DEPARTMENT OF HEALTH AND HUMAN SERVICES

Public Health Service

Centers for Disease Control
and Prevention
Atlanta, GA 30333

Axel Lehrer
651 Ilalo Street
Honolulu, HI 96813

Application # 20191106-4778A & 20191017-4587A

Inspection of: University of Hawaii

Inspection dates: 06 January 2020

This letter is to confirm that inspectors from the Centers for Disease Control and Prevention's (CDC) Import Permit Program are scheduled to inspect your entity beginning at 9:00 AM on the date and at the location listed above. Your entity is conducting activities regulated by 42 CFR Part 71.54 (Import Regulations for Infectious Biological Agents, Infectious Material and Vectors) and you are therefore required to allow inspectors access to all laboratories, biosafety records and personnel related to the use and storage of imported materials listed on your Import Permit Application.

The purpose of this inspection is to verify that the biosafety measures that are in place are commensurate with the hazard(s) posed by the infectious biological agent, infectious substance, and/or vector to be imported, and the level of risk given its intended use.

To facilitate the inspection process the following items should be available for review: risk assessments associated with the imported biological agent(s), biosafety plan(s) and biosafety training records. Implementation of biosafety plans and procedures will be verified by conducting interviews with personnel regarding safety practices. A brief presentation or overview of the type of work conducted by the permittee or designated representative(s) is beneficial.

If you have not already done so, please fax or email directions to the facility, parking instructions, any facility entrance requirements (e.g., immunizations, identification), and any special personal protective equipment requirements necessary to conduct laboratory inspections at your facility.

All CDC inspectors are credentialed representatives of the Director of CDC. The inspectors who will perform the inspection at your entity are identified below:

[redacted] Lead Inspector
[redacted]
[redacted]
[redacted]

Failure to allow these inspectors to conduct an inspection at your entity may result in civil penalties and/or prevent the approval of your import application.

For questions regarding this inspection please contact [redacted] Team Leader/Import Permit Program) at 404.718.2053.

Thank you,

Centers for Disease Control and Prevention
Import Permit Program
Ph: 404.718.2077; Fax: 404.718.8333
importpermit@cdc.gov
www.cdc.gov/od/eaipp

DEPARTMENT OF HEALTH AND HUMAN SERVICES

Public Health Service
Centers for Disease Control
and Prevention (CDC)
Atlanta, GA 30329-4027

January 29, 2020

Axel Lehrer
651 Ilalo Street
Honolulu, HI 96813

RE: Entity Inspection Report
University of Hawaii (Applicant: Axel Lehrer)

Dear Dr. Lehrer:

An inspection of your facility was scheduled in response to your request to import and conduct work with blood/blood products from humans that survived Ebola virus disease and tested negative for the virus but may contain other infectious biological agents (applications 20191017-4587A and 20191106-4778A). The purpose of the Centers for Disease Control and Prevention (CDC) Import Permit Program (IPP) visit was to assess whether the importer's facility has implemented biosafety measures commensurate with the hazard posed by the infectious biological agent, infectious substance, and/or vector to be imported, and the level of risk given its intended use.

Inspectors from the CDC IPP visited your facility located at 651 Ilalo Street Honolulu, HI on January 6, 2020. A list of laboratories inspected on these dates is on file with this letter at CDC.

The following personnel from the CDC IPP inspected the facility:

[redacted], Lead Inspector
[redacted]

A list of individuals from the University of Hawaii present was provided to you at the close of the inspection.

The regulations for the importation of infectious biological agents, infectious substances, and vectors (42 CFR 71.54) require the implementation of biosafety measures commensurate with the hazard posed by the infectious biological agent, infectious substance, and/or vector to be imported, and the level of risk given its intended use. See 42 CFR 71.54(b)(3). To determine whether your facility meets this regulatory requirement, CDC uses the nationally recognized biosafety guidelines contained in the Biosafety in Microbiological and Biomedical Laboratories (BMBL, http://www.cdc.gov/biosafety/publications/bmbl5/index.htm).

