Technical Procedure for General Laboratory Equipment

1.0 Purpose - This procedure specifies the required elements for the use of general laboratory equipment.

2.0 Scope - This procedure applies to all pipettes, pH meters, liquid handling systems, electronic balances, and evidence refrigerator/freezers in the Toxicology Units of the State Crime Laboratory.

3.0 Definitions

- **Calibration -** Checking or adjusting (by comparison with a standard) the accuracy of a measuring instrument. Calibrations are performed by approved service contractors for all pipettes, liquid handling systems, and balances in the State Crime Laboratory Toxicology Unit.
- **Quality control (QC) Check -** Periodic confirmation of the reliability of equipment, instrumentation, and/or reagents.
- **Reference standard –** Measurement standard designated for the calibration of other measurement standards (reference standards or equipment).
- **Primary reference standard weights –** Reference standard weights which have documentation issued by an approved vendor authenticating the calibration status.
- **Secondary reference standard weights** – Reference standard weights that have the calibration status verified by comparison to primary reference standard weights.

4.0 Equipment, Materials and Reagents

4.1 Equipment

- Glass Class A pipettes
- Mechanical pipettes
- pH Meter with Electrode - pH combination, double junction, Ag/AgCl reference and thermometer
- Top Loading Balance
- Analytical Balance
- Liquid Handling Systems
- Toxicology refrigerators/freezers

4.2 Materials and Reagents

- Deionized water
- Beakers
- Reference standard weights
- Weighing vessels
- 10 % bleach solution

4.3 Commercial Reagents

- Buffer solutions, pH 4.00 and pH 7.00, and other buffer solution strengths as needed
- 4 M Potassium Chloride saturated solution

5.0 Pipettes

5.1 New Pipettes

5.1.1 A Certificate of Calibration shall be added to FA prior to use.

5.1.2 A performance check shall be performed prior to use. Refer to **5.5**.

5.2 Maintenance

5.2.1 Clean with deionized water as needed.

5.2.2 Prior to returning a mechanical pipette to use (out of service for any reason - e.g., maintenance, malfunction, leaving the direct control of the Laboratory), correct operation shall be demonstrated by a performance check.

5.3 Calibration

5.3.1 Calibration for all Toxicology Unit mechanical pipettes shall be done on a yearly basis by an approved ISO accredited outside vendor. Certificates of Calibration shall be maintained in the Section shared drive on the network.

5.4 Certification

5.4.1 Mechanical pipettes used in procedures that involve quantitative measurements shall be certified.

5.4.2 Glass pipettes used for quantitative measurements shall be Class A. Class A pipettes do not need to be certified annually.

5.4.2.1 Glass Class A pipettes that are damaged or cannot be cleaned thoroughly must be discarded.

5.5 Performance Checks

5.5.1 Mechanical pipettes

5.5.1.1 The Toxicology Technical Leader or designee shall check the calibration every two months gravimetrically using a calibrated balance and deionized water. Results shall be recorded in the **Pipette and Liquid Handling System Logbook**.

5.5.1.1.1 Place a container on the balance and tare.

5.5.1.1.2 For a single volume mechanical pipette:

5.5.1.1.2.1 Dispense the volume of deionized water into the container and record the results in the pipette logbook along with the identifying information of the pipette and the initials of the person performing the calibration check.

5.5.1.1.2.2 Tare the balance and repeat **5.4.1.1.2.1**.

5.5.1.1.3 For an adjustable mechanical pipette, the calibration check shall be performed at both the lowest and highest settings of the pipette:

5.5.1.1.3.1 Dispense the lowest volume of deionized water into the container and record the results in the pipette logbook along with the identifying information of the pipette and the initials of the person performing the calibration check.

5.5.1.1.3.2 Tare the balance and repeat **5.4.1.1.3.1.**

5.5.1.1.3.3 Repeat 5.4.1.1.3.1 and 5.4.1.1.3.2 for the highest volume setting.

5.5.1.1.3.4 If the results are outside of 3 % for volumes greater than 10 µL or 10 % for volumes equal to or less than 10 µL of expected value, clean the pipette and repeat either 5.4.1.1.3.1 or 5.4.1.1.3.3, whichever applies.

5.5.1.1.3.5 If the results are still outside of 3 % or 10 % (whichever is appropriate for the pipette volume being checked), remove the pipette from service and notify the Toxicology Technical Leader or designee.

5.6 Application of Procedure on Evidence

5.6.1 Traditional Method

5.6.1.1 Set and lock desired volume to be pipetted.

5.6.1.2 Firmly attach the pipette tip.

5.6.1.3 Press plunger to the first stop only.

5.6.1.4 Holding the micropipette vertically (±20 degrees), immerse the pipette tip into sample and slowly return the plunger to its starting position.

