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Vaccine, Immunity and Cancer Directorate Standard Operating Procedure

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| Author Name | Title | Signature/Date |
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| Approver Name | Title | Signature/Date |
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1. PURPOSE

1.1. The purpose of this procedure is to describe the proper use and maintenance of the pH Meter.

2. SCOPE

2.1. This procedure applies to all pH Meters.

3. REFERENCES

- 3.1. Mettler Toledo SevenCompact pH Meter S220 User Manual
- 3.2. Mettler Toledo Question and Answer on pH Electrodes: https://www.mt.com/us/en/home/products/Laboratory_Analytics_Browse/pHmeter/sensor/pH-sensor.html#sensors_question_1
- 3.3. 10007: Non-Routine Equipment Maintenance
- 3.4. 10009: General Record Review
- 3.5. 26016: Operation, Use and Maintenance of the Water Purification Systems
- 3.6. HSL_GL_001: Waste Disposal at the Advanced Technology Research Facility

4. **RESPONSIBILITIES**

- 4.1. The Research Associate, hereafter referred to as Analyst, is responsible for reviewing and following this procedure, and documenting performance of equipment maintenance.
- 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.
- 4.4. Trained personnel perform equipment maintenance record review per "10009: General Record Review."

5. DEFINITIONS

5.1. As Needed Maintenance – maintenance that is performed outside of routine maintenance but is not performed in response to equipment malfunction.

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- 5.2. Non-Routine Maintenance maintenance that is performed in response to equipment malfunction or failure.
- 5.3. Routine Maintenance maintenance that is performed at planned intervals to identify and prevent problems before they result in equipment failure.
- 5.4. TOC Total Oxidizable Carbon
- 5.5. Type II Water Pure/Analytical Grade, used for standard applications (Resistivity >1 M Ω cm and TOC \leq 50 ppb)

6. REAGENTS, CHEMICALS, AND EQUIPMENT

- 6.1. 150 mL Beaker (Nalgene, Cat # Z130966 or equivalent)
- 6.2. Calibration Buffers pH 4.0, 7.0, 10.0 (VWR, Cat # 97007-790 kit or equivalent)
- 6.3. Electrode, pH, InLab Routine Pro-ISM (Mettler Toledo, Cat # 51344155)
- 6.4. Electrolyte Solution (VWR, Cat # 97007-764 or equivalent)
- 6.5. pH meter (Mettler Toledo, SevenCompact, Cat # 30019028 or equivalent)
- 6.6. Printer, RS-P25 (Mettler Toledo, Cat # 11124300 or equivalent)
- 6.7. Tubes, Conical, 50mL (Warehouse, Cat # 66401493 or equivalent)
- 6.8. Wash Bottle, Standard Spout (VWR, Cat # 414004-226 or equivalent)
- 6.9. Water, Type II
- 6.10. Wipe, No-Lint, Kimwipes (VWR, Cat # 21905-026 or equivalent)

7. HEALTH AND SAFETY CONSIDERATIONS

- 7.1. 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.
- 7.2. Refer to the respective Safety Data Sheet (SDS) when working with any chemicals.

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7.3. Refer to "HSL_GL_001: Waste Disposal at the Advanced Technology Research Facility," "EHS-WM-1: Disposal and Minimization of Chemical Waste," and "EHS-WM-2: Biological Waste Handling and Disposal" for waste disposal processes.

8. GENERAL USE

- 8.1. Press the On/Off key to turn pH Meter on. Press and hold the On/Off key for 2 seconds to turn pH Meter off after use.
- 8.2. pH Meter MUST be standardized/calibrated daily prior to use with three (3) calibration buffers (pH 4, 7, 10). Make fresh aliquots daily of each buffer into labeled 50 mL conical tubes prior to calibration. Calibration buffers can be used in any order.

Note: If the reagent is acidic, and the slope passes during calibration for the pH 4 and 7 calibrators (B1/B2, \geq 93%), but not the pH 7 and 10 calibrators (B2/B3, <93%), then the analyst may proceed with the pH of the reagent. However, if the pH 4 and 7 calibrator slope failed (B1/B2, <93%), then the calibration must be repeated, or pH meter fixed before use. The same scenario holds true for a basic solution. The pH 7 and 10 calibrator slope must pass before use (B2/B3, \geq 93%).

