

# SHAROM LABORATORY REGULATIONS AND SAFETY TRAINING RECORD

Department of Molecular and Cellular Biology  
University of Guelph  
New Science Complex Room SCIE 2206

In case of emergency,  
call the University Emergency Phone Line × **52000**  
and contact Dr. Sharom at home (824-2712)

Please keep this memorandum for future reference.

It is also posted at <http://www.uoguelph.ca/~fsharom/>

If you are in doubt about any procedure, please ask for advice before proceeding.

## Environmental Health and Safety (EHS) Department

The Home Page of EHS Department is: <http://www.uoguelph.ca/ehs/>

The EHS Department provides technical information on laboratory hazards and biohazards and advice on how to clean up, inactivate, or dispose of hazardous materials.

<b>Director</b>	Chris White	×53101
<b>Hazardous Materials Safety Officer</b>	Jennifer Minogue	×53190
<b>Hygiene Safety Officer</b>	Dal Persaud	×54855
<b>Occupational Health and Wellness Administrator</b>	Giselle MacNeil	×52133
<b>Radiation Safety Officer</b>	Domenico Barillari	×54888
<b>General Enquiries</b>	Lisa Kohlmeier	×53282
<b>Agricultural / Industrial Safety Officer</b>	Christi Cooper	×52049

The University of Guelph “**Safety Policy Manual**” is available on-line at:

<http://www.uoguelph.ca/hr/ehs/policies/06-10.pdf>

The major topics include: Personal Protective Equipment, Emergencies, Radiation Safety, Biosafety, Fire Safety, Safe Work Procedures, Physical Agents, Incident Reporting and Investigation, Hazardous Materials, Non-Hazardous Waste Management and Occupational Health.

## Department of Molecular and Cellular Biology

<b>Chair:</b>	Dr. Whitfield	SCIE 1252	×53361		
<b>Safety Officer:</b>	Dr. Preston	SCIE 3247	×53478		
<b>Radiation Safety:</b>	Dr. Preston	SCIE 3247	×53478		
<b>Safety Committee contact:</b>					
Dr. Preston	SCIE 3247	×53478	Teresa Partridge	SCIE 3206	×54385
Dr. Mangroo	SCIE 2244	×53432	Gerry Prentice	SCIE 2204	×58163
Deb Flett	SCIE 4113	×54328	Paula Russell	SCIE 3115	×58220
Karen Ingram	SCIE 1203	×53816	Michael Skalski	SCIE 2203	×58181
Jaspreet Kaur	SCIE 3115	×58220	Leona Varga	SCIE 1255	×56246

The Departmental Safety Committee meets to discuss safety issues on a regular basis. If you wish to discuss any aspect of departmental safety, contact a member of the committee.

The Department of Molecular and Cellular Biology provides a manual “**Standard Operating Procedures**” for:

- (1) Autoclaves
- (2) Centrifuges and Ultracentrifuges
- (3) French Press
- (4) SpeedVac Lyophilizer
- (5) Bio-Rad Gel Doc System
- (6) Liquid Scintillation Counters
- (7) Liquid Nitrogen-dispensing into Dewars
- (8) Handling, Use, and Storage of Compressed Gas Cylinders

## 1. GENERAL LAB RULES AND PERSONAL PROTECTION

- 1.1 All research laboratories should be considered as secure areas. All doors must be kept **closed** at all times, and kept **locked** when the laboratory is empty. Persons who are not lab members are permitted entry only in the company of authorized personnel.
- 1.2 NO eating, drinking, smoking, chewing gum or applying cosmetics are allowed in the lab at any time. No storage of food or eating utensils in the laboratory.
- 1.3 All new research personnel are required to attend a WHMIS (Workplace Hazardous Materials Information System) safety training session. Consult with the Departmental Chair Office (see Sandra Good in SCIE 1253) or the EHS Department (×53282) to find out when the next training session will be held.
- 1.4 Lab coats are recommended for everyone working in labs, particularly if you are wearing shorts and/or tank tops.
- 1.5 Proper footwear is mandatory. Do not wear open toed shoes, sandals or canvas topped shoes in labs or workshops. Corrosive reagents can easily drip onto unprotected toes.
- 1.6 Long hair is a safety hazard, and must be tied back.
- 1.7 Disposable gloves should be worn whenever chemicals are handled, and removed as soon as they become contaminated. Gloves must be removed before leaving the lab. You may be contaminating others by wearing gloves in the halls, touching doorknobs, telephones, *etc.*
- 1.8 Safety goggles and a face shield should be worn if there is a possibility of explosion, spattering of acids or bases, chemicals, *etc.* Face shields must be worn over other protective eyewear.
- 1.9 Cover all work surfaces with absorbent paper for easy clean-up and decontamination. Keep the laboratory area CLEAN and ORGANIZED.

- 1.10 Wash your hands regularly, especially before eating or leaving for home. This is an effective way of minimizing the spread of chemicals and biologicals out of the laboratory.
- 1.11 Safety Manual and Charts are available in every laboratory. Take the time to read through and familiarize yourself with the safety precautions relevant to your work.
- 1.12 Hazardous Chemicals are listed on the “**Chemical Inventory**” in each lab. Read the Material Safety Data Sheets before using hazardous chemicals. Make sure the Laboratory Chemical Inventory and HAZCHEM Web are kept up-to-date.
- 1.13 Spill kits with acid/base neutralizers and absorbent pads are kept in the cupboard under the sink in the lab (SCIE 2206).
- 1.14 Know the location of safety shower, eye-wash station and fire extinguisher in the lab. Also know the location of fire blanket, the nearest fire alarm and the nearest exit from the building.
- 1.15 Each lab is equipped with a small First Aid Kit. Some certified first aiders are currently listed in the Department of Molecular and Cellular Biology.