As a condition of being issued and retaining CDC import permits, the importer (Axel Lehrer) at the University of Hawaii must address each of the items described in Attachment 1, including the specific actions or changes to be adopted. A detailed response should be received by this office no later than 30 business days from receipt of this report. Send an electronic copy of your response to Thomas Cremer at importpermit@cdc.gov.

Sincerely,

[redacted]

[redacted]

Division of Select Agents and Toxins
Office of Public Health Preparedness and Response
Centers for Disease Control and Prevention

Attachment 1:
Inspection Observations and Corrective Actions Required

Attachment 2:
Concerns

Attachment 1: Inspection Observations and Corrective Actions Required

A detailed response to each of the following items is due to **DSAT within 30 business days of the receipt of this report.** The items below are ordered by their relative severity - highest to lowest.

1. **Requirement:** To apply for a permit, an individual must submit a signed, completed CDC Form 0.753 (Application for Permit to Import Biological Agents or Vectors of Human Disease into the United States) to the HHS/CDC Import Permit Program. [42 CFR 71.54(g)(1)]

   Observation: The CDC Import Permit Applications for Axel Lehrer was inaccurate: Eye protection was required in the laboratories, but this was not reported in the application (Section F).

   Corrective Actions Required as a Condition of Issuing/Retaining an Import Permit: A response was provided after the inspection, but prior to issuance of this report, no further action is needed.

2. **Requirement:** The importer has implemented biosafety measures commensurate with the hazard posed by the infectious biological agent, infectious substance, and/or vector to be imported, and the level of risk given its intended use. [42 CFR 71.54(b)(3)]

   Observation: The following is an example of the University of Hawaii's failure to provide adequate biosafety and containment procedures consistent with nationally recognized safety standards or equivalent practices:

   a) Staff and visitors did not wash their hands before leaving BSL-2 laboratory. [BMBL: (BSL-2) A2]

   b) There was evidence of improper reuse of sharps in the BSL-2 laboratory; there were open packages of recapped needles discarded on laboratory shelves, not in the sharps containers. [BMBL: (BSL-2) A5]

   c) Two aerosol containment centrifuge buckets were missing o-rings. [BMBL: (BSL-2) C1-b]

   d) Signage was posted at the laboratories, but entry/exit procedures were inadequate. [BMBL: (BSL-3 and BSL-2) A9]

      1) Entry/exit procedures for the BSL-2 did not specifically state the required personal protective equipment (PPE).

      2) The BSL-3 was not active at the time of the inspection, so escorts and inspectors did not follow the posted entry procedures (e.g., sleeve covers and second pair of gloves). Varied entry procedures should be defined and posted.

   e) Chairs in the BSL-2 were damaged, so they could not easily be cleaned and decontaminated [BMBL: (BSL-2) D4-b]

   f) The biosafety manual for the BSL-2 laboratory was from 2017 and was not laboratory-specific. The plan lacked complete information on decontamination, incident reporting, medical surveillance, and use of PPE. [BMBL: (BSL-2) B4, B7, B8, C3]

   g) Visitor training was inadequate. [BMBL: (BSL-2 and BSL-3) B1]

      1) Visitor training did not include the hazards of the BSL-2 laboratory (e.g., Vesicular Stomatitis Virus, Plasmodium species).

      2) Visitor training for the BSL-3 included specific on several infectious agents, but did not include information specific to Ebola virus disease.

Corrective Actions Required as a Condition of Issuing/Retaining an Import Permit: Provide:

a) The measures implemented to ensure that staff and visitors wash their hands before leaving the BSL-2 laboratories.

b) The measures implemented to ensure that staff do not reuse or recap sharps such as needles.

c) Evidence that the centrifuge buckets have o-rings.

d) Copies of the updated signage for BSL-2 and BSL-3 laboratories and evidence (e.g., photograph) that the signage has been posted.

e) Evidence that the damaged chairs have been replaced.

f) An updated copy of the biosafety plan that is laboratory is specific and addresses the following: Routine decontamination, incident reporting, what types of medical surveillance/services are provided, and proper PPE usage (including eye protection).

g) An updated visitor training curriculum and the measures implemented to provide training before visitors enter the laboratory.