8.3. Always rinse the pH electrode with Type II Water before, in-between, and after measurements. May use Wash Bottle with a standard spout when rinsing electrode. Collect liquid run-off in beaker for proper disposal per "HSL_GL_001: Waste Disposal at the Advanced Technology Research Facility."



Image 1: Rinse Electrode with Type II Water

- 8.4. When blotting electrode dry with Kimwipe, do not dry or touch the bottom/underside of the sensor tip. (See Attachment 1: InLab Routine Pro-ISM pH Sensor)
- 8.5. Place sensor tip as close to the middle of the sample as possible and use a large enough sample container so the sensor tip does not touch the bottom or sides of the container.

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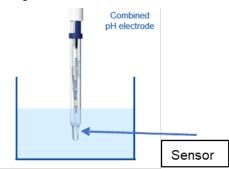


Image 3: Check Electrolyte Level If Needed For pH Electrode Check the electrolyte level



8.7. Verify that the filling hole on the electrode is plugged during storage and use. (See Attachment 1)

Image 4: Ensure Filling Hole is Secured

8.8. Verify that the sensor tip is filled with electrolyte solution and does not have any bubbles. If bubbles are present, flick the sensor until bubbles disappears. (See Attachment 1)

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Image 5: Shake Electrode While Holding Top to Remove Bubbles



8.9. Do not allow external portion of pH electrode to dry out, which will damage the membrane, nor store in Type II Water, which will deplete the ion rich reference electrolyte. Wetting cap, filled with 3 M KCl solution, is to be placed over the end (tip) of the pH electrode during electrode storage and inactivity.

Image 6: Wetting Cap



8.10. The integrated temperature probe measures pH with temperature compensation. See Attachment 4: pH Electrode.

9. EQUIPMENT USE

- 9.1. Standardization/Calibration
 - 9.1.1. Rinse pH electrode with Type II Water then gently blot dry with Kimwipe.
 - 9.1.2. Place electrode in pH 4.0 calibration buffer/standard and press CAL.

Cal 1 appears on the display. The meter endpoints according to preselected endpoint mode after signal has stabilized or after pressing **READ**. The relevant buffer value is shown on the display.

- 9.1.3. Rinse electrode with Type II Water then gently blot dry with Kimwipe.
- 9.1.4. Place electrode in pH 7.0 calibration buffer and press CAL.

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Cal 2 appears on the display.

- 9.1.5. Rinse electrode with Type II Water then gently blot dry with Kimwipe.
- 9.1.6. Place electrode in pH 10.0 calibration buffer and press CAL.

 \rightarrow **Cal 3** appears on the display.

- 9.1.7. Rinse electrode with Type II Water then gently blot dry with Kimwipe.
- 9.1.8. Press **Save** to keep calibration or **EXIT** to reject calibration.

Note: Calibration data will automatically print when pressing **Save**. If it does not print automatically, follow data transfer procedure in section 10.

- 9.1.9. Record calibration information on "26011-01: pH Meter Daily Use Calibration Form" and attach printout.
- 9.1.10. Verify the B1/B2 slope is ≥93% and B2/B3 slope is ≥93%; otherwise, repeat calibration with new pH calibration fluids. If after the second calibration event the slopes fail, regenerate electrode with Electrolyte solution (3 M KCI) and repeat calibration.
- 9.2. Sample Measurements
 - 9.2.1. Rinse electrode with Type II Water then gently blot dry with Kimwipe.
 - 9.2.2. Place sensor in the sample (see Image 2 for placement in container) and press **READ** to start a measurement.
 - 9.2.3. The display shows the readings of the sample. The endpoint format blinks, indicating a measurement is in progress.
 - 9.2.4. As soon as measurement is stable according to the selected stability criterion (Automatic Endpoint), the **Stability** icon appears.

Note: "Automatic endpoint" format is selected, measurement stops automatically once the **Stability** icon appears.

9.2.5. Data automatically prints after measurement is taken. If not, follow data transfer procedure in section 10.

10. DATA TRANSFER PROCEDURE

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10.1. Connect printer cable to pH Meter and the corresponding interface on backside of printer.

Note: Ensure printer's green light is not flashing. A steady green light indicates the printer is synced and ready.

- 10.2. Select interface "**printer**" in data transfer settings menu.
- 10.3. Start transfer in data menu. The data will automatically print.