<b>Certified First Aid Personnel</b>		
Catrien Bouwman	SCIE 4202	×54710
Laleh Hatefi	SCIE 1251	×53362
Karen Ingram	SCIE 1203	×53816
Leanne Krick	SCIE 4104	×53365
Leona Varga	SCIE 1255	×56246

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Leona Varga	SCIE 1255	×56246

## 2. “AFTER-HOURS” AND “WORKING ALONE” PROCEDURES

- 2.1 The University of Guelph general policy and relevant procedures on working “**After Hours**” (*i.e.* before 8:30 am, after 5:00 pm, or on weekends and holidays) is posted on-line:  
<http://www.uoguelph.ca/hr/ehs/policies>
  - (i) Personnel are strongly encouraged to work during regular hours (8:30 a.m. - 5:00 p.m.) and *not to work alone*. If you are working outside of these hours or on weekends, you must notify someone of your whereabouts and your expected time of return. (You can notify the Campus Police - Security Service, at ×52245, if you wish.)
  - (ii) Campus SafeWalk (×53200) can accompany you across campus after dark.
- 2.2 The University of Guelph policy on “**Working Alone**” is documented in detail and may be viewed on the University of Guelph web-site:  
<http://www.uoguelph.ca/hr/policies/06-06.pdf>

Working alone is not the same as working after hours, although in principle, those working out of regular hours should adhere to the same general procedures. The Safety Committee of the Department of Molecular and Cellular Biology draws attention to the following points in relation to “Working Alone” policies applicable to this department.

- (i) Graduate students or employees working after hours should pair up with another individual who is physically close by, although not necessarily working in the same room. Pairs not working in close proximity should periodically check in with each other. Pairs should have a mutually agreed quitting time, or if one party leaves early, inform the other member of the pair. Persons truly working alone should check in and check out with University of Guelph Police - Security Service by phone (×52245).
  - (ii) Undergraduate students are not permitted to work alone.
- 2.3 Do **not** let unauthorized people into the building **after hours** by holding doors open for them when you leave. Politely tell them that college security regulations prevent you from letting them in unless they have a key. Students should be entering via the main doors in the New Science Complex.
- 2.4 Personal safety of the members of the Department of Molecular and Cellular Biology is of paramount importance. Most entrances to the New Science Complex building are secured from 4:30 pm to 8:30 am every day unless a special arrangement has been made. Under no circumstance should doors be “wedged” open after hours or on the weekend.

### 3. MATERIALS HANDLING PROCEDURES

The brochures “**Biohazard Safety Guidelines**” and “**Guidelines for Use of Radioactive Isotopes** (April 1990)” are available in the lab. These are provided by EHS Department, and should be consulted, where appropriate.

- 3.1 Wear disposable gloves whenever hazardous chemicals are handled, and remove them as soon as they become contaminated. Nitrile gloves generally provide superior protection to vinyl or latex gloves. Remove and discard gloves appropriately (biohazard, radioactive waste, *etc.*) when the need for them is over.

Do not wear gloves **outside** the laboratory. Do not touch the door handles, telephones and computers with the gloves on.

- 3.2 Two vented solvent storage cabinets under the fumehoods are available in SCIE 2206. Large quantities (litres or more) of flammable and/or volatile solvents must be stored inside these cabinets. Note that the amount of ventilation provided by these cabinets is limited, as they are meant to protect the contents from external fire and explosion rather than vent fumes. Also note that there is a limit of 50 L of flammable liquids per lab; no more than 5 L total outside the cabinet.

Organic solvent use must be confined to fumehoods, to the maximum extent practical. Organic solvent bottles can develop pressure on hot days in the summer, so the tops should be left slightly loose. Make sure no flames (bunsen burner) are on while you (or others near you) are using flammable solvents. Bunsen burner flames are nearly invisible and should be turned off when not in use. If you smell gas, inform other people in the lab and look for a source.

- 3.3 Handle hazardous chemicals carefully in the fumehood. Chemical fumehoods provide protection from the toxic fumes emitted by some hazardous materials. Proper functioning requires that the sash be lowered to approximately six inches from the apron and that the fan is turned on. When working in a fumehood, keep movement in and around the hood to a minimum. Keep clutter away from the air slots at the back of the fumehood to ensure an adequate level of air flow. Check the flow alarm regularly.

Always keep the fumehood tidy and spacious for working. Note that one fumehood is designated for radioactive handling. Do not use it for storage.

- 3.4 If you are using any compressed gases, read the departmental manual “**Standard Operating Procedure for Handling, Use, and Storage of Compressed Gas Cylinders**”.

Compressed gas tanks can be extremely dangerous if they are knocked over and the valve assembly is broken off. Because the gas in the cylinder is under pressure, the tank will blow through several concrete walls killing anyone in its path. For this reason, gas cylinders must be securely clamped to a wall or bench. When not in use, or when in transport, the valve cover must be in place. This will protect the fragile valve stem should the tank fall over. Use cylinder carts at stockroom when transporting gas tanks from place to place. Make sure you are instructed in the proper method of handling gas cylinders and changing a cylinder regulator.