Attachment 2: Additional Concerns

DSAT has the following additional items of concern for your consideration. While these items do not currently represent a departure from the biosafety standards required by the import regulations, if left unaddressed, they may constitute such a departure in the future. No responses are required regarding the items below.

1. During the inspection, available University of Hawaii staff were not aware proposed work with convalescent blood products from survivors of Ebola virus disease at BSL-2 (see application 20191106-4778A). IBC minutes from 2019 stated that the Principal Investigator was only approved for such activities at BSL-3 (see application 20191017-4587A). The permittee should obtain all relevant institutional approvals for work at BSL-2 prior to initiation.

2. Inspectors passed through Room B of the BSL-3, though this space was not included on the import permit application. Signage at Room B indicated that N-95s are required for entry, but staff reported that PAPRs are required. Consider updating the signage to Room B of the BSL-3 to reflect the current practices.

3. During interviews staff reported that they were enrolled in a medical surveillance/occupational health program, but they stated it was due to unrelated work that they conduct in the vivarium. In discussions with organization staff it was acknowledged that staff that only work in the BSL-2 are not provided any medical surveillance/occupational health services. Consider implementing measures that ensure that all laboratory staff (BSL-2 and BSL-3) are provided medical surveillance/occupational health services.

4. N-95 respirators were reused, and the plan stated that reuse was allowed for up to 8 hours. However, it was not clear how hourly usage was being tracked. Consider re-evaluating procedures for reuse of N-95 respirators.

Attachment 1: Inspection Observations and Corrective Actions Required

A detailed response to each of the following items is due to **DSAT within 30 business days of the receipt of this report.** The items below are ordered by their relative severity - highest to lowest.

1. Requirement: To apply for a permit, an individual must submit a signed, completed CDC Form 0.753 (Application for Permit to Import Biological Agents or Vectors of Human Disease into the United States) to the HHS/CDC Import Permit Program. [42 CFR 71.54(g)(1)]

Observation: The CDC Import Permit Applications for Axel Lehrer was inaccurate: Eye protection was required in the laboratories, but this was not reported in the application (Section F).

Corrective Actions Required as a Condition of Issuing/Retaining an Import Permit: A response was provided after the inspection, but prior to issuance of this report, no further action is needed.

2. Requirement: The importer has implemented biosafety measures commensurate with the hazard posed by the infectious biological agent, infectious substance, and/or vector to be imported, and the level of risk given its intended use. [42 CFR 71.54(b)(3)]

Observation: The following is an example of the University of Hawaii's failure to provide adequate biosafety and containment procedures consistent with nationally recognized safety standards or equivalent practices:

a) Staff and visitors did not wash their hands before leaving BSL-2 laboratory. [BMBL: (BSL-2) A2]

Corrective Actions Required: The measures implemented to ensure that staff and visitors wash their hands before leaving the BSL-2 laboratories.

RESPONSE: The BioSafety Manual has been updated to include a section on visitor training (see below) and new signs have been put up to remind all staff and visitors to wash their hands when leaving the laboratory. See included picture of new signage on door of BSB 336.

b) There was evidence of improper reuse of sharps in the BSL-2 laboratory; there were open packages of recapped needles discarded on laboratory shelves, not in the sharps containers. [BMBL: (BSL-2) A5]

Corrective Actions Required: The measures implemented to ensure that staff do not reuse or recap sharps such as needles.

RESPONSE: A retraining session has been held for members of the Lehrer laboratory to reiterate the importance of NOT reusing and recapping needles. List of participants attached.

c) Two aerosol containment centrifuge buckets were missing o-rings. [BMBL: (BSL-2) C1-b]

Corrective Actions Required: Evidence that the centrifuge buckets have o-rings.