5.6.1.5 Remove the pipette from the sample and place in its target container.

5.6.1.6 Press the plunger past the first stop to dispense the desired volume.

5.6.1.7 Remove the pipette from the target container and eject the pipette tip into an appropriate waste container.

5.6.2 Reverse Method

5.6.2.1 Set and lock desired volume to be pipetted.

5.6.2.2 Firmly attach the pipette tip.

5.6.2.3 Press plunger past the first stop to the second stop.

5.6.2.4 Holding the micropipette vertically (±20 degrees), immerse the pipette tip into the sample and slowly return the plunger to its starting position.

5.6.2.5 Remove the pipette from the sample and place in its target container.

5.6.2.6 Press the plunger to the first stop to dispense the desired volume.

5.6.2.7 Remove the pipette from the target container and eject the pipette tip into an appropriate waste container.

6.0 Liquid Handling Systems

6.1 New Liquid Handling Systems

6.1.1 A Certificate of Calibration shall be added to FA prior to use.

6.1.2 A performance check shall be added to FA prior to use. Refer to **6.4**.

6.2 Maintenance

6.2.1 The Blood Alcohol Key Operator or designee shall flush the tubing with a 10 % bleach solution, or equivalent, once every three months to remove protein build-up and prevent bacterial growth in the tubing.

6.2.2 Syringes shall be replaced yearly.

6.2.3 Tubing shall be replaced as needed.

6.2.4 Prior to returning a liquid handling system to use (out of service for any reason - e.g., maintenance, malfunction, leaving the direct control of the Laboratory), correct operation shall be demonstrated by performance verification.

6.3 Calibration

6.3.1 New liquid handling systems shall have a Certificate of Calibration added to the Manage Files for the resource in the Resource Manager of FA.

6.3.2 The liquid handling systems shall be calibrated annually by a vendor or contractor and the Certificate of Calibration maintained in the Pipette Calibration Reports folder in the Section shared drive on the network.

6.4 Performance Check

6.4.1 The Key Operator or designee shall check the calibration every two months gravimetrically using a calibrated balance and deionized water. Results shall be recorded in the Pipette and Liquid Handling System Logbook.

6.4.2 Place a container on the balance and tare.

6.4.3 Draw the appropriate sample volume of deionized water into the syringe on the liquid handling system.

6.4.4 Dispense the total volume into the container.

6.4.5 Record the results in the Pipette and Liquid Handling System Logbook along with the identifying information of the pipette and the initials of the person performing the calibration check. This weight shall be within 3 % of the expected value.

6.4.6 Draw the appropriate volume of deionized water from the container and re-weigh the container.

6.4.7 Record the result in the pipette logbook and subtract from the value obtained in **6.4.5**. Record the result of the subtraction. The result will be equivalent to the volume of sample removed in **6.4.3**. This result shall be within 3 % of the expected value.

6.4.8 Repeat steps **6.4.2** thru **6.4.7**.

6.4.9 If the results are outside of 3 %, remove the liquid handling system from service and notify the Key Operator or the Toxicology Technical Leader.

6.5 Application of Procedure on Evidence

6.5.1 Ensure that the liquid to be measured is well mixed using shaking and/or vortexing prior to measuring.

6.5.2 Refer to the appropriate technical procedure for sample preparation instructions.

6.5.3 The liquids being analyzed shall be at room temperature before use.

6.5.4 Liquids containing volatile substances shall be covered or sealed to prevent evaporation of the volatiles.

6.5.5 The reservoir containing the diluent (internal standard) solution shall be covered, but not sealed, to prevent a vacuum from forming in the reservoir.

6.5.6 Place clean wand/pipette tip in sample to be aspirated.

6.5.7 The pipette tip shall remain in liquid during entire aspiration.

6.5.8 Remove wand/pipette tip from sample and remove excess sample residue from wand/pipette tip with a clean Kimwipe.

6.5.9 Dispense sample into proper vial/tube.

6.5.10 Place cap on collection vial/tube and seal.

6.5.11 Discard pipette tip/wash tubing between sample collections by aspirating an air sample and dispense in a waste container.

7.0 Balances

7.1 New Balances

7.1.1 New balances shall be installed and leveled according to manufacturer's specifications. A calibration shall be performed by an outside vendor prior to use.

