / 210 g, reads to 0.00001 g or .01 mg, reproducibility # 0.02 / 0.05 / 0.1 mg (Manufacturer=s Claims).

7.4 Sartorius Master<sup>Pro</sup> Series Model LP 420 (Serial Number 81107148): Weighs 0 to 420 g, reads to 0.01 g or 10 mg, reproducibility # 0.01 g (Manufacturer=s Claims).

7.5 Microsoft Excel Spreadsheet (2003)

#### 8.0 INSTRUMENT OR METHOD CALIBRATION:

8.1 The weigh balances are calibrated annually by a professional calibration service and are checked quarterly for accuracy using a reference weight set certified by NIST (see SOP EQ-03, Weigh Balances).

9.0 SAMPLE HANDLING AND STORAGE: Not applicable

#### 10.0 PROCEDURE AND ANALYSIS:

10.1 Wash and rinse the buret with de-ionized water. Place a clean 50 mL beaker on the balance and tare it. Fill the buret with room temperature de-ionized water and adjust the level of the water to the zero mark while allowing the rest of the buret to become filled. Remove any air bubbles. Record the temperature of the water on the Calibration of Kimble Class A Buret Form (see 16.1).

10.2 For the 10 mL buret, dispense into the pre-tared beaker 5 serial aliquots (2 mL each) of water from the filled buret. Weigh the pre-tared beaker after the addition of each 2 mL aliquot. Record results on the Calibration of Kimble Class A Buret Form (see 16.1).

10.3 For the 50 mL buret, dispense into the pre-tared beaker 5 serial aliquots (10 mL each) of water from the filled buret. Weigh the pre-tared beaker after the addition of each 10 mL aliquot. Record results on the Calibration of Kimble Class A Buret Form (see 16.1).

10.4 Weights are plotted against the independent variable of the volume reading on the buret using Microsoft Excel.

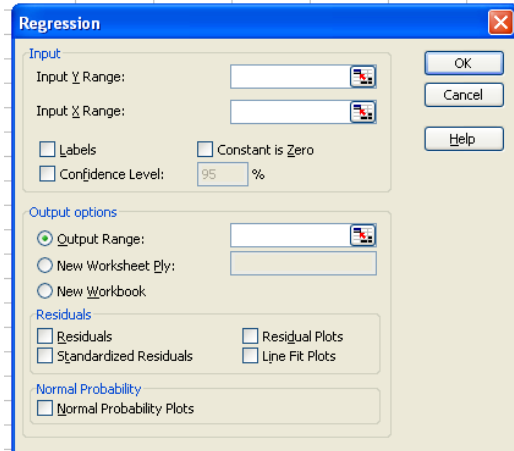
#### 11.0 DATA ANALYSIS/CALCULATIONS:

11.1 Simple linear regression is calculated by Microsoft Excel.

11.1.1 For the 10 mL buret, enter X values in column A of the spreadsheet and Y values in column B.

11.1.2 In Microsoft Excel, select Tools → Data Analysis → Regression → OK.

### 11.1.3 Pop-up menu:



### 11.1.4 In the pop-up menu under *Input*:

- Click once in the blank cell to the right of *Input Y Range*, and then highlight the column of 5 y-values in the spreadsheet.
- Click once in the blank cell to the right of *Input X Range*, and then highlight the column of 5 x-values in the spreadsheet.

### 11.1.5 In the same pop-up menu under *Output*:

- Select the *Output Range* option.
- Click once in the blank cell to the right of *Output Range*, and then highlight a single empty cell beneath the “Vol. X” column in the spreadsheet (e.g., cell A16).