RESPONSE: New o-rings have been bought for the buckets. Buckets without o-rings have been removed and are currently not in service. Attached Purchase Order form for new o-rings.

d) Signage was posted at the laboratories, but entry/exit procedures were inadequate. [BMBL: (BSL-3 and BSL-2) A9]
   1. Entry/exit procedures for the BSL-2 did not specifically state the required personal protective equipment (PPE).
   2. The BSL-3 was not active at the time of the inspection, so escorts and inspectors did not follow the posted entry procedures (e.g., sleeve covers and second pair of gloves). Varied entry procedures should be defined and posted.

Corrective Actions Required: Copies of the updated signage for BSL-2 and BSL-3 laboratories and evidence (e.g., photograph) that the signage has been posted.

RESPONSE: New signage have been placed on BSL2 (336-3) and BSL3 (Suite AB and AC) doors to indicate PPE requirements for working in the lab. Photos of posted signs are attached. Please note that at the time of inspection, the laboratory was inactive. Therefore, PPE for active studies was not utilized.

e) Chairs in the BSL-2 were damaged, so they could not easily be cleaned and decontaminated [BMBL: (BSL-2) D4-b]

Corrective Actions Required: Evidence that the damaged chairs have been replaced.

RESPONSE: The seats and chair backs have been exchanged. Photos of replacement chairs attached.

f) The biosafety manual for the BSL-2 laboratory was from 2017 and was not laboratory- specific. The plan lacked complete information on decontamination,

incident reporting, medical surveillance, and use of PPE. [BMBL: (BSL-2) B4, B7, B8, C3]

Corrective Actions Required: An updated copy of the biosafety plan that is laboratory is specific and addresses the following: Routine decontamination, incident reporting, what types of medical surveillance/services are provided, and proper PPE usage (including eye protection).

RESPONSE: The BSL2 Biosafety Manual has been updated to include medical surveillance/services provided. Should a breach in laboratory procedures occur, the BSL2 researcher is being instructed to be vigilant and if necessary self-quarantine and monitor temperature and other relevant signs of disease specific symptoms. Should a laboratory acquired infection (LAI) occur, workman's Compensation procedures will then be implemented. Additionally, we have included specifics on incident reporting from the JABSOM EHSO manual. Because infectious agents are used in multiple labs, we have included a section similar to that found in the JBF Operations Manual on Agent Specific Information which include Laboratory Safety and Containment Recommendations, Decontamination Procedures, and PPE requirements.

g) Visitor training was inadequate. [BMBL: (BSL-2 and BSL-3) B1]

1. Visitor training did not include the hazards of the BSL-2 laboratory (e.g., Vesicular Stomatitis Virus, Plasmodium species).
2. Visitor training for the BSL-3 included specific on several infectious agents, but did not include information specific to Ebola virus disease.

Corrective Actions Required: An updated visitor training curriculum and the measures implemented to provide training before visitors enter the laboratory.

RESPONSE: We have written a Risk Assessment Hazard Communication for VSV. Plasmodium is no longer cultured in the laboratory. We have written a RAHC for Ebola virus and added it to the JBF RAHC. Both are attached. Please note that during the visit, samples from healthy Ebola virus disease (EVD) survivers were not in the BSL-3 facility. It was our intention that once the CDC approved our application, we were going to have signs and information in place for EVD.

Attachment 2: Additional Concerns

DSAT has the following additional items of concern for your consideration. While these items do not currently represent a departure from the biosafety standards required by the import regulations, if left unaddressed, they may constitute such a departure in the future. No responses are required regarding the items below.

1. During the inspection, available University of Hawaii staff were not aware proposed work with convalescent blood products from survivors of Ebola virus disease at BSL-2 (see application 20191106-4778A). IBC minutes from 2019 stated that the Principal Investigator was only approved for such activities at BSL-3 (see application 20191017-4587A). The permittee should obtain all relevant institutional approvals for work at BSL-2 prior to initiation.

