Toxins of Biologic Origin

**Good News:**
Toxins do not:
1. Replicate
2. Are not infectious
3. Difficult to transmit from person to person
4. Most have very low volatilities
5. Since most are proteins, unstable in environment (depends upon the toxin though)

**Bad News:**
Toxins can:
1. Cause death or severe incapacitation at relatively low exposure levels

**Main Laboratory Risks**
1. Accidental exposure by direct contamination of the mouth, eyes, other mucous membranes
2. Inadvertent aerosol generation
3. Needle-sticks or other accidents that compromise the skin barrier
4. Cutaneous absorption

**Risk Assessment**
As with any biohazardous agent, a risk assessment should be done to identify risks inherent to whatever procedures will be done. Examples of risk include autoinoculation during animal procedures, inhalation of aerosols (inadvertently or deliberately created) and static build-up when working with powders.

Questions to ask:
What is the probability of generating an aerosol?
What is the amount of toxin to be used?
How available are successful treatments?
How available are vaccines or antitoxins?
How experienced are personnel in working with toxins?
What is the intoxication/lethality dose?
What are the effects of exposure – both acute and chronic?
Is there a record of accidents indicating an increased probability of exposure?
What are the engineering controls (negative air pressure, biological safety cabinet etc.)?
What safety equipment is available and how effective is it?
What PPE is available and how effective is it?
What is the worst case scenario and how would you react?

It is imperative that the risk assessment is communicated to laboratory personnel so that everyone will be aware of each hazard and associated risk as well as give input on policies and procedures to either eliminate or reduce the hazard to an acceptable level.

**Biological Safety Cabinets vs. Chemical Fume Hoods**

BSCs are preferable to fume hoods for a number of reasons. Since BSCs are HEPA-filtered, the potential contamination of downstream ductwork, exhaust ventilation equipment and the environment is avoided. Not only do BSCs protect the environment and worker, but due to the HEPA-filtered supply air, the
integrity of the toxin is also protected. The recommended BSC types for toxin work include the Class II, Type B2 and the Class III.

The chemical fume hood is designed to safely ventilate volatile chemical fumes and vapors. Biological toxins, however, are particulates (protein) and do not produce fumes/vapors. Fume hoods can be used for certain kinds of toxin work if the following criteria are met:

- The average face velocity is 100 linear feet per minutes (lfpm) +/- 10%. Higher face velocities may increase air turbulence, thereby reducing the capture efficiency of the fume hood.

- Fume hood should only be used for toxins in solution. Dry powdered toxins should not be used since they can aerosolize easily due to air turbulence caused by the high volume of flow. If there is no other option to a fume hood, then a NIOSH approved full face respirator with HEPA canister filter should be worn during the operation.

- Ideally, the fume hood should be connected to a HEPA filtration system before release into the environment.

- Follow the 15 cm (6 inch) line rule. The 15 cm line is located 15 cm from the face of the fume hood. Mark with tape or paint. The area in front of the 15 cm is a zone of air turbulence, and toxin particles can easily be ejected from the fume hood into the lab. So all work with the toxin should occur behind the line towards the rear of the hood.

Both horizontal and vertical laminar flow “clean benches” SHOULD NOT be used since air flow is from the cabinet out towards the worker. Go to the Biological Safety Cabinet section for more information.

**Recommendations for Personnel Working with Toxins**

Training for toxin workers should include:
- Discussion of the physical and biological characteristics of the toxin(s)
- Instruction in the use of the appropriate PPE
- Instruction in the use, care and maintenance of equipment related to toxin procedures
- Discussion of the decontamination/cleaning procedures
- Discussion of the signs/symptoms of exposure
- Review of the written emergency response plan as well as response exercises
- Training in first aid
- Discussion of the various types of prophylaxis/immunization
- Review of the written waste disposal plan

**Good Work Practices**

Develop standard operating procedures that are clear, in chronological order, discuss PPE and decontamination procedures.

