

## **Biosafety in the Museum of Vertebrate Zoology Standard Operating Procedures**

### **A. BIOSAFETY LEVEL 2**

The Museum of Vertebrate Zoology (MVZ) is required to be a Biosafety Level 2 (BSL2) facility to comply with U.S. Department of Agriculture (USDA) import and transport permits for restricted products. The Museum's USDA permits covers all avian material of foreign. Anyone working in a lab designated as BSL2 within the MVZ must read this Standard Operating Procedures (SOP) and sign the sheet in the relevant lab(s) before handling USDA-restricted products. The MVZ may require BSL2 procedures for additional materials that are not specifically regulated by the USDA (e.g., potentially infectious agents of non-foreign origin).

For complete BSL2 regulations, see <http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4s2.htm>

Additional information:

UC Berkeley fact sheet on biohazard waste:

<http://www.ehs.berkeley.edu/sites/default/files/lines-of-services/biosafety/01biohazwaste.pdf>

UC Berkeley fact sheet on handling and disposal of sharps:

<http://www.ehs.berkeley.edu/sites/default/files/lines-of-services/biosafety/12sharps.pdf>

### **B. BSL2 LABS IN THE MVZ**

The following labs in the MVZ are designated as BSL2 because they are used to store and/or process avian materials that might be regulated by the Museum's USDA permits:

Room 1143 – Tissue Collection Storage Facility/Tissue Collection Processing Lab

Room 1168 – Specimen Preparation Lab

Room 4123 – Tissue Collection Storage Facility

Room 4123A – RNA RNA Extraction Lab

Room 4140 – Historical DNA Extraction Lab

Room 4141 – Modern DNA Extraction and PCR Setup Lab

### **C. RISK LEVELS**

#### **1. High risk animals:**

Risk will vary depending on the animal and the area of origin. As a general rule, the MVZ considers the following groups to be higher risk, and thus precautions should be taken when working with this material. Only bird specimens from foreign countries are considered high risk from the standpoint of the USDA BSL2 procedures.

- All carnivores
- Birds (especially corvids)
- Rodents (especially *Peromyscus* and *Neotoma* species)
- Bats

- Primates
- Specimens from foreign countries
- Fresh specimens that have never been frozen

## **2. Becoming familiar with Zoonoses:**

A zoonosis is an infectious disease transmittable between animals and humans. Zoonoses with a wildlife reservoir are typically caused by various bacteria, viruses, parasites, and perhaps fungi. In the MVZ Prep Lab (Rm. 1168), we mostly work with animals that have been frozen in the -26°C freezer for several months or years, and therefore do not need to worry as much about bacteria, parasites, or fungi. We do have to worry about viruses, which have been shown to survive freezing.

In 2001, of the catalogued 1,415 known human pathogens, 62% were of zoonotic origin. There is a reference book on Zoonoses available in the Prep Lab. You may also want to refer to the CDC website on zoonoses: <http://www.cdc.gov/healthypets/diseases/index.html> or go to <http://www.cdc.gov> and search for the disease in which you are interested.

Here are some zoonoses to become familiar with (ones in italics are of particular concern in the MVZ Prep Lab):

1. *Rabies*
2. *West Nile Virus*
3. *Hanta Virus*
4. *Tularemia*
5. *Arenaviruses*
6. *Salmonella*
7. Histoplasmosis
8. Plague
9. Herpes B
10. Leptospirosis
11. Cryptococcosis
12. Histoplasmosis
13. Monkeypox
14. Q Fever

## **D. GENERAL PROCEDURES FOR BSL2 LABS**

### **1. Dealing with animal materials:**

- Don't assume anything is safe. Always wear gloves and other protection as needed (see below). DO NOT assume animal materials are safe because they have been frozen (research has shown that certain zoonoses can survive freezing).
- Dispose of sharps (hypodermic needle tips, razor blades, etc.) in a sharps disposable container. Never reuse sharps. ALL biohazard contaminated sharps must go into a designated biohazard sharps container.

- Clean all utensils and spaces after use. Use bleach to clean counters that have been in contact with potentially biohazardous materials.
- DO NOT touch general use equipment with gloves on (i.e., faucets, knobs, telephones, light switches, etc.).
- Always wash your hands thoroughly and with soap before leaving the room.

We encourage anyone, but in particular people who are immunocompromised and women of child-bearing age, to discuss concerns about their potential susceptibility to infection with the manager of the lab in which they are working.

## **2. Cuts or spray/aerosol exposure:**

Treat any exposure seriously.