   RESPONSE: Importation of the WHO Ebola Serum Standards (20191106-4778A) and importation of serum samples from Liberia (20191106-4587A) are part of two different IBC approved projects. WHO Ebola Serum Standards are approved for BSL-2 containment by the University of Hawaii IBC; a recombinant protein expression project. As such, this work will be conducted in BSL-2 facilities in room BSB 336.

2. Inspectors passed through Room B of the BSL-3, though this space was not included on the import permit application. Signage at Room B indicated that N-95s are required for entry, but staff reported that PAPRs are required. Consider updating the signage to Room B of the BSL-3 to reflect the current practices.

   RESPONSE: We have updated the signage for the BSL3 suites AB and AC to reflect, PPE requirements for active manipulations and non-active manipulations. See attached new signage and pictures of these signs on the doors.

3. During interviews staff reported that they were enrolled in a medical surveillance/occupational health program, but they stated it was due to unrelated work that they conduct in the vivarium. In discussions with organization staff it was acknowledged that staff that only work in the BSL-2 are not provided any medical surveillance/occupational health services. Consider implementing measures that ensure that all laboratory staff (BSL-2 and BSL-3) are provided medical surveillance/occupational health services.

   RESPONSE: For all JBF researchers (BSL-3), we are investigating the possibility of using the current Animal Veterinary Service (AVS) Occupational Health Program for JBF users. The University of Hawaii AVS has contracted through a local hospital (Straub Hospital) to review a self-survey, and determine

suitability to work with animals. While the survey is animal centered, it does have questions on use of infectious agents, which is relevant to work in the BSL3.

4. N-95 respirators were reused, and the plan stated that reuse was allowed for up to 8 hours. However, it was not clear how hourly usage was being tracked. Consider re-evaluating procedures for reuse of N-95 respirators.

   RESPONSE: We have change our procedures and have now instituted a single use policy for N-95 respirators. A discussion of the new policy was presented at a training session on 2/25/2020. The sign-in sheet is attached.

Attachments:

a) visitor training
   - added visitor training section to the Trop Med BSL2 Biosafety Manual

b) improper reuse of sharps.
   - List of Lehrer training session participants

c) missing o-rings
   - o-ring purchase order

d) BSL2 and BSL3 signage
   - BSL2 – PPE requirements
   - Picture -- posted on door – BSL2 – PPE requirements
   - BSL2 – exit requirements
   - Picture -- posted on door – BSL2 – exit requirements
   - BSL3 Suite AB – PPE entry requirements
   - Picture -- posted on door – BSL3 Suite AB – PPE entry requirement
   - BSL3 Suite AC – PPE entry requirements
   - Picture – posted on door – BSL3 Suite AC – PPE entry requirements

e) damaged chairs
   - Picture of laboratory chairs with new seats and chair backs

f) update biosafety manual to include decontamination, incident reporting, medical surveillance, and use of PPE
   - Updated sections are included

g) visitor Risk Assessment Hazard Communication lacks VSV and Ebola

   - VSV Risk Assessment Hazard Communication

   - Ebola Risk Assessment Hazard Communication

10.0. Visitors, Contractors, Inspectors

Visitors include all those who do not have Biosciences Building (BSB) card access, including but not limited to, contractors, vendors, inspectors, guest, family members, etc. If visitor is entering an area of active research, they must sign the Security, Safety, and Emergency Guidelines for Visitors form. Please refer to the JBF Operations Manual for specific information regarding JBF visitor access.

- All visitors, contractors, inspectors, guest, family members etc, must be given an orientation to include, potential hazards and risks associated with entering an active BSL2 laboratory, locations of PPE, telephones, and safety equipment, and emergency procedures.
- All visitors to any laboratory working with infectious agents must complete the form – BioSafety, Biosecurity, and Emergency Response/Incident Response Acknowledgement for All Visitors.