- 3.5 When handling radioactive materials, wear disposable gloves and obey the rules and regulations for safe and responsible laboratory activity. Use shielding, absorbent papers, absorbent jug, Decon detergent and fumehoods when appropriate. Monitor contamination with a Geiger counter and wipe tests measured in a liquid scintillation counter.

Users of radioactive isotopes **must** have appropriate training and must be registered through the EHS Department. Contact Lisa Kohlmeier at ×53282 for detailed information. All research projects using radioisotopes must be approved by the University Radiation Safety Committee. The Radioisotope Project includes an inventory of all radioactive substances held at any time in the laboratory. It also includes instructions for the disposal of radioactive waste. All users should familiarize themselves with the specifics of their Radioisotope Project as well as the university “**Guidelines for the Responsible Use of Radionuclides**”. The University is licensed by the Canadian Nuclear Safety Commission (CNSC), and CNSC inspectors can inspect laboratories at any time.

- 3.6 Pay extreme care and attention when handling electrical equipment. Always read the appropriate instruction manuals thoroughly prior to using the equipment. Get help from experienced users, if necessary.

Many separation technologies employ electrical conditions that are lethal if they should short through the experimenter. It is current that kills, not voltage, and currents in the 100 to 200 mA range (blotting conditions and some large agarose gels) are absolutely lethal. You should never use any form of electrical equipment unless it is equipped with safety interlocks. Interlocks prevent accidental contact with electrically “live” surfaces unless the power has been disconnected. All commercial equipment has these interlocks.

Other good working practices involve turning off the power, keeping benches and floors dry, and keeping one hand in your pocket while disconnecting leads. (Electrical shocks can cause the victim to involuntarily clamp a hand onto a grounded surface. If they have one hand clamped onto a live source and the second onto a ground, this will cause the current to short through their heart.) Modern power supplies are also equipped with ground-fault detection circuitry. This shuts off the power should the machine sense a short circuit through an external ground (*i.e.* you). Where possible, older power supplies should be replaced with new ones. Do not use any electrical equipment that has frayed wires or loose connections.

#### 4. HAZARDOUS CHEMICALS USED ROUTINELY IN OUR LAB

Hazardous chemicals are listed on the “**Chemical Inventory**” in the lab (SCIE 2206). Make sure the laboratory inventory is kept up-to-date. It is good practice to read through the appropriate section of Material Safety Data Sheets describing any compound or chemical that is new to you before use. The following chemicals are used regularly in the lab. Their principal hazard, primary treatment, primary protection, relevant safety apparatus and comment are listed.

##### 4.1 Acids

**Typical examples:** HCl, HNO<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, H<sub>3</sub>PO<sub>4</sub>. **Principal hazard:** Concentrated acids can blind if splashed into eyes. They can also create corrosive burns on the skin. **Primary treatment:** Irrigate affected tissue extensively with plenty of water for at least 15 minutes. Consult a doctor if splashed in the eyes or if the burn warrants medical attention. **Primary protection:** Dispense acids in the fumehood. Wear gloves, aprons, and goggles when dispensing. **Relevant safety apparatus:** Gloves and goggles should always be accessible. Eye-wash fountains, in good working condition, must be nearby. Absorbent pads are available in the lab to clean up spills. **Comment:** Buy in plastic coated bottles. Segregate away from bases. Always add acidic solutions to basic solutions and water, NOT *vice versa*.

##### 4.2 Acrylamide

**Typical examples:** Acrylamide and bis-acrylamide. **Principal hazard:** Slow acting neurotoxin, can cause contact dermatitis. **Primary treatment:** Wash off spilled material with plenty of soap and water. **Primary protection:** Wear gloves and a mask when dispensing. Clean up spilled powder. Pour electrophoresis gels on absorbent paper. **Relevant safety apparatus:** Disposable gloves and masks. **Comment:** Much safer when polymerized, however, some monomer is left behind after polymerization. Premixed solutions are available and are much safer than powder. Precast gels are available for most electrophoresis systems.

##### 4.3 Bases

**Typical examples:** NaOH, NH<sub>4</sub>OH, KOH. **Principal hazard:** Concentrated bases can blind if splashed into the eyes. They also cause corrosive burns on the skin. Ammonia can burn lung tissue if inhaled. **Primary treatment:** Wash affected tissue extensively with plenty of water for at least 15 minutes. See a doctor if splashed in the eyes or if burn warrants medical attention. **Primary protection:** Dispense bases in the fumehood. Wear gloves, aprons, and goggles when dispensing. **Relevant safety apparatus:** Gloves and goggles should always be worn. Eye-wash fountains, in good working condition, must be nearby. Absorbent pads are available in the lab. **Comment:** Segregate away from acids. Always add acidic solutions to basic solutions, NOT *vice versa*.

4.4 **Chromic-Sulfuric Acid Cleaning Solution**

**Principal hazard:** Corrosive oxidizer, Fire hazard. **Primary treatment:** Flush skin or eyes with plenty of water for 15 minutes. **Primary protection:** Do not get on eyes, skin or clothing. Do not breathe mists. Store in a tightly closed container. Use with adequate ventilation. Remove and wash contaminated clothing promptly. Wash thoroughly after handling. Do not store near combustibles. **Relevant safety apparatus:** Wear gloves and work in fumehood. **Comment:** This is a dangerous chemical solution prepared by combining chromic acid and concentrated H<sub>2</sub>SO<sub>4</sub>. Handle with extreme care.