7.1.1.1 Initial calibration by the vendor shall be documented in Resource Manager in FA by Section personnel.

7.2 Maintenance

7.2.1 Maintain analytical balance level using the air bubble.

7.2.2 If air bubble is not centered, use leveling feet to make adjustments.

7.2.3 Always place weighing sample in the middle of the weighing pan to prevent corner load errors.

7.2.4 Prior to returning a balance to use (out of service for any reason - e.g., maintenance, malfunction, leaving the direct control of the Laboratory), correct operation shall be demonstrated by a performance check.

7.3 Calibration

7.3.1 Calibration for all Toxicology Unit balances shall be done on a yearly basis by an approved ISO accredited outside vendor. Certificates of Calibration shall be maintained in Forensic Advantage Resource Manager.

7.4 Performance Checks

7.4.1 Prior to use, a QC check using two reference standards that bracket the weight of the items of interest shall be performed. Results shall be recorded in the Balance Log book. Acceptance criteria for reference weight: +/-3 % of target weight.

7.4.1.1 If acceptance criteria are not met, balance shall be placed out of service until all necessary steps have been taken to bring the balance back into compliance.

7.4.1.2 Primary Reference Standard Weights

7.4.1.2.1 Primary reference standard weights are maintained by the Drug Chemistry Balance Coordinator.

7.4.1.3 Secondary reference standard weights

7.4.1.3.1 Secondary reference standard weights shall be checked annually, during the month of September, against the primary reference standard set of weights. See the Procedure for Measurement Assurance for requirements of a successful recheck and documentation procedures.

7.4.2 Reference standard weights calibration certificates shall be filed and maintained by the Balances Coordinator for each Laboratory.

7.5 Application of Procedure

7.5.1 Check that balance displays exactly zero at the start of each weighing. Tare if needed.

7.5.2 Tare the weighing vessel.

7.5.3 Place weight in/on the tared weighing vessel.

8.0 Toxicology Refrigerators and Freezers

8.1 New Refrigerators and Freezers

8.1.1 New refrigerators and freezers shall have the temperature checked using a traceable thermometer prior to use.

8.1.1.1 The acceptable temperature range of the refrigerators shall be 2 °C – 8 °C.

8.1.1.2 The freezers shall be at 0 °C or below.

8.2 Maintenance

8.2.1 Any maintenance to the coolant systems must be performed by a qualified refrigeration specialist.

8.2.2 Prior to maintenance, the contents of the coolant system shall be transferred in accordance to **8.4.11.**

8.2.3 Prior to returning a refrigerator/freezer to use (out of service for any reason - e.g., maintenance, malfunction, leaving the direct control of the Laboratory), correct operation shall be demonstrated by a performance check with a NIST traceable thermometer.

8.3 Calibration, Certification and Application of Procedure on Evidence– N/A

8.4 Performance Check

8.4.1 Each refrigerator/freezer in the Toxicology Unit shall have the temperature monitored by a Chemistry Technician III, Evidence Technician, or designee.

8.4.2 The acceptable temperature range of the refrigerators shall be 2 °C – 8 °C.

8.4.3 The freezers shall be at 0 °C or below.

8.4.4 All refrigerators and freezers shall have accompanying daily temperature monitoring devices checked by a NIST traceable thermometer prior to initial use.

8.4.5 Refrigerators/freezers with a manual temperature chart shall have the temperature monitored each working day by a Chemistry Technician III, Evidence Technician, or designee. The chart shall be initialed to signify the temperature is in compliance with the criteria established in **8.4.2** and **8.4.3**.

8.4.6 Automatic temperature charts shall be changed weekly by a Chemistry Technician III, Evidence Technician, or designee.

8.4.7 All charts shall be labeled with the refrigerator serial number, if present.

8.4.8 Records of the temperature monitoring shall be maintained in the FA Manage Files for the resource.

8.4.9 The thermometers monitoring the daily temperature of the refrigerators and freezers shall be checked annually with a NIST traceable thermometer by a Chemistry Technician III, Evidence Technician, or designee. If a refrigerator or freezer is found to be out of compliance with the criteria established in **8.4.2** and **8.4.3**, the Toxicology Technical Leader shall be notified immediately.

8.4.10 Any refrigerators or freezers that are out of tolerance or show any other refrigeration problems shall be reported to the Toxicology Technical Leader or designee, who will evaluate the nature of the problem and determine a solution.

8.4.11 Any refrigerator or freezer found to be out of tolerance or having maintenance performed shall have its contents transferred to a comparable refrigerator/freezer.

9.0 pH Meter

9.1 New pH meter

9.1.1 New pH meters shall have a calibration performed prior to use. Refer to **9.3.**

9.2 Maintenance

9.2.1 Fill the electrode with 4 M potassium chloride saturated solution as needed.

9.2.2 Prior to returning a pH meter to use (out of service for any reason - e.g., maintenance, malfunction, leaving the direct control of the Laboratory), correct operation shall be demonstrated by a successful calibration.