11. MAINTENANCE

- 11.1. Monthly Maintenance
- 11.1.1. Verify pH electrode is always completely covered by appropriate filling solution (3 M KCl) using the wetting cap and do not allow electrode to dry out.
 - 11.1.2. Verify pH electrode is always kept filled with appropriate filling solution (3 M KCI).
 - 11.1.3. Verify that the filling hole on the electrode is plugged during storage and use. (See Attachment 1)
 - 11.2. As Needed Maintenance
 - 11.2.1. Meter Maintenance
 - 11.2.1.1. Spray cavicide on a low-lint towel and wipe exterior surfaces.
 - 11.2.2. External Electrode Maintenance
 - 11.2.2.1. Ensure pH electrode is always completely covered by appropriate filling solution (3 M KCI) and do not allow electrode to dry out.
 - 11.2.2.2. Rinse pH electrode using Type II Water as needed.
 - 11.2.3. Spills
 - 11.2.3.1. Turn pH Meter off.
 - 11.2.3.2. Spray cavicide on a low-lint towel and wipe exterior surfaces.
 - 11.2.3.3. Use low-lint absorbent wipe to dry components.

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- 11.2.4. Document As Needed Maintenance in its respective section on form "26011-02 pH Maintenance Form."
- 11.3. Non-Routine Maintenance
 - 11.3.1. In the case that the pH Meter is not operating correctly, transition processes being performed to another unit (when applicable), post a sign stating the equipment is out of service and initiate non-routine maintenance documentation per "10007: Non-Routine Equipment Maintenance."
 - 11.3.2. Document the nature of any failures or malfunctions, how and when it was discovered, and the personnel involved on "10007-01: Non-Routine Equipment Maintenance Form."
 - 11.3.3. Initiate a service request and complete the non-routine maintenance process following 10007.

12. SETTINGS

12.1. pH stability reading is set at "Automatic Endpoint."

13. ATTACHMENTS

- 13.1. Attachment 1: InLab Routine Pro-ISM pH Sensor
- 13.2. Attachment 2: 26011-01: pH Meter Daily Use Calibration Form
- 13.3. Attachment 3: How to Detect When a pH Sensor Needs to Be Replaced
- 13.4. Attachment 4: pH Electrode
- 13.5. Attachment 5: pH Maintenance Form

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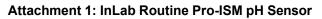
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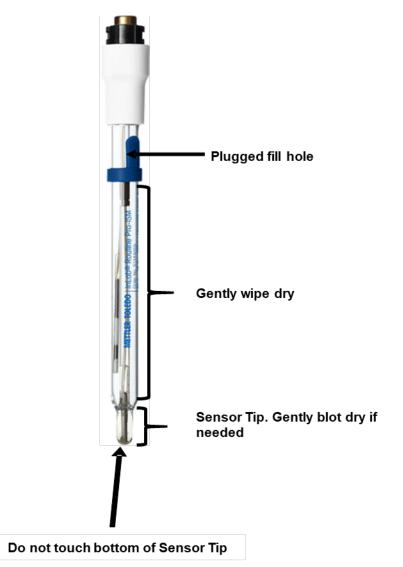
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14. **REVISION HISTORY**

| Revision Start Date | Version # | Changes | Reasons |
|------------------------|-----------|---|-------------------------------|
| 15Mar17 | New | Create new SOP for Use and Maintenance of a pH meter | Currently no SOP |
| 25Mar19 | 2.0 | Updated Maintenance Procedures | Reflect current GDP practices |
| 04Aug21 | 3.0 | Added Images and edited procedure | Clarity |

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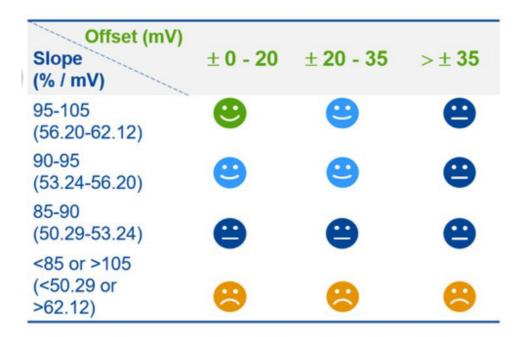
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Attachment 2: 26011-01: pH Meter Daily Use Calibration Form