Security

- Visitors must check-in at the security desk and provide a photo ID and cell number.
- Visitors must wear their visitor badge at all times.
- Visitors must be escorted by an authorized individual while in the BSB – BSL-2 laboratory space.
- Visitors may not enter the Vivarium without training and authorization by LAS (BSL-2 containment).

Safety

- Visitors entering a BSL2 laboratory must be escorted by trained authorized personnel.
- Visitors entering a BSL2 laboratory must wear personal protective clothing (covered shoes, labcoat, gloves, etc.) if active research work is taking place during their visit.
- If active infectious research is taking place while a visitor is in the laboratory, the visitor must be provided the Risk Assessment Hazard Communication form pertaining to the specific infectious agent.
- Visitors under the age of 14 years old are not allowed in any laboratory (they are only allowed in the offices, breakrooms, restrooms, and lobby areas).
- Visitors that are not in the facility to conduct research or service laboratory equipment shall not handle any biological material, hazardous chemical, radioactive material, or sharps (needles, scalpels, etc).

Emergency

- Visitors will evacuate the facility if the building's fire alarm is sounded, or if instructed to do so by Security or emergency services (Fire Dept, Police Dept, etc).
- If prompted to evacuate, visitors will use the emergency exits (follow illuminated 'EXIT' signs) and follow their escort to an evacuation gathering area

BioSafety, Biosecurity, and Emergency Response/Incidence Response Acknowledgement for All Visitors to the Department of Tropical Medicine, Medical Medicine, & Pharmacology BSL2 laboratories.

Due to the nature of the infectious agents handled in the TM3P BSL2 Laboratories, all visitors must be alerted to the potential dangers associated with these biological agents. Special precautions must be taken to avoid infection or injury while visiting the laboratory areas.

BioSafety:

- Laboratory testing involves handling of infectious materials with the potential for infections. Diseases can be spread through skin contact, through open lesions, or wounds, or through infectious aerosols and airborne particles.
- Visitors must use appropriate Person Protective Equipment when entering the laboratory.
    - covered shoes
    - disposable lab coat
    - gloves
- Visitors that are not in the facility to conduct research or service laboratory equipment shall not handle any biological material, hazardous chemicals, or sharps (needles, scalpels, etc.)

BioSecurity:

- All visitors must give the Security desk a photo ID, sign in, and obtain a Visitor's Badge.
- Visitors must be escorted by an authorized TM3P user at all times.

Emergency

- In case of an emergency/Incident Visitors must follow proper esit procedures as directed by their escort.

Attestation:

I have read and understand the biosafety, incident response, and biosecurity issues for the TM3P BSL2 laboratories and agree to abide by the laboratory policies. I acknowledge that there are risks and hazards associated with entering the facility and will follow proper safety procedures.

Name of Visitor	Signature and Date
Name of Escort	Signature and Date

JABSOM Biocontainment Facility

JBF Retraining Sign-in Sheet

Sharps re-training session: These members were trained to not recap and reuse needles.

Date: Wednesday 15 January 2020, 3:30 PM, BSB 320N and remote

Name	Signature	Affiliation
Albert To		TRMD / Grad student
Vamey M. Komara		TRMD
Olivia Smith		TRMD / Grad Student
Taylor Tashiro		TRMD / Grad Student
Chih-Yun Lai		TRMD / Instructor
Aquena Ball		TRMD / Grad Student
Caitlin Williams		TRMD / student
Eleanore Chwang		TRMD / postdoc
Brien Haun		CMB / Student
Teri Ann Wong		TRMD
Michael Fanardo		TRMD / Grad Student

100 Matsonford Road
Radnor, PA 19087
t: +1 800 932 5000 f: +1 866 329 2897

Order Confirmation

Click your order number 8361019998 to view current order status

Sold-To-Party
HI UNIV OF JABSOM
TROPICAL MEDICINE
BSB 3R FL
651 ILALO ST RM BSB 320
HONOLULU HI 96813-5525

Ship-To-Party
Lehrer Lab
HI UNIV OF JABSOM
TROPICAL MEDICINE
TROPICAL MEDICINE BSB 3RD FL
651 ILALO ST
HONOLULU HI 96813-5525