1. Wash the exposed area thoroughly (at least 10 minutes).
2. Alert your supervisor.
3. Apply first aid if needed.
4. Go to the Tang Center (or after 6 p.m., to Alta Bates) and have the exposure checked.
5. Your supervisor will follow through by contacting EH&S and making an accident report.

## **3. Biohazard disposal:**

All materials related to the processing and cleanup of specimens or tissues should be considered potential biohazards (this includes any animal materials, and gloves and paper towels used). Biohazards must be placed in a designated biohazard container. They MUST NOT be put in the regular garbage or down the sink.

## **4. Personnel Animal Contact Policy:**

Personnel should refrain from direct contact with "like species" outside the lab for several days when working on foreign animal materials that are potentially infectious. For example, if a person is manipulating foreign non-processed bird carcasses, he/she should avoid contact with a pet bird at home. The primary method of preventing transmission of potentially infectious agents to "like species" is through adherence to proper basic lab procedures (e.g., wearing gloves, hand washing, limiting aerosol production, proper biohazard disposal, etc.) as stated in this SOP.

## **5. Evaluation of Plan**

The MVZ Biosafety SOP will be evaluated at least annually to ensure that it is up-to-date with current procedures. In addition, MVZ personnel, under direction of the Safety Officer, will conduct drills or exercises annually to test and evaluate the effectiveness of the plan. The plan will be reviewed and revised, as necessary, after each exercise and after any incident.

## **E. Room 1143: TISSUE COLLECTION LIQUID NITROGEN (LN2) STORAGE FACILITY AND PROCESSING LAB**

### **1. Access:**

Access to the facility is at the discretion of the MVZ Faculty and Staff Curators (see <http://mvz.berkeley.edu/Directory.php>), and failure to follow Standard Operating Procedures may result in restriction of access.

Outside doors that provide access to the facility must be kept closed except when trained personnel are in the facility. Anyone working in the facility must be trained by a Staff Curator prior to starting their work.

### **2. Protection:**

All workers should wear lab gloves (either latex or nitrile) when handling tissue samples. Lab coats and face protection are required when working with USDA-restricted materials (e.g., avian samples of foreign origin). Lab coats and protective face shields are also recommended when working with other types of potentially "high risk" samples (see "High Risk Animals" above). Lab coats, gloves, and disposable face shields are available in the lab.

USDA-restricted materials must be handled only in the bench spaces delimited by red tape and marked with biohazard stickers. Avian tissues of foreign origin MAY NOT be worked on in any other part of the labs. When work is completed, these areas must be thoroughly bleached and cleaned by the user.

All workers must be trained by a Staff Curator on the additional hazards and safety requirements when working with or around LN2 prior to starting their work (see Rm. 1143 Standard Operating Procedures binder located in Rm. 1143).

### **3. Biohazard Disposal:**

Biohazards must be thrown in a biohazard box in the lab. They MUST NOT be put in the regular garbage or down the sink. Biohazard waste is taken to Room 1168 (Specimen Preparation Lab) weekly for disposal by the UC Berkeley Department of Environmental Health and Safety.

## **F. ROOM 1168: SPECIMEN PREPARATION LAB**

### **1. Access:**

Access to the lab is at the discretion of the manager of this facility (Senior Museum Scientist Terri Barclay, [tlynnwong@berkeley.edu](mailto:tlynnwong@berkeley.edu)), and failure to follow Standard Operating Procedures may result in restriction of access.

Outside doors that provide access to the lab must be kept closed except when trained personnel are in the lab. Anyone working in the lab must be trained by Terri Barclay prior to starting their work. Users should read the General Lab Protocol and agree to all terms and conditions within it. Also, users should view the safety bulletin board for any announcements or notices concerning lab safety and zoonoses. (Copies of all printed materials are available upon request).

## **2. Protection:**

Standard Outfit: All workers should wear lab gloves (either latex or nitrile). Lab coats and face protection are recommended (the disposable face shields are the most comfortable, but we also have goggles and face masks for filtering air). Lab coats, gloves, and face protection are available in the lab.

Special Cases:

- When working with sharp knives/large cutting edges, it's recommended to wear metal mesh gloves under your latex/nitrile gloves.
- When working with animals that potentially carry zoonoses, workers must work in the Biological Safety Cabinet, wear double gloves, and a lab coat.

## **3. Biohazard disposal:**

Biohazards must be thrown in a biohazard bin in the lab. They **MUST NOT** be put in the regular garbage or down the sink. Take special care when washing/flushing specimens with water -- catch all materials in a strainer and dispose of that in the biohazard bin. There is a separate biohazard sharps container for razors and scalpel blades.