4.5 **Dimethylsulfoxide (DMSO)**

**Comment:** DMSO, in the pure state, is a relatively harmless chemical. However, it greatly increases the transport of dissolved chemicals through the skin barrier and thus potentiates the hazard of any dissolved compound. DMSO also permeates latex gloves, use neoprene or natural rubber gloves when using DMSO as a solvent for hazardous (poisonous or mutagenic) chemicals.

4.6 **Dry Ice (Frozen Carbon Dioxide)**

**Principal hazard:** Asphyxiation, skin burns (frost bite). **Primary treatment:** In cases of asphyxiation, remove victim to fresh air and apply artificial respiration. See a doctor immediately. Treatment for burns depends upon severity. **Primary protection:** Wear insulating gloves when handling dry ice. **Relevant safety apparatus:** Insulated gloves. **Comment:** The sublimation of dry ices creates CO<sub>2</sub> gas. In an enclosed space, with limited air circulation, this can asphyxiate. For this reason, all parcels containing dry ice must be labelled so that transport companies are aware of the potential hazard to other cargoes. Do not place dry ice shipments in the walk-in Cold Room Suite (SCIE 2226). Make sure that the CO<sub>2</sub> container allows for escape of the CO<sub>2</sub> gas to avoid risk of rupture.

4.7 **Ethanol**

**Principal hazard:** Fire, toxicity. **Primary protection:** Ethanol is kept in Solvent Storage Room on the first floor, which is in charge by Karen Ingram in SCIE 1203. A detailed log of ethanol usage is kept at Store Room by Karen Ingram. It is subject to periodic inspection by Canada Customs and Excise. **Comment:** To reduce the fire hazard, do not keep more than 4 L of ethanol in the laboratory at any one time. For your convenience, ethanol is provided in 4 L plastic bottles. Some high purity grades of ethanol contain traces of benzene because it is purified by distillation as a benzene/ethanol azeotrope. Do not drink “*Ultrapure*” ethanol as benzene is highly toxic.

4.8 **Ethidium bromide**

**Principal hazard:** Mutagen. **Primary protection:** Disposable gloves. Measure out or dispense in fumehood. **Comment:** Solutions of ethidium bromide should be sent to EHS Department for disposal.

4.9 **Formaldehydes**

**Typical examples:** Paraformaldehyde, formaldehyde, glutaraldehyde. **Principal hazard:** Carcinogen, dermatitis, allergen. Fumes can “fix” corneal tissue. **Primary protection:** Avoid contact by using latex or nitrile gloves and wear goggles. Work in a fumehood. **Relevant safety apparatus:** Disposable gloves, fumehood. **Comment:** These agents are typically used as fixatives. Formaldehyde gels are also used to size fractionate RNA. Since formaldehyde is volatile, solutions should always be handled in a fumehood. Gels should also be run in a

fumehood. Note that long-term exposure can cause severe contact dermatitis.

#### 4.10 **Liquid Nitrogen**

**Principal hazard:** Burns (frost bite), asphyxiation, implosion hazard (Dewar flasks). **Primary treatment:** In case of asphyxiation, remove the victim to fresh air and apply artificial respiration. Burn treatment depends upon severity. **Primary protection:** Insulated gloves, face shield and goggles. **Relevant safety apparatus:** Insulated gloves, ventilated working area. Dewar flasks must have a protective shell or be taped. Do not use “Thermos” bottles from hardware stores.

#### 4.11 **Methanol**

**Principal hazard:** Fire, toxicity. **Primary protection:** Avoid breathing vapours and skin contact. Wear disposable gloves. Store large volumes (greater than 4 L) in the yellow vented organic solvent cabinet in the lab. **Relevant safety apparatus:** Gloves, fumehood. **Comment:** Methanol vapours are heavier than air. Sparks or open flames can cause ignition and flashback if it is not stored properly. Methanol is easily absorbed through the skin, thus increasing the toxic hazard.

#### 4.12 **Organic Solvents**

**Principal hazard:** The low flash point of most organic solvents results in a risk of fire if they are exposed to an open flame or spark. As volatile compounds, they present a significant risk associated with inhaling the vapours. **Primary protection:** All work with solvents must be performed in a working fumehood. Cap or enclose vessels containing organic solvents. Clean up spills promptly. Store, closed, in a yellow vented chemical storage cabinet.

#### 4.13 **Phenol**

**Principal hazard:** Chemical burns. Death can result from contact over a large amount of skin area. **Primary treatment:** Wash off spilled material with plenty of soap and water. Contact a doctor immediately if the spill covers an extensive area of skin. **Primary protection:** Wear gloves. Apron and safety goggles should also be worn when distilling phenol. Work in a fumehood. **Relevant safety apparatus:** Gloves, apron, safety goggles, safety shower. **Comment:** Phenol causes unpleasant burns that may not be noticed until next day. This is because it numbs the skin as it burns. Look carefully for whitish patches of skin if you suspect you have spilled phenol on yourself. Phenol is absorbed through the skin and has killed people following large spills. If you should pour phenol over a large area of skin, wash it off immediately and have someone call a doctor.

