Standard Operating Procedure
for work with

**Chemical name/class:** Osmium Tetroxide  
**CAS #:** 20816-12-0

**PI:** Mark Walters  
**Date:** November 1, 2017

**Building:** Fitzpatrick CIEMAS  
**Room #:** 1562 (Sample Prep Lab)

**Designated Work:** TEM Sample Preparation

1. **Circumstances of Use:**

Osmium tetroxide is a volatile, highly toxic solid that is used as a stain for electron microscopy, and as a fixative for biological samples.

In SMIF, Osmium tetroxide is only used in a diluted liquid form. SMIF receives this chemical as a 4% solution in vacuum sealed vials within secondary containment. It is stored in this fashion in a refrigerator in a secure location where only SMIF staff have access. When needed by SMIF users, SMIF staff further dilutes the osmium tetroxide to a 1% solution in a single 40 mL bottle. SMIF users are only allowed to use the 1% solution out of the 40 mL bottles.

2. **Potential Hazards:**

Consult the Safety Data Sheet (SDS) and the Laboratory Chemical Safety Summary for Osmium Tetroxide from Prudent Practices in the Laboratory (The National Academies Press).

Be aware of these specific hazards:

- Osmium tetroxide is a strong oxidizer that will sublime (pass directly from solid to vapor and back to solid) readily at room temperature and significantly when refrigerated.
- It is highly toxic (LD50 oral [rat] 14 mg/kg) – ingesting very small amounts can cause death. It is also a severe eye and respiratory irritant – acute exposure can cause severe eye damage, even blindness, or chemical burns to the respiratory tract. It can also cause dermatitis or lung or kidney damage.
- The OSHA Permissible Exposure Limit is 0.002 mg/m³, and the ACGIH Threshold Limit Value is 0.0002 ppm over 8 hours or 0.0006 ppm over 15 minutes.
- Chronic exposure to osmium tetroxide can result in accumulation of osmium compounds in the liver and kidney and damage to these organs. Osmium tetroxide has been reported to cause reproductive toxicity in animals; this substance has not been shown to be carcinogenic or to show reproductive or developmental toxicity in humans.

3. **Engineering Controls:**

- Always work with osmium tetroxide in the designated chemical fume hood in the Sample Preparation Lab.
- An eyewash and safety shower are available in the immediate area.

4. **Work Practice Controls:**

- In SMIF, Osmium tetroxide is only used in a diluted liquid form. SMIF receives this chemical as a 4% solution in vacuum sealed vials within secondary containment. It is stored in this fashion in a refrigerator in a secure location where only SMIF staff have access.
- When needed by SMIF users, SMIF staff further dilutes the osmium tetroxide to a 1% solution in a single 40 mL bottle.
- SMIF users are only allowed to use the 1% solution out of the 40 mL bottles.
- Use only in the designated chemical hood that is labeled with the following: DANGER: Osmium Tetroxide in use. Oxidizing Agent, Severe Irritant, Causes Eye Damage, Toxic to Liver and Kidney, Authorized Personnel Only.
- Line work surfaces in the hood with plastic-backed absorbent pads.
- Keep containers closed as much as possible.
- Corn oil is kept in the storage area below the designated chemical hood to use for decontamination and in case of a spill – it deactivates osmium tetroxide.
- Change gloves regularly (at least every two hours) and wash hands at the time of the glove change.
- Wash hands thoroughly immediately after working with any concentration of osmium tetroxide.
- Contaminated items are to be disposed of properly as hazardous waste, following SMIF’s hazardous waste policy.
5. **Personal protective equipment (PPE):**
   - Two pairs of standard nitrile laboratory gloves are recommended.
   - Wear chemical splash goggles (safety glasses are not sufficient). If there is risk of splash, also wear a face shield.
   - Wear a chemical-protective gown with sleeves.

6. **Transportation and Storage:**
   - Solutions must be in sealed shatter-resistant containers, within secondary containment, during storage and transportation. Dry powders are not allowed in SMIF.
   - Osmium tetroxide solutions should be stored in a location that is secure (no unauthorized access).
   - Osmium tetroxide can penetrate plastic, so should be stored in a sealed glass container (such as a vacuum-type blood collection tube), and placed inside a secondary container.
   - Osmium tetroxide should be kept in a refrigerator, and should be stored separately from hydrochloric acid as well as other acids, bases, organic materials, metals, strong reducing agents, and strong oxidizing agents.

7. **Waste Disposal:**
   Unwanted osmium tetroxide solutions and contaminated items must be disposed of following the SMIF chemical waste disposal plan, Duke University’s [Chemical Waste Policy](#) and the [Laboratory Chemical Waste Management Practices](#). These policies cover container management and procedures and timelines for chemical waste pickup.

8. **Exposures/Unintended contact:**
   Contact Employee Occupational Health and Wellness (EOHW) at 919-684-3136 for medical advice on occupational chemical exposures. For an actual chemical exposure
   - Flush exposed eyes or skin with water for at least 15 minutes.
   - If there is respiratory irritation associated with exposure, remove all persons from the contaminated area and contact the OESO spill team.
   - Exposed persons should seek immediate medical attention at the nearest emergency department/
     - Call 911 from a campus phone or 919-684-2444 from any phone to request assistance if needed. Contact Employee Occupational Health and Wellness at 919-684-8115 for exposure-related advice.
   The work-related injury or illness report found at: [http://www.hr.duke.edu/benefits/medical/workcomp/report.php](http://www.hr.duke.edu/benefits/medical/workcomp/report.php) should be completed within 24 hours. Follow-up medical attention should be sought through Duke Employee Occupational Health and Wellness (919-684-3136).

9. **Spill Procedure:**
   In the event of a spill, follow SMIF spill procedures and immediately contact SMIF staff. Only SMIF staff and/or appropriate OESO personnel should clean up spills
   - Contaminated containers and equipment may be decontaminated by dipping in corn oil before removing from the hood. The corn oil will turn black. Paper soaked with corn oil may be used to test if the osmium tetroxide is fully neutralized – if the paper blackens, osmium tetroxide is still present and more corn oil should be added.
   - Contaminated work surfaces may be decontaminated with corn oil or an aqueous solution of sodium sulfite, followed by a cleaning with detergent and water.
   For cleaning up a small spill (<2 ml) of osmium tetroxide solution, cover the spill with corn oil-soaked kitty litter, then scoop up the material and place it in a plastic bag. After spill has been absorbed, wipe down area again with corn-oil, then soap and water solution to decontaminate. Contact OESO Environmental Programs at 919-684-2794 for pick-up.
   Spills of osmium tetroxide powder or spills of solution > 2 ml outside of a chemical fume hood or other enclosure should be referred to the OESO spill response team by calling 911 from a campus phone or 919-684-2444 from any phone.

10. **Training of personnel:**

- All personnel are required to complete the SMIF General Lab Safety session and the SMIF Chemical Safety and Wet Hood training session.
- Training on lab-specific procedures and the hazards of osmium tetroxide is required for all personnel working with these materials, and is documented in the CoreResearch@Duke on-line system.
- All personnel shall read and fully adhere to the laboratory-specific SOP, and shall document that they have read it by signing and dating the SOP.

“I have read and understand this SOP. I agree to fully adhere to its requirements.”

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