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Revision: 2.0	Subject: Vacuum and aspiration equipment	

1. Introduction

Experimental procedures involving biohazardous material often require vacuum pumps or building vacuum systems to help researchers filter reagents and dispose of waste. Such procedures can result in accidental contamination of the pump, vacuum system, laboratory air or plumbing system with biohazardous aerosols or fluids. The purpose of this procedure is to describe the proper set up and safe handling of portable vacuum aspiration equipment used in laboratories to prevent accidental exposure of lab personnel to biohazardous materials and chemicals substances, but also building personnel, and the environment.

2. Scope

This procedure applies to all laboratory personnel – including students, instructional faculty staff, and research teams - using a two-flask aspirator system in their experiments and procedures.

When the procedure is completed using a commercial apparatus, relevant sections of this procedure can be modified by the laboratory supervisor including references to the manufacturer instructions.

3. Applicable Legislation, Standards, Guidelines:

- Ontario Occupational Health and Safety Act -R.S.O. 1990
- Control of Exposure to Biological or Chemical Agents -Regulation 833
- Canadian Biosafety Standards 3rd Edition, 2022 Matrix 3.6.8

4. Responsibilities

4.1 Responsibilities of Directors, Department Heads and Managers

- Ensure that supervisors, employees, and students are notified about the provisions of this SOP,
- Support and assist, when necessary, Principal Investigators to ensure the components of this SOP and the applicable legislation are implemented in all facilities under the Head's authority.
- When necessary, assist the Department of Environmental Health and safety in addressing outstanding non-compliance issues that have been identified in a facility under the Head's authority.

4.2 Responsibilities of Supervisors

- Reviewing this SOP on a regular basis. Review is to consider and mitigate the risks of spill, loss of containment and exposure or other harm.
- Providing the equipment required for use of a two-flask aspirator system if its use is prescribed by the Supervisor.
- Replace or allow replacement of any broken equipment.
- Ensuring that all workers under their supervision are trained on and are proficient in performing the steps of this SOP.

4.3 Responsibilities of Staff and Students

- Following this SOP as approved by their supervisor.
- Reporting any broken equipment immediately to their supervisor.

5. Definitions

Filtration - A filtration operation involves passing a mixture of a liquid with undissolved solids through a filter medium. The filter medium is supported by a funnel or embedded in a filter housing, the exit of which is attached to a collection vessel. The liquid containing the undissolved solids passes through the filtration medium into the vessel below. The liquid that is collected in the vessel is called the FILTRATE. The undissolved solids are trapped on the filter medium. The trapped solid matter is called the FILTRAND. With the right kind of equipment, a vacuum can be drawn on the collection vessel, facilitating faster filtration separation as well as faster drying of the filtrate.

Aspiration - An aspiration operation involves transfer of a supernatant liquid, through suction, away from settled insoluble matter in a mixture. The suction can be applied manually with a hand-operated suction bulb, or with a vacuum system.

Vacuum systems - Vacuum systems create a pressure differential that causes flow from atmospheric pressure side to the vacuum side. When this pressure differential is high enough, a suction effect is created.

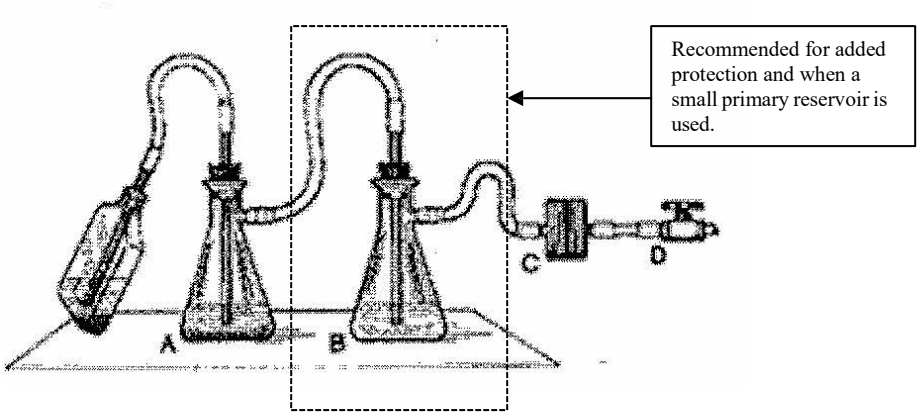

Figure 1.0 Optimal Protection for Vacuum Systems

The above arrangement provides optimal protection for vacuum systems during aspiration. The left suction flask (A) is used to collect fluids into a suitable decontamination solution; [the right flask (B), would be recommended in cases when flask (A) is small and prone to overfilling and serves as a fluid overflow collection vessel--A glass sparger in flask B minimizes splatter].

An in-line HEPA filter (C) is essential to protect the vacuum system (D) from aerosolized materials.