| | Frede | Frick National Labo for Cancer Re sponsored by the National Ca | | | HPV Serology Laboratory Standard Operating Procedure Form | | | |
|--------------------------|-----------|--|-------------------------------------|---------------------------------|---|---------------------------|------------------------------|--|
| | Form Tit | le: pH Meter Daily Use | Calibration Form | | | | | |
| | Docume | nt ID: 26011-01 | | Version: | | 3.0 | | |
| | Associate | ed SOP: 26011 | | Effective Dat | e: | 12Aug21 | | |
| | Superse | des Version: | 2.0 | | Page 1 of | 1 | | |
| Equipmen | it ID: H | ISL_ | | | | | | |
| Calibration Buffer pH | | Part Number | Lot Number | Expiration Date | B1/B2 Slope (%) (≥93%) | B2/B3 Slope (%) (≥93%) | Comments: | |
| □ N/A 4 7 | | | | | | | ☐ Pass ☐ Fail ☐ Other: | |
| ⊐N/A 10 | | | | | | | | |
| Print out: □ | N/A | | | | | | | |
| | | | | | | | | |
| | Performe | d by/date: | | | | | | |
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| | | N | erify current version prior to use. | Use of a superseded or obsolete | document is prohibited. | | | |
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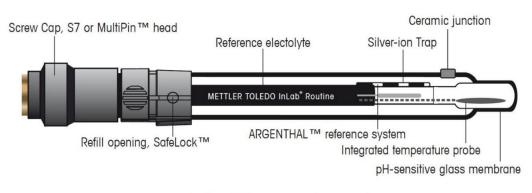
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Attachment 3: How to Detect When a pH Sensor Needs to Be Replaced.



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Attachment 4: pH Electrode



Typical combination pH electrode with inner pH sensor and outer reference element

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Attachment 5: pH Maintenance Form

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|----------|--|---------------------|--------------|--------------------------------|-----------------------|--------------|-------------------------------|-------------|--|----------------------------------|
| | Form Title: p | H Mainter | nance Form | | | | | | | |
| | Document ID | : 26011-0 | 2 | | | ١ | Version: | | 3.0 | |
| | Associated SC | DP: 26011 | L | | | Effe | ctive Date: | | 12Aug2 | 1 |
| | Supersedes \ | Version: | | 2.0 | | | | Pa | ge 1 of 2 | |
| | Maintenance | e Year: | | | | Equipment ID | D: | | | |
| | Monthly Maint | | | - | | | | | | |
| | Month | | nuary | February | | larch | April | | Мау | June U YES |
| | etting Cap filled ed on electrode | _ YES | with 3 M KCI | YES Refilled with 3 M KCI | YES Refilled a | vith 3 M KCl | TYES | | YES Refilled with 3 M KCI | □ Refilled with 3 M KCI |
| Verify e | electrode filled h 3 M KCI | _ YES | with 3 M KCl | YES Refilled with 3 M KCI | □ YES | vith 3 M KCl | 🗆 YES | | YES Refilled with 3 M KCI | YES Refilled with 3 M KCI |
| plug | filling hole is ged during storage | _ YES | | ⊂ YES | U YES | | □ YES | | □ YES | □ YES |
| Perform | med By/Date: | | | | | | | | | |
| Review | wed By/Date: | | | | | | | | | |
| | Month | | July | August | Sep | tember | Oct | ober | November | December |
| | etting Cap filled ed on electrode | _ YES _ Refilled | with 3 M KCI | L YES Refilled with 3 M KCI | ☐ YES ☐ Refilled v | vith 3 M KCl | U YES | ith 3 M KCl | ☐ YES ☐ Refilled with 3 M KCI | ☐ YES ☐ Refilled with 3 M KCI |
| | electrode filled h 3 M KCI | _ YES _ Refilled | with 3 M KCI | C YES Refilled with 3 M KCI | ☐ YES ☐ Refilled v | vith 3 M KCI | YES Refilled with 3 M KCI | | YES Refilled with 3 M KCI | YES Refilled with 3 M KCI |
| plug | filling hole is ged during storage | _ YES | | C YES | □ YES | | TYES | | I YES | □ YES |
| Perform | med By/Date: | | | | | | | | | |
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|---|---------------------|-------------|------------------|------------------|
| Form Title: pH Maintenance For | m | | | |
| Document ID: 26011-02 | | Version: | 3.0 | |
| Associated SOP: 26011 | sociated SOP: 26011 | | 12Aug21 | |
| Supersedes Version: | 2.0 | Page 2 of 2 | | |
| As Needed Maintenance: 🗆 N/A | | | | |
| Date Activity Performed | | | Recorded by/date | Reviewed by/date |

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