#### 4.14 **Silane (Dimethyldichlorosilane)**

**Principal hazard:** HCl vapour is released upon reaction with glass surfaces and water. **Primary protection:** Wear protective gloves and work in a fumehood. **Relevant safety apparatus:** Gloves, working fumehood. **Comment:** Dimethyldichlorosilane is used to coat glass and plastic surfaces, thus reduces the non-specific adsorption of macromolecules. It should be handled with care as the silanizing reaction releases HCl vapour. Traces of residual dimethyldichlorosilane can be inactivated with copious quantities of water, but do not mix the silane concentrate with water. An explosive release of HCl may occur.

4.15 **Silver Nitrate (AgNO<sub>3</sub>)**

**Principal hazard:** Strong oxidizer. May form explosive material when mixed with ammonia or methanol, especially when dry. **Comment:** AgNO<sub>3</sub> is a component of silver staining protocols for electrophoresis gels. Used solutions should not be stored in the lab for long periods as shock-sensitive silver compounds (silver fulminates) can accumulate on standing. Used silver-staining solutions must be disposed of as hazardous waste by EHS Department.

4.16 **Trichloroacetic acid (TCA)**

**Principal hazard:** Blindness, chemical burns. **Primary treatment:** Wash extensively with water. See a doctor if splashed in the eyes or if the burn warrants medical attention. **Primary protection:** Wear eye-protection and disposable gloves. **Relevant safety apparatus:** Disposable gloves and protective goggles. **Comment:** TCA, like phenol, numbs the skin as it burns. Burns may not be obvious until next day.

## 5. WASTE DISPOSAL PROCEDURES

Many types of waste are generated in laboratories, and each must be disposed of by appropriate procedures. *Your work is not completed until all the waste materials are safely disposed of.*

5.1 Warning tapes (such as Autoclave, Biohazard, Corrosive, Radioactive Toxic, *etc.*), warning tags (*e.g.* Hazardous Chemical Waste), caution signs (such as Emergency, Fire Hazard, Radioactive Materials; Experiment in Progress *etc.*) and labels (*e.g.* Workplace Label, Fire Hazard Label *etc.*) are available in the laboratory and should be used as appropriate.

5.2 **Solids:** Regular garbage is deposited in the waste cans. Note that the lab garbage is sorted for “**Garbage Only**” (black bag) and “**DRY - Recyclables Only**” (clear bag). Broken or intact glass must be deposited in the white plastic pails containing yellow bags, labelled “**Glass Only**” to avoid injury to housekeeping personnel as they empty garbage. “**Sharps**” (*e.g.* needles) are placed in the bright-red sharps containers. Sharps or yellow containers are autoclaved before pick up for disposal by EHS Department.

5.3 **Solvents:** Large red safety can containers are available for each of: (i) non-halogenated solvent waste; (ii) halogenated solvent waste (*e.g.* chloroform); and (iii) radioactive solvent waste (*e.g.* scintillant waste). Aqueous waste may be stored in glass or plastic jugs. A record sheet is kept for each container, and any additions of more than a few millilitre should be recorded. As containers become full, a disposal form (**Request for “Sharps” and Waste / Surplus Chemical Disposal**) must be filled out, signed by the Laboratory Supervisor, and forwarded to the EHS Department. An attached warning tag for identification of the waste is required for each disposal. Hazardous waste is picked up regularly on Friday mornings by the EHS Department for disposal.

*Do not allow large inventories of waste solvent to accumulate!*

- 5.4 **Biohazard waste:** Biohazard-contaminated materials (*e.g.* Petri dishes, cultures) must be collected in appropriate containers (*e.g.* orange autoclave bags for solids), and autoclaved when full. Close autoclave bags **loosely** with a piece of autoclave tape. Solid waste bags should be autoclaved for 60 minutes in “Gravity” cycle. All materials must be placed in an autoclavable plastic or metal tray to prevent spillage from contaminating the autoclave. After sterilization, the biohazard bags are placed in the garbage can (black bag). Alternatively, bring the waste to Leanne Krick (Prep Room SCIE 4104) before noon for disposal.
- 5.5 **Radioactive waste:** A copy of the valid radioisotope permit issued by EHS Department is required before any radioactive experiments can be carried out in the laboratories. If you intend to use radioisotopes, you must contact Domenico Barillari, EHS Department (×54888) and familiarize yourself with the appropriate handling procedures. A training session is mandatory for any new radioisotope users. Follow the radioactive waste guidelines, **UG-ORSOG-101**.
- (i) For **solid** radioactive waste, wrap inside bench paper and place it in a plastic bag, then bring the radioactive waste to the Radioactive Preparation Room (MacN 229) for disposal. Note that there are two different types of radioactive waste bins, which are separated as “<sup>32</sup>P” or “\*NO\* <sup>32</sup>P”. When full, call the EHS Department at ×53282 for weekly disposal. The regular pick-up time is scheduled on Friday mornings.
  - (ii) For **liquid** radioactive waste, there are a few large red safety can containers (7.6 L size) available in the lab for disposal. Note that the radioactive waste containers are separated as <sup>3</sup>H & <sup>14</sup>C, <sup>125</sup>I and organic waste. Fill out a “**Request for Radioactive Waste Disposal**” form, obtain the signature of the Laboratory Supervisor, then forward to the EHS Department. Place a fully labelled warning tag on each radioactive waste container for disposal.

