Released by/Date Effective:

Author Name	Title	Signature/Date

Approver Name	Title	Signature/Date

1. PURPOSE

1.1. The purpose of this procedure is to describe the use and maintenance of Rainin brand pipettes used in the HPV Serology Laboratory.

2. SCOPE

2.1. This procedure applies to the HPV Serology Laboratory located at the Advanced Technology Research Facility, Room C2007.

2.2. This procedure applies to Rainin Brand pipettes, both manual and electronic types; however, this SOP may be applicable for other brands as well, if needed.

3. REFERENCES

3.1. Rainin Original Certificate of Conformity

3.2. HSL_EQ_015: Use and Maintenance of an Analytical & Precision Balance

3.3. HSL_EQ_019_Use and Maintenance of the Milli-Q Integral 3 Water System

3.4. HSL_GL_001: Waste Disposal at the Advanced Technology Research Facility

3.5. HSL_GL_002: Equipment Qualification and Calibration in the HPV Serology Laboratory

3.6. HSL_GL_003: Good Documentation Practices for the HPV Serology Laboratory

3.7. HSL_GL_004 Laboratory Notebook Control and Use for the HPV Serology Laboratory

3.8. HSL_GL_007: Reagent and Chemical Expiry in the HPV Serology Laboratory

3.9. HSL_GL_008: Laboratory Flow and Gowning Procedures for the HPV Serology Laboratory

3.10. HSL_GL_009: HPV Serology Laboratory BSL-2 Procedures

3.11. HSL_GL_010: Control and Request of Documents in the HPV Serology Laboratory

4. RESPONSIBILITIES

4.1. The Research Associate, hereafter referred to as analyst, is responsible for reviewing and following this procedure.

4.2. The Scientific Manager or designee is responsible for training personnel in this procedure and reviewing associated documentation.

4.3. The Quality Assurance Specialist is responsible for quality oversight and approval of this procedure.

5. REAGENTS, CHEMICALS AND EQUIPMENT

5.1. Rainin Pipettes

5.2. Rainin Tips

5.3. Analytical Balance

5.4. 15 mL Conical Tubes (Warehouse, Cat # 66401479 or equivalent)

5.5. Bleach, concentrated (Warehouse, Cat # 68100251)

5.6. Ster-ahol (Fisher, Cat # 19-546-862 or equivalent)

5.7. Weigh Boats (VWR, Cat # 89106-768 or equivalent)

5.8. Weigh Paper (VWR, Cat # 12578-121 or equivalent)

5.9. Wypalls paper towel (Warehouse, Cat # 79300335 or equivalent)

6. HEALTH AND SAFETY CONSIDERATIONS

6.1. Proper safety precautions should be taken while working in a laboratory setting. This includes, but is not limited to, proper protective equipment such as lab coats, safety glasses, closed-toe shoes, and non-latex gloves.

6.2. Refer to the respective SDS when working with any chemicals.

6.3. Refer to “HSL_GL_001: Waste Disposal at the Advanced Technology Research Facility” regarding waste disposal processes at the ATRF.

7. DEFINITIONS

Term	Definition
BioClean	Free or almost free from harmful microorganisms
FME	Facilities, Maintenance and Engineering
HPV	Human Papillomavirus
HSL	HPV Serology Laboratory
SDS	Safety Data Sheets
SOP	Standard Operating Procedure
Type II water	Pure/Analytical Grade, used for standard applications

8. EQUIPMENT USE

8.1. Only use Rainin Brand tips with Rainin Brand pipettes. Rainin pipettes and tips are designed together as a pipetting system, both in LTS and traditional versions.

8.1.1. All Rainin tips are BioClean and totally inert, to assure the best pipetting results.

8.1.2. To mount a tip, press the pipette shaft into the end of the tip with light force. With both LTS and traditional versions, the tip will seal properly on the shaft with minimal force. Do not use more force than is required.

8.1.3. Don't invert or lay the pipette flat with liquid in the tip.

8.2. Minimize heat transfer from user to pipette, by returning the pipette to the stand between deliveries.

8.3. When using a pipette, immerse the tip to the proper depth (typically 2-6 mm below the meniscus) to prevent improper measurement.

