

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-06-05

Date Revised: 10-05-11

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

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Initiated By:		Date://
	Print Name:	
Technical Review:		Date://
	Print Name:	
QA Review:	Technical Staff	Date://
	Print Name: QA Officer	
Approved By:		Date:/
	Print Name: Branch Chief	<u></u>
Effective Date:	//	
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1.0 <u>SCOPE AND APPLICATION</u>:

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 <u>DEFINITIONS</u>:

- 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 ISO = International Organization for Standardization
- 3.0 <u>HEALTH AND SAFETY</u>: 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 <u>SPECIAL APPARATUS AND MATERIALS</u>:

- 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 BP211D (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^{Pro} 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 (2007)

8.0 INSTRUMENT OR METHOD CALIBRATION:

- 8.1 The weigh balances are calibrated annually by an ISO accredited vendor and are checked quarterly for accuracy using a reference weight set (see SOP EQ-03, Weigh Balances).
- 9.0 <u>SAMPLE HANDLING AND STORAGE</u>: 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.

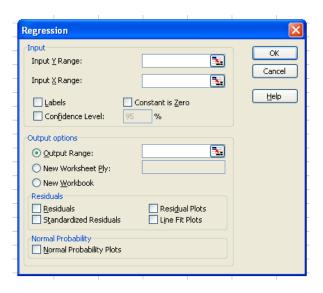
11.0 DATA ANALYSIS/CALCULATIONS:

- 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, where X = theoretical value (volume in mL) and Y =

actual value (weight in g).

11.1.2 In Microsoft Excel, select the "Data" tab of Excel's ribbon. Then from the "Analysis" group, click the "Data Analysis" button. From the pop-up menu, select "Regression" option then 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

Regression Statistics							
Multiple R	0.999994087						
R Square	0.999988173						
Adjusted R Square	0.999984231						
Standard Error	0.012564167						
Observations	5						

ANOVA

	af .	SS	MS'	F	Significance F
Regression	1	40.04241124	40.04241124	253660.4849	1.72618E-08
Residual	3	0.000473575	0.000157858		
Total	4	40.04288481			

	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95.0%	Upper 95.0%
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 correction factor, 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 table within the spreadsheet generates a correction factor for each volume evaluated using the aforementioned calculation. The average correction factor is calculated and used to determine the correction factor for the buret.
- 11.2.2 Place a label displaying the date of calibration and the correction factor (even if it is zero) for any volume dispensed by the buret 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 <u>REFERENCES</u>: 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 Ana	<u>alysis</u>							
Calibration Date:	00/00/00							
Analyst Name:	J. Smith							
Temperature of water:	23°C							
Thermometer #:	#12345							
Buret Volume:	10 mL							
Buret ID:	#1		Corrected Y Vah	<u>ie</u>		Correction Factor	Avg. Correc	tion Factor
			Vol. $X = 2 mL$	y =	2.0	0.0	- 0.	.0
Volume X (mL)	Weight Y (g)	,	Vol. $X = 4 \text{ mL}$	y =	4.0	0.0		
2	1.99465		Vol. $X = 6 \text{ mL}$	y =	6.0	0.0		
4	4.01910		Vol. $X = 8 \text{ mL}$	y =	8.0	0.0		
6	6.01814		Vol. X = 10 mL	y =	10.0	0.0		
8	8.01560			-				
10	10.00170		To determine co	rrected y va	flue: $y = mx + b$			
Regression S								
	0.999994087							
Multiple R								
R Square	0.999988173 0.999984231							
Adjusted R Square Standard Error								
Observations	0.012564167							
Observations		•						
ANOVA								
	df	SS	MS	F	Significance F	-		
Regression	1	40.04241124	40.04241124	253660.5	1.72618E-08	-		
Residual	3	0.000473575	0.000157858					
Total	4	40.04288481						
	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95.0%	Upper 95.0%
Intercept	0.006658	0.01317741	0.505258638	0.648159	-0.035278398			0.048594398
X Variable 1	1.00053	0.001986569	503.647183	1.73E-08	0.99420785	1.00685215		1.00685215