We are required by the conditions of our radioisotope licence to monitor the lab for radioactive contamination on a weekly basis. Note that one of the two fumehoods in SCIE 2206 is **only to be used for working with radioactive materials**. The other fumehood in SCIE 2206 is reserved for non-radioactive work, such as TLC, solvent handling, *etc.*

Note that the Radioactive Preparation Room (MacN 229) is available for work with larger quantities of radioactive materials (see below). Anyone who uses this room must follow the safety rules exactly.

- 5.6 Waste containing strong acids or bases should **NEVER** be mixed with each other **OR** with organic solvents. This is the **most common cause** of laboratory fires and explosions. It is illegal to dilute acids or bases with copious amounts of water and pour them down the sink. Large quantities of acid or base waste should be stored in separate containers and disposed of by the EHS Department. Small quantities (100 mL or less) can be neutralized and put to drain as long as the neutralized solution is non-toxic (*e.g.* no heavy metals).
- 5.7 Samples stored in the refrigerator, freezer, or cold room must be clearly marked with initials of the user, description of contents, and date.
- (i) Sample vials should be stored in sealed “Tupperware”-type or Nalgene (screw-cap) containers.

- (ii) Petri dishes must be sealed with Parafilm or in a securely-tied plastic bag, such as the plastic sleeves in which the dishes are shipped.
- (iii) Solutions, buffers, *etc.* stored in glass or plastic bottles or flasks must be labelled. Label the containers by writing with permanent marker on a piece of coloured labelling tape. Stick the tape to the bottle, opposite to the measuring scale, folding over the end so that the tape can be removed easily. Do **not** mark on the white frosted areas, as marker cannot be removed from those areas.

*Unmarked samples may be disposed of without notification.*

- 5.8 Disposable gloves are meant to protect the wearer from hazardous substances and to protect sensitive work from contamination. They are, by definition, contaminated as soon as they are worn. Remove and discard gloves appropriately (biohazard, bypass, radioactive waste *etc.*) when the need for them is over. Do not allow them to come into contact with any surface that would pass on the contamination to other workers or yourself via door knobs, telephone, fridge, computer keyboard *etc.*

## 6. USE OF COMMUNAL EQUIPMENT ROOM (SCIE 2202)

Most of our equipment is shared voluntarily. Remember that a faculty member may have sweated blood to get funding for the instrument from NSERC, CIHR, or NCIC. They will clearly not be thrilled when it is abused by users. When a key instrument, such as a counter or centrifuge, is down for repair, many labs will be unable to carry out experiments. If you are responsible for damage caused by carelessness, your supervisor (*i.e.* Dr. Sharom) may be asked to pay for it from her research grant.

Before using a piece of equipment with which you are unfamiliar, *always* consult a senior member of your research lab or the person in charge of the instrument for instructions. Contact a faculty member in the Department of Molecular and Cellular Biology for any questions on the communal equipment such as autoclave, centrifuges, counters, Super-Q water system, *etc.* (See below for further instructions regarding Departmental Biochemistry Facilities). If the person is **not** a member of the department, he/she must sign a “Request to Use Equipment” form to accept responsibility for costs/damage to equipment.

As a user of the communal equipment, you must obey the following rules:

- 6.1 Sign the log book **immediately before** beginning to use the instrument. If there is a problem during your run, how will anyone know who to contact if you haven't signed in yet? Centrifuge Log is especially important to keep track of hours.
- 6.2 Use full names (no initials), contact phone number and lab location on logs and scheduling sheets. If there is a problem with your run, how will anyone find you? Also, research labs are billed for the use of some equipment, such as use of fluorimeters, particle sizer, imaging system and plate readers, so someone has to be able to read your name, and the lab you work in, at a much later date when billing is being done.

- 6.3 Inspect the instrument before and after use. If you notice something odd or different before you start, DO NOT SIMPLY GO AHEAD, you will probably damage the machine; *always* consult a senior member of your research lab or a biochemistry faculty member.
- 6.4 Always remember to turn equipment off. Do not leave any instrument on all weekend.
- 6.5 Leave the work area clean. Take your paper towels, pipette tips, tissues, gloves, microtubes, scintillation vials, *etc.* away with you, or dispose of them (sorted correctly) in the garbage. Housekeeping will not clean up after you, so if you don't, who will?
- 6.6 The following section describes standard procedures for general use of the communal Departmental Facilities such as centrifuges, autoclaves and Super-Q water system.

### 6.6.1 CENTRIFUGES

Before operating any low- or high-speed centrifuges and ultracentrifuges, users are required to read through the “**Standard Operating Procedures**” Manual.

Rotor failures at centrifuge speed creates an explosion hazard. Failures are caused by three common faults: (1) an unbalanced rotor bouncing off the spindle, (2) a rotor being operated at higher than design speeds, or (3) a rotor breaking apart as a result of age embrittlement and corrosion. Always carefully balance centrifuge tubes and bottles. Leaks will also create an imbalance, so check that the tubes are not cracked and that all the seals are in place. Be especially careful with swinging-bucket rotors. The buckets must swing freely, the tubes must be balanced, and all the buckets must be in place for the rotor to balance as designed. Note that each bucket is numbered and goes on a special position, they are not freely interchangeable. Be sure you use the correct buckets with the correct rotor. Get assistance from experienced users and read the instruction manuals carefully. Contact a faculty member immediately if there is a problem with any centrifuges.