6. Safety precautions

- At a minimum, the vacuum pump or vacuum system must be protected with at **least one primary reservoir and HEPA** filter assembly when the possibility for contamination exists. For an extra margin of safety or when the primary reservoir is a small flask that is prone to overfilling, it is recommended that a two-reservoir system (Figure 1.0) be used.
- Make sure ALL COMPONENTS of the vacuum train are VACUUM RESISTANT, specifically, glassware.
- Use heavy duty vacuum hoses. Make sure tubing for filtration and aspirate extends well below the height of the vacuum port, and don't let the collected liquid reach the exit point of this tubing.
- Glass traps SHOULD BE covered in plastic tape or with a plastic coating to contain accidental implosion. House vacuum systems MUST BE protected from chemicals by in-line HEPA filters.
- Make sure tubing for filtrate and aspirate extends to well below the height of the vacuum port.
- NEVER let the liquid level of filtrate or aspirate get near the height of the vacuum port.
- The filter should have a rated capacity to remove particles 0.2 um or larger. Filters can be either disposable or the type that can be autoclaved for reuse. All filters that are potentially contaminated with biohazardous material should be autoclaved before disposal. Information regarding filters can be obtained by contacting the Department of Environmental Health and Safety.
- Where possible, the secondary containment flask should be placed higher than the experiment so that liquid accidentally aspirated into the flask can drain back to the primary reservoir when the connection to the vacuum line is broken.
- The whole assembly should be placed into a secondary containment tray to prevent accidental release and spill of aspirated solutions onto the floor.
- Inactivation of aspirated materials can be accomplished by placing sufficient chemical decontamination solution into the flask prior to use to kill microorganisms as they are collected.
- Bleach (sodium hypochlorite) is a corrosive agent that can cause severe skin burns and eye damage. Read SDS before using the product. Wear skin, hand, face and eye protection when handling concentrated bleach solution as indicated in the PPE section below. Ensure adequate ventilation. Do not eat, drink, or smoke when using this product.
- WARNING: Some chemicals like phenol, chloroform, Trizol, acetone or xylene can melt plastic and are NOT to be used with aspirator systems. Instead, these chemicals must be collected in a glass container and disposed of as liquid chemical waste. AVOID vacuum filtration of highly volatile liquids.

6.1 Personal Protective Equipment (PPE)

- Hand Protection - Disposable nitrile gloves provide sufficient protection for most routine lab operations. They should be changed if liquid is splashed onto them. Gloves must be inspected prior to use for signs of wear and damage. Such gloves should be disposed of in accordance with appropriate laboratory disposal practices.

- Eye Protection - Use ANSI Z87.1-compliant safety glasses or safety goggles with side shields or tightly

fitting whenever working in the laboratory.

- Skin and Body Protection - Long pants, closed toe/heel shoes, and a lab coat must be worn whenever working in the laboratory. Fully extend sleeves to the wrists and keep always buttoned. Avoid wearing synthetic clothing when practicable.

7. Work practices.

7.1 Before using the vacuum aspiration,

1. Determine the maximum fill volume on your primary flask (80% of total volume). Mark the maximum fill volume on the flask (such as with a marker or label tape).
2. Add a second mark for the level of bleach required to achieve a 10% final concentration when the primary flask will be at maximum fill volume (i.e. 8.3% to 10% final dilution of stock bleach, depending on the concentration of stock bleach - 5% to 6% hypochlorite). (SOP-Biosafety-03 pp. 10).
3. Wearing proper PPE, add fresh bleach in the vacuum primary collection flask and seal the container with the lid or rubber stopper.

7.2 Pre-Use Check

1. Check volume of waste in primary flask. If a risk of overflow is likely, disassemble and clean as described below.
2. Ensure all hose connections are secure.
3. Ensure any slack hose or cord is secured.
4. Ensure any rubber stoppers are on tight.
5. Verify HEPA filter is connected and does not appear to be wet or has changed in colour.
6. Inspect hose before affixing a Pasteur pipette or tip. There may be glass shards present. Use forceps to remove shards and discard in sharps waste.

7.3 Using the aspirator

1. Turn the vacuum pump on only when you want to use the aspirator. Please keep the pump off when not in use.
2. Use a pedal if available.
3. Affix Pasteur pipette or tip to the hose.
4. Aspirate fluids into vacuum primary flask using the aspirating pipette.
5. Change aspiration tips between samples and dispose of as biohazardous waste. In the case of using a glass Pasteur pipette dispose of it in the appropriate sharp container after rinsing with 10% bleach.

7.4 When You Are Done

1. Aspirate 100ml of 10% bleach solution to disinfect the hose.

2. Aspirate 100ml of tap water to rinse the bleach out of the hose.
3. Remove and discard tip into sharps from aspirator hose.
4. Secure aspirator hose.
5. Complete cleanup of the BSC work area.

7.5 Disposing primary flask content

1. After completing your work, check if the primary flask is not full or if the solution has changed color. If any of these conditions are found proceed to empty the primary flask.
2. Biohazardous liquid materials can be disposed of as non-infectious waste after 30 minutes of contact time with 10% bleach. To dispose the content of the flask, ensure that the vacuum pump is not in operation.
3. Wearing gloves and safety glasses or googles, remove all connections to the flask and empty the collector flask into the sink. Always flush the drain with tap water before and after use
4. Rinse the primary flask with plenty of tap water before re-connecting it to the vacuum system or apparatus.

7.6 Maintenance:

- Regularly check the vacuum line for contamination such as bacterial growth or mold. Replace the line, and ensure all users are rinsing the line with fresh 10% bleach after each use.
- Regularly check the online HEPA filter for contamination such as mold growth, Replace the filter before the expiration date (printed on filter) or when contamination is present.

8. Training requirements

- Users of this procedure must have taken WHMIS training as per O. Reg 860.
- No student or lab personnel should operate a vacuum trap before training and receiving their faculty, PI, or lab manager approval.

9. References

- Aspiration Vacuum Flask Set up SOP UMass Amherst Environmental Health and Safety.pdf (umass.edu)
- SOP 001 Two Flask Aspirator System McMaster University. Revised: February 27, 2018
- SOP Vacuum-Assisted (suction) Filtration or Aspiration. California State University.

10. Revision History

Date	Revised by	Changes
January 2006: Initial Release		
2.0 May 2025 - Modifications to procedure.	Raico Lamela, Natalie Roy	Content and formatting. Safety precautions, step procedures and maintenance requirements.

11. Appendix

N/A