

ESS INO GENOP 200 Pipette Performance Checks

1. Purpose

- 1.1 Air displacement pipettes used for the measurement of liquids in the laboratory must be tested on a regular basis to insure volumes delivered are accurate and precise. This method describes two different means of monitoring the performance of air displacement pipettes.

2. Summary

- 2.1 Pipettes are tested by pipetting aliquots of a reagent and comparing the resulting mean and standard deviation to the nominal or "true" value.
- 2.2 All pipettes must be tested when first purchased, following any major service and once annually by analyzing ten replicates.
- 2.3 All pipettes must be tested quarterly by analyzing four replicates.
- 2.4 Replicate analyses must meet acceptance criteria or use of the pipette should be discontinued until the problem has been corrected.
 - 2.4.1 % Inaccuracy $[(\text{Corr. Mean} - \text{true value}) \div \text{true value} \times 100]$ must be less than 2%
 - 2.4.2 No single replicate may be greater than 2% from the true value.
 - 2.4.3 %CV $(\text{Standard Deviation} \div \text{Corr. Mean} \times 100)$ must be less than 1.00
- 2.5 The gravimetric method is used for the analysis of volumes of 1 mL or greater. The Artel PCS 2 Pipette Calibration System is used for the analysis of volumes of 1 mL or less.
- 2.6 If you are checking an adjustable volume pipette, three different volumes are tested; 10% of maximum volume, mid volume and maximum volume.

3. Apparatus

- 3.1 Pipette Tips
- 3.2 Balance (NBS certified) connected to a PC with the BalLink software installed.
- 3.3 Weight table
- 3.4 Disposable beakers
- 3.5 Thermometer
- 3.6 250 mL beaker

4. Procedure

- 4.1 Each analyst performing the pipette calibration must have an operator ID number and each pipette being calibrated must have a unique pipette ID number. The PCS 2 will only accept numeric characters for either of these ID numbers.
 - 4.1.1 Open the Excel worksheet M:\EHD\Forms\Pipette Calibration System\Pipette Calibration Info.xls.
 - 4.1.2 On the "Pipettes" page, enter the pipette #, manufacturer, type, volume and serial number of any pipette which has not had an ID number assigned to it. The suggested format for the pipette # is the room number the pipette is stored in followed by a two digit number for the pipette. For example: 11901, 11902, etc. This number should then be placed on the pipette also.

- 4.1.3 On the "Analyst" page, enter your name and department on the next available line to obtain an operator ID number.
- 4.1.4 Save the changes to the spreadsheet and close Excel.
- 4.2 Gravimetric analysis
 - 4.2.1 Log on to N.T and double click the BalLink icon on the desktop.
 - 4.2.2 Click on Setup and chose "Data String" from the menu. In the "Movement after Data Entry" box, enter 0 for movement horizontally and 1 for the movement vertically.
 - 4.2.3 If using the balance in the Solids area in room 117, the settings for the balance must be changed for the capture of the readings.
 - 4.2.3.1 Click on Setup and chose "Data Initialization File" from the menu. Chose "weights" and double click on "pipet.met".
 - 4.2.3.2 These settings **must be returned to the original ones** using the path M:\EHD\ESS(4900)\ESS Inorg(4910)\General Chemistry\Solids\Balance\weights\at200.MET once pipette calibration has been completed.
 - 4.2.4 Click on "Open Excel Worksheet". The worksheet is found at M:\EHD\Forms\Pipette Calibration System\ESS Pipette Cal.xls
 - 4.2.5 Using the tabs at the bottom of the screen, choose the appropriate page (Annual or Quarterly).
 - 4.2.5.1 Enter the date, the temperature of the reagent water to be used, and the analyst initials or operator ID number. The spreadsheet will automatically find the correct Z-Factor.
 - 4.2.5.2 Enter the pipette # and the volume you will be pipetting.
 - 4.2.5.3 Place cursor in the top empty cell of the weight column.
 - 4.2.6 Place a clean, dry disposable beaker on the balance, close and tare the balance.
 - 4.2.7 Apply a clean tip to the pipette.
 - 4.2.8 Operate the pipette's action a few times prior to using. This will redistribute the lubricant and ensure a smooth positive action.
 - 4.2.9 Wet the tip by drawing up an aliquot of reagent water and discard.
 - 4.2.10 Open the balance and pipette an aliquot of reagent water into the disposable beaker taking care to touch off any remaining liquid on the tip.
 - 4.2.11 Close the balance. Once the weight has stabilized, depress the foot pedal or hand button to send the weight to the spreadsheet.
 - 4.2.12 Tare the balance and repeat for as many replicates are needed. If a mistake was made during one replicate, move the cursor to that weight and pipette a new replicate.
 - 4.2.13 Check if results fall within the Acceptance Criteria (2.4.1 - 2.4.3).
 - 4.2.14 If the pipette fails the criteria, take corrective action and perform the analysis again, recording the results in a new portion of the same spreadsheet. Record the corrective action taken next to the failed analysis on the hard copy when it has been printed.
 - 4.2.15 If the criteria has been met, print the worksheet and place the hard copy in an appropriate binder.
 - 4.2.16 Close Excel. **Do not save changes to the spreadsheet!**
 - 4.2.17 If needed, return balance settings to the original settings (4.2.3.1, 4.2.3.2).
- 4.3 Photometric analysis using the Artel PCS 2 Pipette Calibration System.
 - 4.3.1 Turn on the PCS 2 and the printer. Be sure the printer is on line.
 - 4.3.2 The PCS 2 instrument calibration should be checked every thirty days.