#### **Important notes:**

- (i) All rotors should be stored upside down with absolutely no moisture in or on them.
- (ii) All spills in rotors and centrifuges should be cleaned up immediately with mild Beckman detergent and water only. This includes radioactive spills; under no circumstances use “Decon”, it destroys rotors. Do **not** soak rotors or parts.

### 6.6.2 AUTOCLAVES

Autoclaves are very dangerous devices which can malfunction and should only be used by people familiar with their operation. If you do not know what is wrong, contact Karen Ingram (SCIE 1203, ×53816), rather than punching buttons at random. Superheated liquids can “bump” when they are removed from the autoclave. This can spray you with boiling liquid if proper containers are not used. Stand back when opening the autoclave door to avoid being scalded by escaping steam. Lids must let air flow back into a solution as it cools; otherwise there is an implosion risk as a vacuum is created above a cooling solution. For a detailed description how to operate the autoclave properly, read the Departmental “Standard Operating Procedures” Manual.

Follow the rules listed below when using the autoclave:

- (i) Do not autoclave any vessel where the volume of liquid is more than half that of the container; remember liquids expand when heated.
- (ii) All liquids are to be autoclaved in deep trays ONLY to contain possible spills. If anything is spilled, scrub it with a dry scouring pad, wipe the material from the interior of the autoclave, and clean the drain.
- (iii) Remove material from the autoclave promptly to prevent smells from pervading the area. Not everyone likes the smell of sweaty socks!
- (iv) When including a biological standard in your run on request, place it directly in the CENTRE of the load. Otherwise, we will be unable to ascertain if the load has been properly sterilized.

### 6.6.3 Super-Q WATER SYSTEM

Pay attention to the following rules when using the Super-Q water system:

- (i) Users are required to sign in a log book.
- (ii) Turn off the water system after use to prevent flooding on the floor. Follow the posted instructions exactly.
- (iii) Place the black plastic tubing back in the sink after you have finished using the system, otherwise the end can become contaminated. This will affect some research groups' experiments.
- (iv) Please clean up any water that you spill on the stainless stain sink-top. Housekeeping does not do this, so the users have to keep the area clean.

## 6.7 GENERAL SECURITY

Our equipment is expensive, and the possibility of vandalism or theft is always present. The following security rules are applicable to this department.

- 6.7.1 All doors in the Communal Equipment Rooms, both SCIE 2202 and SCIE 2207, are kept locked at all times. If the lights are off when you enter an instrument room, that is an indicator that you are responsible for making sure it is in the same state when you leave.
- 6.7.2 Under no circumstances are wedges to be left under doors to expedite entry when you are working in the room or working after hours.
- 6.7.3 Do **not** let unauthorized people into the equipment room **at any time** by holding doors open for them when you leave. According to the department security regulations, a person must own a key to get entry into a specific equipment room.

## 7. USE OF THE INTERMEDIATE LEVEL RADIOACTIVE PREPARATION ROOM (MacN 229)

The Radioactive Preparation Room (MacN 229) is available for work with larger quantities of radioactive materials such as iodination, aliquot radioactive samples, *etc.*

- 7.1 Log in as soon as you enter the room. This is essential for tracking room usage and to protect workers in case of accidents or spills.
- 7.2 Absorbent bench paper must be placed on any surface where you intend to use isotopes. This includes dispensing from stock bottles on the counter, as well as working in the fumehood. Bench paper and scissors are available in the room. This is common sense; would you rather spend hours decontaminating the counter-top in the event of spill, or put contaminated bench paper in a waste bag?
- 7.3 Consider safety aspects when working in the room outside normal working hours. Remember that EHS personnel will not be available to help you in the event of an accident! Always consider the worst case scenario. If you are using high levels of isotope, what will you do if you drop the entire 10 mCi stock bottle on the floor? What if the fumehood ceases to function in the middle of an experiment with a volatile isotope? In case you think accidents can't happen to you, experience has shown that they are more likely to happen if you are tired, or working late at night.
- 7.4 This room is not a dumping ground for waste! Do not bring waste from the basic level lab here. Other people working in the room do not appreciate being irradiated unnecessarily. You must label all radioactive waste *clearly* and *precisely*. There are two types of radioactive trash bins for **solid** waste: a “<sup>32</sup>P” or “\*NO\* <sup>32</sup>P”, sort your radioactive waste accordingly. When a radioactive **liquid** waste bottle is full, tag it and get it picked up by EHS Department within a week (call ×53282). The regular pick-up time is on Friday mornings. Note that this room is not designed for long-term storage of radioactive waste. Refer to UG-ORSOG-101 (revised) for management of radioactive waste.
- 7.5 This room is wipe-tested on a weekly basis by Angela Wilson (SCIE 4104, ×53365).

## 8. ROUTINE CLEANING AND DISINFECTION OF LAB SPACE AND EQUIPMENT

- 8.1 Water baths must be cleaned regularly with 70% ethanol.
- 8.2 Centrifuges: Use tightly capped centrifuge tubes. Avoid spills by filling tubes only to three-quarters level. Do not overfill tubes. Check for spills in buckets and chamber. Clean up the buckets and inside of chamber with mild detergent immediately after spills.
- 8.3 Microscope: Use cover slips at all times.