No eating, drinking, applying of cosmetics, storage of foodstuffs, contact lens manipulation in the work area. Strict adherence to these procedures has been shown to prevent accidental ingestion of lab materials.
Entrances into the work area should be posted with a sign such as “CAUTION, TOXIN USE AREA”. Only personnel performing the operations are permitted in the lab during the toxin work.

Whenever possible, design protocols requiring less than the human lethal dose.

Use a biological safety cabinet if possible but a chemical fume hood can be used under certain circumstances.

When removing a syringe from a stoppered bottle, place a cotton ball or gauze pad wetted with decon solution at the junction of the needle and stopper to prevent any pressurized, aerosolized toxin material from escaping.

Whenever possible, keep toxin containers secured in a rack or foam block to prevent accidental spills during the operation.

Special care must be taken with powders or lyophilized toxins since static and ventilation air flow can easily cause the toxin to form a particulate cloud. Since powdered toxins are generally highly concentrated, it is likely a lethal dose could be delivered. It is best to work with the powder form in an enclosed glove box.

Have a clear and organized work area when injecting animals or manipulating sharps. Practice the procedure in dry runs until the technique is perfected.

Toxins should be stored in double containment configuration. The outer container should be leak proof and unbreakable. Toxin storage areas should maintain an inventory of toxins and quantities.

Minimize the use of sharps. If there is no alternative to the use of sharps, follow the BMBL guidelines under standard microbiological practices.

Use caution with disposable pipette tips. Tips can easily be ejected from a BSC or fume hood during operations. Tips should be injected into a container of decon solution within the hood.

In some cases work with toxins should follow the “two person” rule which requires that two authorized personnel be present during operations. This is especially important when the protocol requires an amount greater than the human lethal dose or for any operation where toxin aerosolization is probable or intentional.

Air circulating fans, window air conditioners or open windows can produce cross currents which may interfere with the capture efficiency of BSCs or fume hoods. Fans or window air conditioners should be turned off during toxin operations and windows kept closed.

Personnel should always wear a lab coat during toxin operations. Keep them in the lab until processed for laundering.

Personnel should always wear goggles and face shield during work.

Use the 15 cm line concept for all work with toxins in fume hoods or BSCs.

Do not work with dry, powdered toxins in a fume hood.
Perform a pre-operational checklist before work. Contact BSO for the checklist.

**Decontamination Recommendations**

The use of appropriate decontamination methods depends upon the toxin in use and what needs to be deconned (personnel, equipment, work areas). Appendix I of the BMBL has several tables on toxins and effective decon methods.

**Personnel Decontamination:** In most cases, the first step should be removal of clothing followed by a soap and water wash of the contaminated area (not eyes). Eye splashes should be immediately treated with copious of water from an eyewash station.

**Equipment/Work Surfaces:** For most toxins, 0.5% sodium hypochlorite (household bleach) is an effective decontaminant but verify through a literature search for your particular toxin.

**Glassware:** Soak all toxin contaminated glassware in a mixture of 2.5% sodium hypochlorite and 0.25N sodium hydroxide for 16 hours or more. Alternatively, soak in a 5% solution of bleach for 8 hours.

**Liquids:** Decontaminate toxin solutions 1:1 with a mixture of 5% bleach and 0.5N NaOH, mix well and hold for 8 hours. Contact the Office for Regulatory Compliance for pick-up.

**Emergency Response Recommendations**

The risk assessment should address emergency procedures which are specific to the operations, the type of toxin used and the configuration of the facility. Things to consider include:

- Access to readily available medical support.
- Posted phone numbers of emergency response personnel including the Longest Student Health Center.
- Other topics such as first aid, skin contamination, eye and mucous membrane contamination, spills, fume hood or BSC failure, theft, and building power failure.

**Security Considerations for Toxin Laboratories**

This will vary based on the type, amount, location and perceived potential for terrorist threat. A security program should be documented and consider some of the following:

- Toxin storage areas (such as freezers) should be locked at all times. The facility should institute a key control plan.
- Facilities with a high public relations risk factor, a concern of industrial sabotage or other types of threats can employ a motion detector system with alarms for off-duty surveillance.
- A facility security force, punch locks, card/palm print readers or similar devices may be used where a higher level of security is desired.