Information

Order	8361019998
Date	02/27/2020
Sold To Customer No.	80083505
Ship To Customer No.	80129915
Contact Name	cmlum@hawaii.edu
Telephone	
Email	cmlum@hawaii.edu
Currency	USD
Sales Rep	James Mccord
Cust ref	
Reference	W16739371
PO No	Cassidy Tabata 022720
PO Date	02/27/2020

Information

Term: Standard terms and conditions unless otherwise agreed.
ATTN: TERI WONG--BILL TO: CASSIDY TABATA

Item	Ref.	Catalog Number	Quantity	Unit Price	Ext. Amount
		CC_REFERENCE - CassidyTabata022720			
100		BK345366	2 EA	10.02	20.04
		O-RING CANISTER REPLC AEROSOLVE			
		Shipping Direct from Manufacturer, Estimated Delivery Date 03/04/2020			
		Item Total			20.04
		Total			**20.04**

BIOSAFETY LEVEL 2 LABORATORY

ACTIVE STATUS

Date	Ongoing active status	
Room(s)	336 C	
Protocol(s)		
Lab Director	Vivek Nerurkar	
Contact Information	*Office*: 808-692-1668 (Dr. Nerurkar),	*24/7*: [redacted] (Dr. Nerurkar),
Infectious Agents	**VSV; recVSV**	
Hazardous Chemicals	70% Ethanol; Sodium Hypochlorite Germicidal Bleach; 4% Paraformaldehyde;	
General Occupational Health Requirements	Risk Assessment and Hazard Communication	

Admittance to Authorized Personnel Only

1. No eating, drinking, applying cosmetics, handling contacts, or smoking.
2. Ankle length pants and closed toed shoes are required.
3. Do not bring personal or non-essential items into the room.
4. Follow the ENTRY & EXIT procedures as specified on the signage
5. Report all incidents and emergencies to the Lab Director

Personal Protective Equipment (PPE) Requirements

1. Primary Gloves
2. Gown – Front closing; fully buttoned
3. Don eye protection if executing high risk manipulations that generate aerosols

Emergency & Incident Contacts

Title	Name	Office Phone	24/7 Phone
Lab Director	Vivek Nerurkar	692-1668	[redacted]
PI	Axel Lehrer	692-1614	[redacted]
PI	Sandra Chang	692-1607	[redacted]
Research Assistant	Teri Ann Wong	692-1651	[redacted]
Research Support	Eileen Nakano	692-1612	[redacted]
Kakkako EHSO	Lisa Johns	692-1855	[redacted]
Biosafety Officer	Hubert Oliparus	956-3197	[redacted]
Security	-	692-1911	[redacted]

BSL-2 Room 336 C Laboratory Status Sign

BIOSAFETY LEVEL 2 LABORATORY

ACTIVE STATUS

Date	Ongoing active status	
Room(s)	336 C	
Protocol(s)		
Lab Director	Vivek Nerurkar	
Contact Information	*Office*: 808-692-1668 (Dr. Nerurkar),	*24/7:* [redacted] (Dr. Nerurkar),
Infectious Agents	**VSV; recVSV**	
Hazardous Chemicals	70% Ethanol, Sodium Hypochlorite Germicidal Bleach; 4% Paraformaldehyde; Ethidium Bromide, Sodium Azide	
General Occupational Health Requirements	Risk Assessment and Hazard Communication	

Admittance to Authorized Personnel Only

1. No eating, drinking, applying cosmetics, handling contacts, or smoking.
2. Ankle length pants and closed toed shoes are required.
3. Do not bring personal or non-essential items into the room.
4. Follow the ENTRY & EXIT procedures as specified on the signage
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Emergency & Incident Contacts