- 8.4 Vortexer: Ensure that all centrifuge tubes are securely capped before vortexing.
- 8.5 Pipettor Aid: If cell cultures are aspirated into or past the cotton plug of the pipette, discard the filter component, and decontaminate the pipettor head **immediately**.
- 8.6 Decontamination of Biohazardous Materials, Cell Cultures and Disposables
- (i) Used glass pipettes: Soak in a pipette holder filled with soapy water.
  - (ii) Cell cultures: Discard in a container with 10% bleach.
  - (iii) Disposable plastic cell culture flasks: Discard into biohazard autoclave bag.
- When a biohazard bag is two-thirds full, autoclave for 60 minutes in “Gravity” cycle. Then bring the autoclaved waste materials to the garbage container (black bag) in the lab for disposal. Alternatively, consult Leanne Krick (SCIE 4104, ×53365) and bring the waste to Departmental Prep Room for disposal.

## 9. GENERAL PROCEDURES FOR CLEANING SPILLS OF BIOHAZARDOUS MATERIALS

### 9.1 Small spills

- (i) Wear double pairs of gloves and a lab coat before cleaning.
- (ii) Immediately soak up **all** of the spills by covering the spill with paper towels. **Do not lift** the towels.
- (iii) Then use 10% bleach to soak the paper towels, and leave for 5 min to allow disinfection of the cell cultures.
- (iv) Dispose of paper towels into autoclavable biohazard bag, then discard the top pair of gloves into the bag for later autoclaving.
- (v) Put on a new second pair of gloves, then clean up the area of spills again with 10% bleach. Finally, wipe the work area with 70% ethanol.

### 9.2 Large spills

- (i) Wear double pairs of gloves and a lab coat before cleaning.
- (ii) Soak up most of the spill by covering with paper towels to reduce the amount of cell culture. Then use 10% bleach to soak the paper towels. Discard paper towels into autoclavable biohazard bag for later autoclaving.
- (iii) Soak up the rest of the spill with paper towels in 10% bleach. Allow to disinfect for 5 min.
- (iv) Dispose of paper towels into autoclavable biohazard bag, then discard the top pair of gloves into the bag for later autoclaving.
- (v) Put on a new second pair of gloves, then clean up the area of spills again with 10% bleach. Finally, wipe the work area with 70% ethanol.

*If the spills are too large to control, evacuate the lab and call Emergency ×52000 immediately.  
Leave the clean-up to experts.*

### 9.3 Spills in the Biocontainment Cabinet

Biocontainment cabinets are designed to keep contamination out of cultures and hazardous

biological materials out of the environment. They do this by filtering and recirculating the air within the cabinet. Do not confuse biocontainment cabinets with regular lab chemical fumehoods or clean benches. Biocontainment cabinets vent a variable portion of the air in the hood, out into the lab through particulate filters. Chemical fumehoods suck air from the lab and vent it outside. Clean benches protect product only, not the worker. A biocontainment cabinet will **not** protect you from volatile chemical hazards. Conversely, a chemical fumehood will blow potentially hazardous biological materials out into the atmosphere.

Biocontainment cabinets are subjected to routine inspection, annually, to verify they are functioning properly. Do not interfere with the laminar airflow by placing objects on the mesh grill than runs across the front of the hood. Keep movement near the cabinet to a minimum. Work in the centre of the bench, not near the front. As with all equipment, it is important to learn the proper use of the laminar flowhood to achieve and maintain sterile conditions for tissue culture. Instructional videotapes are available from EHS Department (contact Lisa Kohlmeier at ×53282).

The general clean-up procedures of any spills in the Biocontainment cabinet are as follows:

- (i) Stop working **immediately**.
- (ii) Soak up the spill by adding more paper towels on the area of spill. Then soak paper towels in 10% bleach. Allow to decontaminate for 5 min. Then discard all paper towels into biohazard bag.
- (iii) Clean the work area three times with 10% bleach, followed by 70% ethanol. Discard paper towels into biohazard bag each time.

#### 9.4 **Spills in Lab Equipment**

##### 9.4.1 **Bench-top Centrifuge**

- (i) Soak up the spill with paper towels containing 10% bleach. Allow to decontaminate for 5 min. Then discard paper towels into biohazard bag.
- (ii) Remove buckets and soak in 20% bleach for 10 min. Then rinse with 70% ethanol.
- (iii) Clean the inside chamber with 10% bleach and rinse with 70% ethanol.

##### 9.4.2 **Water Bath**

- (i) Soak in 30% bleach for 10 min. Then discard all the water from bath.
- (ii) Repeat decontamination with 10% bleach and rinse with 70% ethanol.

#### 9.5 **Spills on your lab coat**

- (i) Immediately remove lab coat. Thoroughly soak the area of spill on the lab coat with 30% bleach, and allow to decontaminate for 10 min.
- (ii) Take to the Store Room for laundry (see Karen Ingram, SCIE 1203, ×53816).

## **Acknowledgment of Laboratory Regulations**

Dr. Sharom Laboratory  
Department of Molecular and Cellular Biology  
University of Guelph

I have read and understood the Laboratory Regulations Document and the Department of Molecular and Cellular Biology Safety Handbook. I have kept a copy of this Laboratory Regulations Document for future reference. I agree to cooperate in ensuring the diligent implementation of these regulations. Any questions which I have at the present time concerning laboratory safety have been answered to my satisfaction.

Name (printed): \_\_\_\_\_

Signature: \_\_\_\_\_

Name of Witness (printed): \_\_\_\_\_

Witness Signature: \_\_\_\_\_

Date signed: 200\_\_ \_\_ \_\_  
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(Copies of this acknowledgment will be kept on file in the Department Office.)