### 11.1.6 Select OK.

### 11.1.7 Summary output:

SUMMARY OUTPUT

<i>Regression Statistics</i>	
Multiple R	0.999994087
R Square	0.999988173
Adjusted R Square	0.999984231
Standard Error	0.012564167
Observations	5

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	1	40.04241124	40.04241124	253660.4849	1.72618E-08
Residual	3	0.000473575	0.000157858		
Total	4	40.04288481			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	0.006658	0.01317741	0.505258638	0.648159495	-0.035278398	0.048594398	-0.035278398	0.048594398
X Variable 1	1.00053	0.001986569	503.647183	1.72618E-08	0.99420785	1.00685215	0.99420785	1.00685215

11.1.8 Repeat steps 11.1.1 through 11.1.7 for the 50 mL buret.

11.2 The corrected value, or adjustment, for any volume (y) dispensed by the buret is obtained by inserting the volume reading (in mL) on the buret into the following equation:

$y = mx + b$ ; where y = corrected volume, m = slope (x variable 1 coefficient), x = volume reading on buret, and b = y-intercept (intercept coefficient). The numbers are rounded to the nearest tenth of an mL.

11.2.1 A label displaying the date of calibration and corrected value (even if it is zero) for any volume dispensed by the buret is placed around the top of the buret.

12.0 DATA MANAGEMENT/RECORDS MANAGEMENT:

12.1 Data will be recorded promptly, legibly and in indelible ink on the Calibration of Kimble Class A Buret Form. The accompanying spreadsheet will be archived with the raw data sheet. Completed forms are archived in notebooks kept in secured file cabinets in the file room D 217. Only authorized personnel have access to the secured files. Archived data are subject to OPP=s official retention schedule contained in SOP ADM-03, Records and Archives.

13.0 QUALITY CONTROL:

13.1 The calibration of burets is performed annually and the information is documented on the appropriate record form(s) (see 16.1).

14.0 NONCONFORMANCE AND CORRECTIVE ACTION:

- 14.1 Burets exhibiting chips and cracks will not be used in the laboratory and will be discarded.
- 14.2 When routinely using the burets in the laboratory to dispense liquids, analysts must record the volume dispensed by the buret plus the corrected value, or adjustment, for that volume.
- 15.0 REFERENCES: None
- 16.0 FORMS AND DATA SHEETS:
  - 16.1 Calibration of Kimble Class A Buret Form.
  - 16.2 Example of Regression Analysis Spreadsheet

Calibration of Kimble Class A Buret  
 OPP Microbiology Laboratory

Date/Initials _____ Temperature of Water _____
--

10 mL Buret ID. \_\_\_\_\_

Volume X (mL Buret )	Weight Y (gm)
2 ml	
4 mL	
6 mL	
8 mL	
10 mL	

50 mL Buret ID. \_\_\_\_\_

Volume X (mL Buret )	Weight Y (gm)
10 mL	
20 mL	
30 mL	
40 mL	
50 mL	

The corrected value for any volume (y) dispensed by the buret is obtained by inserting the volume reading (in mL) on the buret into the following equation:  $y = mx + b$ ; where y = corrected volume, m = slope (x variable 1 coefficient), x = volume reading on buret, and b = y-intercept (intercept coefficient). The numbers are rounded to the nearest tenth of an mL.

## Example of Regression Analysis Spreadsheet

### **Buret Regression Analysis**

Calibration Date: 00/00/00  
 Analyst Name: J. Smith  
 Temperature of water: 23°C  
 Thermometer #: #12345  
 Buret Volume: 10 mL  
 Buret ID: #1

Vol. X	Vol. Y
2	1.99465
4	4.01910
6	6.01814
8	8.01560
10	10.00170

### Corrected Y Value

Vol. X = 2 mL      y = 2.0  
 Vol. X = 4 mL      y = 4.0  
 Vol. X = 6 mL      y = 6.0  
 Vol. X = 8 mL      y = 8.0  
 Vol. X = 10 mL     y = 10.0

To determine corrected y value:  $y = mx + b$

### SUMMARY OUTPUT

<i>Regression Statistics</i>	
Multiple R	0.999994087
R Square	0.999988173
Adjusted R Square	0.999984231
Standard Error	0.012564167
Observations	5

### ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	1	40.04241124	40.04241124	253660.5	1.72618E-08
Residual	3	0.000473575	0.000157858		
Total	4	40.04288481			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	0.006658	0.01317741	0.505258638	0.648159	-0.035278398	0.048594398	-0.0352784	0.048594398
X Variable 1	1.00053	0.001986569	503.647183	1.73E-08	0.99420785	1.00685215	0.99420785	1.00685215