8.4. Use consistent plunger pressure and speed while pipetting.

8.5. Hold pipette upright at a 90° angle when pipetting.

8.6. Clean each pipette prior to use, with either a 10% bleach solution or alcohol solution such as Ster-ahol or if contamination occurs.

8.7. To eliminate errors due to mechanical backlash when setting the desired volume, first turn the knob 1⁄3 turn above the desired volume. Then turn the knob slowly clockwise until the desired volume is displayed. Always dial down to the desired volume.

8.8. Pipettes will be assigned an equipment number, and recorded on appropriate data capture forms when used.

8.9. A binder will be created for pipettes used in C2007, and each pipette will have a section for storage of calibration or maintenance information.

9. PIPETTE PREVENTATIVE MAINTENANCE AND CALIBRATION

9.1. All Rainin pipettes come with a Certificate of Conformity, which indicates a date that the pipette was tested prior to shipping, to show the pipette met with all pipette specifications. In the event that a pipette cannot be calibrated immediately on receipt, the Certificate of Conformity can be used in place of a pipette calibration for up to 6 months after the date the testing occurred.

9.2. Pipettes should be calibrated by a qualified vendor every 6 months, or if damage to the pipette occurred and it does not seem to be pipetting correctly.

9.3. Dry seal and O Ring replacement should occur yearly, and be replaced by the qualified vendor performing the calibration.

9.4. The vendor should perform an "As Found" and "As Left" check on each pipette, and perform the calibration at a minimum of the 10% and 100% volume settings, which will change for each pipette based on the pipette volume.

9.5. Pipette calibration certificates will be stored with the equipment files in HSL Archive.

10. CONFIRMATION OF A PIPETTE ACCURACY

Note: If a pipette is dropped or mishandled, it can disrupt the accuracy of the pipette. If a pipette is ever suspected to not be dispensing the correct volume, it is recommended to confirm the pipette's accuracy.

10.1. Check Method Using Water

Note: The density of water is 1 g/mL. This means that every microliter (µL) should weigh 0.001 g (or 1 mg).

10.1.1. Perform the daily calibration check on the Analytical Balance, per HSL_EQ_015: Use and Maintenance of an Analytical & Precision Balance.

10.1.2. Fill a 15 mL conical tube with Type II water, and allow water to equalize to room temperature next to the analytical balance and the pipette being tested.

10.1.3. It is recommended the pipette be checked at a minimum of two settings, at the low and high point of the pipette's range; however, ideally a mid-point should be checked as well.

10.1.4. Once the water, pipette and balance are all equilibrated to the same relative temperature, place a weigh boat or weigh paper onto the deck of the balance and press **«→0/T←»** to zero the balance.

10.1.5. Set the pipette to the volume being checked and carefully aspirate the water into the tip, then dispense the water onto the weigh boat or weigh paper. (Note: Weigh paper can be used for small volumes, under 20 µL. Otherwise, a weigh boat should be used).

10.1.5.1. Measure weight of water to one decimal place.

10.1.6. Wait for the weight to stabilize then print the reading. Repeat that two more times, for a total of three weights at a single volume.

10.1.7. Repeat the above steps at the other volumes being checked.

10.1.8. Average the results for each volume together using the following equation:

$$\text{Average} = \frac{Volume\ 1 + Volume\ 2 + Volume\ 3}{3}$$

10.1.9. Determine the accuracy using the following equation:

$$\text{Accuracy} = \frac{|Measured\ Average - Actual\ Volume|}{Actual\ Volume} \times 100$$

10.1.10. The accuracy for a pipette should be less than 2% to be considered acceptable. If a pipette does not meet this requirement, the check should be repeated, preferably by another analyst.

10.1.11. If the pipette fails a second time, remove it from service and give to Scientific Manager to handle further actions required.

10.1.12. Record calibration check in a Laboratory Notebook, include calculations and balance print outs.

11. ATTACHMENTS

11.1. Not applicable.

12. REVISION HISTORY

Revision Start Date	Version #	Changes	Reasons
12Apr17	New	New procedure for the use and maintenance of pipettes.	New SOP.