Biohazard waste bins are placed in the lab's walk-in cold room (4°C) at the end of each day, and then moved to the lab's walk-in freezer (-25°C) at the end of each week. The manager of this facility ensures that the bin contents are disposed of by the UC Berkeley Department of Environmental Health and Safety in accordance with UC Berkeley and state regulations.

## **4. Biological safety cabinet:**

The MVZ Prep Lab has a Class 2 biosafety cabinet, 4 foot with no integration of air ducts. All work with USDA-restricted or high risk animals should be done in this cabinet.

## **G. ROOM 4123: TISSUE COLLECTION ULTRALOW FREEZER STORAGE FACILITY**

This facility should be used **ONLY** for taking closed tissue vials out of the collection, **NOT** for handling of tissue samples.

### **1. Access:**

Access to the facility is at the discretion of the MVZ Faculty and Staff Curators (see <http://mvz.berkeley.edu/Directory.php>), and failure to follow Standard Operating Procedures may result in restriction of access.

Outside doors that provide access to the facility must be kept closed except when trained personnel are in the facility or adjoining labs. Anyone working in the facility must be trained by a Staff Curator prior to starting their work.

## **H. ROOM 4123A: RNA EXTRACTION LAB; ROOM 4140: HISTORICAL DNA EXTRACTION LAB; ROOM 4141: MODERN DNA EXTRACTION AND PCR SETUP LAB**

### **1. Access:**

Access to these facilities is at the discretion of the manager of the Evolutionary Genetics Lab and the MVZ Faculty and Staff Curators (see <http://mvz.berkeley.edu/Directory.php>). Failure to follow Standard Operating Procedures may result in restriction of access.

Doors that provide access to these rooms are kept locked at all times. Lab Manager Lydia Smith must give anyone working in the facilities safety training prior to starting their work. Only those who have completed this training are given the access codes for the outside doors.

All researchers who use this facility are trained in the SOP for handling tissue from birds collected outside the United States, regardless if they expect to need them. We keep a record of their signature in room 4141 indicating training completion and an understanding of the regulations in this SOP.

### **2. Protection:**

All workers should wear lab gloves (either latex or nitrile) when handling tissues for DNA extraction. Lab coats and face protection are required when working with USDA-restricted materials (e.g., avian samples of foreign origin). Lab coats and face protection are also recommended when working with other types of potentially “high risk” samples (see “High Risk Animals” above). Lab coats, gloves, and other disposable face shields are available in the lab.

USDA-restricted materials must be handled only in the bench spaces delimited by red tape and marked with biohazard stickers. Avian tissues of foreign origin MAY NOT be worked on in any other part of the labs. When extractions are completed, these areas must be thoroughly bleached and cleaned by the user.

### **3. Biohazard disposal:**

Biohazards must be discarded into a biohazard box in the lab. They MUST NOT be put in the regular garbage or down the sink. These biohazard boxes are to remain closed at all times when they are not actively in use. There is a separate biohazard sharps container for razors and scalpel blades.

Biohazard items are all items that come into contact with USDA-restricted samples during the handling of tissues for cell lysis or during the precipitation and purification of these cell lysis solutions for DNA extraction. This includes but is not limited to:

- Pipette tips
- Microcentrifuge tubes
- Parafilm
- Kimwipes/paper towels
- Spin columns
- Scalpel and razor blades (in biohazard sharps waste only)
- Gloves

When full, biohazard containers should be placed in a bag inside the labeled bin in room 4141. The lab manager ensures that the bin contents are disposed of in accordance with UC Berkeley and state regulations. All biohazard containers are frozen within one week of being used and are taken to room 1168 for disposal and eventual incineration through the UC Berkeley Environmental Health & Safety Department.

#### **4. Subsequent laboratory research with these specimens:**

Once the USDA extraction procedure is completed, the tissue will have been subjected to one or more of the following treatments:

- Immersion in 70% ethanol, and/or
- Immersion in phenol/chloroform, and/or
- Heating to a minimum of 56°C for at least 3 hours, and/or
- Immersion in a minimum of 2% sodium dodecyl sulfate (SDS) for at least 30 minutes, and/or
- Treatment with proteinase K, and/or
- Treatment with guanidine HCL

At this point, the material is no longer considered a biohazard and it is safe to remove the DNA extracts from Biosafety Level 2 areas to be used in different areas of the lab.