9.3 Calibration

9.3.1 Perform a pH meter calibration daily when in use.

9.3.2 Set the function selector to the pH position.

9.3.3 Obtain two buffer solutions with values that bracket the desired measuring range (i.e., pH 4.00 and pH 7.00 for samples that fall between pH 4 and 7).

9.3.4 Open the fill hole on the electrode.

9.3.5 Place a beaker containing the buffer nearest in value to pH 7 in position and immerse the electrode and thermometer into the solution.

9.3.6 Standardize the pH meter to the buffer.

9.3.7 Remove the electrode and thermometer from the buffer solution.

9.3.8 Rinse the electrode and thermometer with deionized water.

9.3.9 Place a beaker containing the second buffer in position, and immerse the electrode and thermometer into the solution.

9.3.10 Standardize the pH meter to the buffer.

9.3.11 Remove the electrode and thermometer from the buffer and rinse with distilled water.

9.3.12 Close the fill hole on the electrode.

9.3.13 The slope (electrode efficiency) must be greater than 95 %. The calibration may be repeated if necessary to meet the requirement.

9.3.14 Notify the Toxicology Technical Leader or designee if a slope greater than 95 % cannot be obtained.

9.3.15 The Toxicology Technical Leader or designee shall evaluate the instrument and replace the electrode if necessary.

9.3.16 If the problem cannot be corrected by replacing the electrode, the Toxicology Technical Leader or designee shall obtain service or replacement. The instrument shall be placed out of service until a slope greater than 95 % is obtained.

9.3.17 Record all calibrations in the instrument logbook with the date, operator initials, lot number, expiration date and pH of buffers used and the slope obtained.

9.4 Application of Procedure on Evidence

9.4.1 Set the function selector to pH and open the fill hole on the electrode.

9.4.2 Immerse the electrode and thermometer into the sample solution.

9.4.3 Read the pH of the sample from the display and record value.

9.4.4 Remove the electrode and thermometer from the solution.

9.4.5 Rinse the electrode and thermometer with deionized water before proceeding with the next measurement.

9.4.6 Close the fill hole on the electrode.

9.4.7 Record the date, initials and liquid measured in the pH logbook.

10.0 Calculations - N/A

11.0 Uncertainty of Measurement - N/A

12.0 Limitations - N/A

13.0 Safety – Use gloves when dealing with equipment which has been used with blood.

14.0 References - Operator Manuals for equipment used.

15.0 Records

- Temperature control record for Toxicology refrigerator/freezers
- Pipette certificates
- Pipette and Liquid Handling System Logbook
- pH Meter Logbook
- Balance Logbook

16.0 Attachments – N/A

Revision History		
Effective Date	Version Number	Reason
09/17/2012	1	Original Document – Technical Procedure O-01 was converted to ISO Standards.
10/26/2012	2	Inserted 5.2.4.7 for annual temperature check with NIST traceable thermometer; grammar
02/15/2013	3	Adjusted header for consistency with other Toxicology procedures 2.0 - inserted "Toxicology Unit", removed specific location 5.1.2.1.1.4 and 5.1.2.1.1.5 - inserted 10 % range for 10 µL and below Removed 5.1.3 - reference to sampling 5.2.1.1, 5.2.4.3, 5.2.4.4, and 5.2.4.7 - inserted Evidence Technician
05/14/2013	4	5.2.4 - changed 4 (+/- 5˚C) to an acceptable range of 2 - 8 ˚C
05/30/2013	5	5.2.4.1 – inserted ˚C after 2 5.2.4.2 – changed zero to numerical 0 Inserted new 5.2.4.3 – created initial check of refrigerators and freezers New 5.2.4.8 – inserted "thermometers monitoring the daily"
11/15/2013	6	Added issuing authority to header
08/29/2014	7	5.3 - added
03/20/2015	8	The Technical Procedure for General Laboratory Equipment was combined with the Liquid Handling Systems procedure, and the pH Meter procedure 3.0 - definitions added 4.0 - additions made applicable to equipment 5.0, 6.0 - renumbered to fit procedure. 5.6 - Traditional and Reverse pipette method added 6.2.2 and 6.2.3 - Syringe and Tubing requirements added

		7.0 - Balance Section added 8.2 - Maintenance for refrigerators and freezers added 13.0 - Safety – removal of broken pipette and addition of gloves 14.0 - Reference – the references were condensed to state: Operator Manuals. 15.0 – Records - additions applicable to instruments referenced in procedure. 5.1, 6.1, 7.1, 8.1, 9.1 - procedures for new equipment added.