- 4.3.2.1 Check the calibration log for the PCS 2 to determine when the instrument calibration was last checked. If needed, perform the calibration check (refer to the PCS 2 Procedure Guide, page 18).
- 4.3.2.2 Enter all required information in the log. Record any failures in the comment field.
- 4.3.2.3 If any failures have occurred take corrective action and recheck the calibration.
- 4.3.2.4 Attach the instrument calibration check printout to a blank piece of paper and place it in the back of the log.
- 4.3.3 Choose a Reagent Kit. The Reagent Kit must be at the same ambient temperature as the instrument. If it is not, allow time for the temperatures to equilibrate.
- 4.3.4 At the prompt "Pipette Calibration" press YES.
- 4.3.5 Enter your operator ID number (4.1.3)
- 4.3.6 Enter the reagent lot code. It can be found inside the cover of the Reagent Kit.
- 4.3.7 If this reagent lot code matches a previous one entered, the pipette calibration will continue. If not, the instrument will prompt "New Lot?". If no, the correct lot code will need to be reentered. If yes, a new calibration code must be entered. It can be found in the reagent kit insert.
- 4.3.8 At the prompt, insert CAL A vial into the instrument with the label facing toward you and close the cover. Take care to not touch the lower portion of any vial as smudges on the glass will affect the results. If there are smudges on the glass, clean carefully with a Kimwipe® before inserting into the instrument.
- 4.3.9 At prompt, remove CAL A vial. Mix a blank vial, remove the cap and insert into the instrument.
- 4.3.10 At the "Enter Pipette Serial #" prompt, enter the pipette number (4.1.2).
- 4.3.11 Enter the pipette volume.
- 4.3.12 The number of samples which can be performed at this pipette volume will now be displayed followed by "Proceed?". Press YES.
- 4.3.13 The instrument will display the correct Range (1,2,3 or 4) Solution to be used at the volume you entered. Find the appropriate range solution in the Reagent Kit. Mix by inverting the vial several times, pour a small amount into a clean disposable beaker and recap the solution.
- 4.3.14 Operate the pipette's action a few times prior to using. This will redistribute the lubricant and ensure a smooth positive action.
- 4.3.15 Wet the tip by drawing up an aliquot of Range Solution and discard.
- 4.3.16 Open the cover of the instrument and pipette the first sample into the Blank vial without touching or removing it from the instrument. Close the cover.
- 4.3.17 The instrument will display the previous reading as it prompts you for the next sample addition. Repeat the above procedure for the desired number of sample additions. If a mistake is made on any sample addition, add another addition at the end. The bad replicate can be deleted from the calculations later.
- 4.3.18 When the correct number of sample additions has been reached, press END OF RUN. The instrument will calculate and print the statistical summary based on all sample additions.
- 4.3.19 The instrument will prompt "Reprint Results?" If you wish to eliminate a sample addition result from the summary, press YES. Follow the instrument prompts to delete any unwanted data points.
- 4.3.20 If all data points are satisfactory, press NO at the "Reprint Results" prompt.
- 4.3.21 Check if results fall within the Acceptance Criteria (2.4.1 - 2.4.3).

- 4.3.22 If the pipette fails the criteria, take corrective action and perform the analysis again. Record the corrective action taken next to the failed analysis on the hard copy when it has been printed.
- 4.3.23 If the criteria has been met, attach the hard copy to a header sheet and place the sheet in an appropriate binder.
- 4.3.24 At the "Another Cal w/Vial?:" prompt, press NO, turn off the instrument and the printer. Place all reagent and range solution vials back in the Reagent Kit and dispose of all used blank vials.

5. Definitions

- 5.1 Mean weight (mg): Total of individual weight measurements divided by the total number of measurements.
- 5.2 Corrected Mean Volume (μl): Mean Weight (mg) \times Z-Factor. The Z-Factor is a correction for the change in weight of water due to the temperature. See Table 200.1.
- 5.3 % Inaccuracy: $(\text{Corr. Mean} - \text{true value}) \div \text{true value} \times 100$. This is the difference between the corrected mean and the true (ideal) value of the pipette.
- 5.4 Standard Deviation (Std. Dev.): Standard Deviation quantifies the magnitude of scatter due to random error.
- 5.5 Coefficient of Variation (% C.V.):

$$\% \text{ C.V.} = \frac{\text{Std. Dev.}}{\text{Corr. Mean } (\mu\text{L})} \times 100$$

6. Related Documents

- 6.1 Performance Assurance for Air Displacement Pipettes, by Liquid Measurement Quality Control, Rainin Instrument Co., Inc.
- 6.2 EDP Plus Motorized Microliter Pipette User Guide and EDP 2 Instructions, Rainin Instrument Co., Inc.
- 6.3 PCS 2 Pipette Calibration System Procedure Guide, Artel, Inc. March 1997.
- 6.4 Oxford Sampler Single and Multi-Range Micropipetting Systems, by Monoject Scientific, St. Louis, MO, June 1984.
- 6.5 Quality Assurance Procedures and Policies, Wisconsin State Laboratory of Hygiene, Revision 3.1, February 15, 2001. \\slhehd\grp\EHD\ESS(4900)\Admin\QA\QA Manual\WSLH ESS Quality Assurance Manual Rev 3_1.doc
- 6.6 National Environmental Laboratory Accreditation Conference, Constitution, Bylaws, and Standards, United States Environmental Protection Agency, Offices of Research and Development, Washington DC 20460, July, 1999.

Table 200.1
Z – Factors

Water Temperature (°C)	Z-Factor (µl/mg)
16.0	1.0021
16.5	1.0022
17.0	1.0023
17.5	1.0024
18.0	1.0025
18.5	1.0026
19.0	1.0027
19.5	1.0028
20.0	1.0029
20.5	1.0030
21.0	1.0031
21.5	1.0032
22.0	1.0033
22.5	1.0034
23.0	1.0035

(Values from ISO Standard.)

Pipette Performance Checks
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