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PI	Axel Lehrer	692-1614	[redacted]
PI	Sandra Chang	692-1607	[redacted]
Research Assistant	Teri Ann Wong	692-1651	[redacted]
Research Support	Eileen Nakano	692-1612	[redacted]
JABSOM EHSO	Lisa Johns	692-1855	[redacted]
Biosafety Officer	Hubert Olipares	956-3197	[redacted]
Security	-	692-1911	[redacted]

336

Research Laboratory

CAUTION

For ALL Emergencies
Call Security 692-1911 (24 hrs)

BUILDING: BSB

ROOM: 336 -3

DEPARTMENT: Tropical Medicine

ADMITTANCE TO AUTHORIZED PERSONNEL ONLY

Name	Office Phone	Emergency Phone	Office Location	Position
Dr. Vivek Nerurkar	692-1668	[redacted]	320	Emergency Contact
Dr. Axel Lehrer	692-1614	[redacted]	320B	Emergency Contact
Dr. Sandra Chang	692-1607	[redacted]	320H	Emergency Contact
Teri Wong	692-1651	[redacted]	320-30	Emergency Contact
Albert To	692-1634	[redacted]	320-24	Emergency Contact

Special Notes:

BIOSAFETY LEVEL 2 LABORATORY

PPE to be worn while working in 336

1. Fully fastened front opening long sleeve lab coat

2. Gloves

HIGH-RISK ACTIVE MANIPULATION WITH AEROSOL PRODUCING ACTIVITIES – Additional PPE required

Eye Protection

BIOSAFETY LEVEL 2 LABORATORY

PPE to be worn while working in 336

1. Fully fastened front opening long sleeve lab coat

2. Gloves

HIGH-RISK ACTIVE MANIPULATION WITH AEROSOL PRODUCING ACTIVITIES – Additional PPE required

Eye Protection

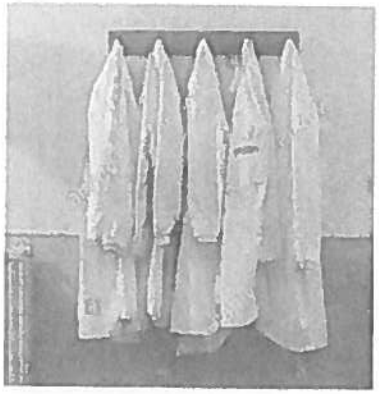
BIOSAFETY LEVEL 2 LABORATORY

Room 336 Exit Procedures

1. Clean up your work area

2. Discard gloves

3. Hang up lab coat



4. Wash hands

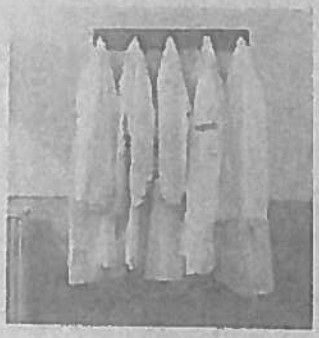
BIOSAFETY LEVEL 2 LABORATORY

Room 336 Exit Procedures

1. Clean up your work area

2. Discard gloves

3. Hang up lab coat



4. Wash hands

BIOSAFETY LEVEL 3 LABORATORY

PPE Required in the Preproom

Wrap-around gown

Booties

Black Gloves

PPE to be Worn in Suite AB

ACTIVE MANIPULATION
Additional PPE required

White Gloves

NO ACTIVE MANIPULATION
Additional PPE required

White Gloves

PAPR

OR

N95

Bonnet

Eye Protection

BIOSAFETY LEVEL 3 LABORATORY

PPE Required in the Preproom

Wrap-around gown

Booties

Black Gloves

PPE to be Worn inside Suite AC

ACTIVE MANIPULATION
Additional PPE required

Tyvec Sleeves

White Gloves

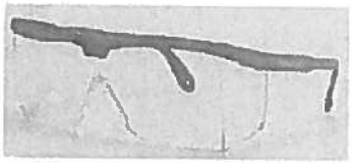
NO ACTIVE MANIPULATION
Additional PPE required

White Gloves

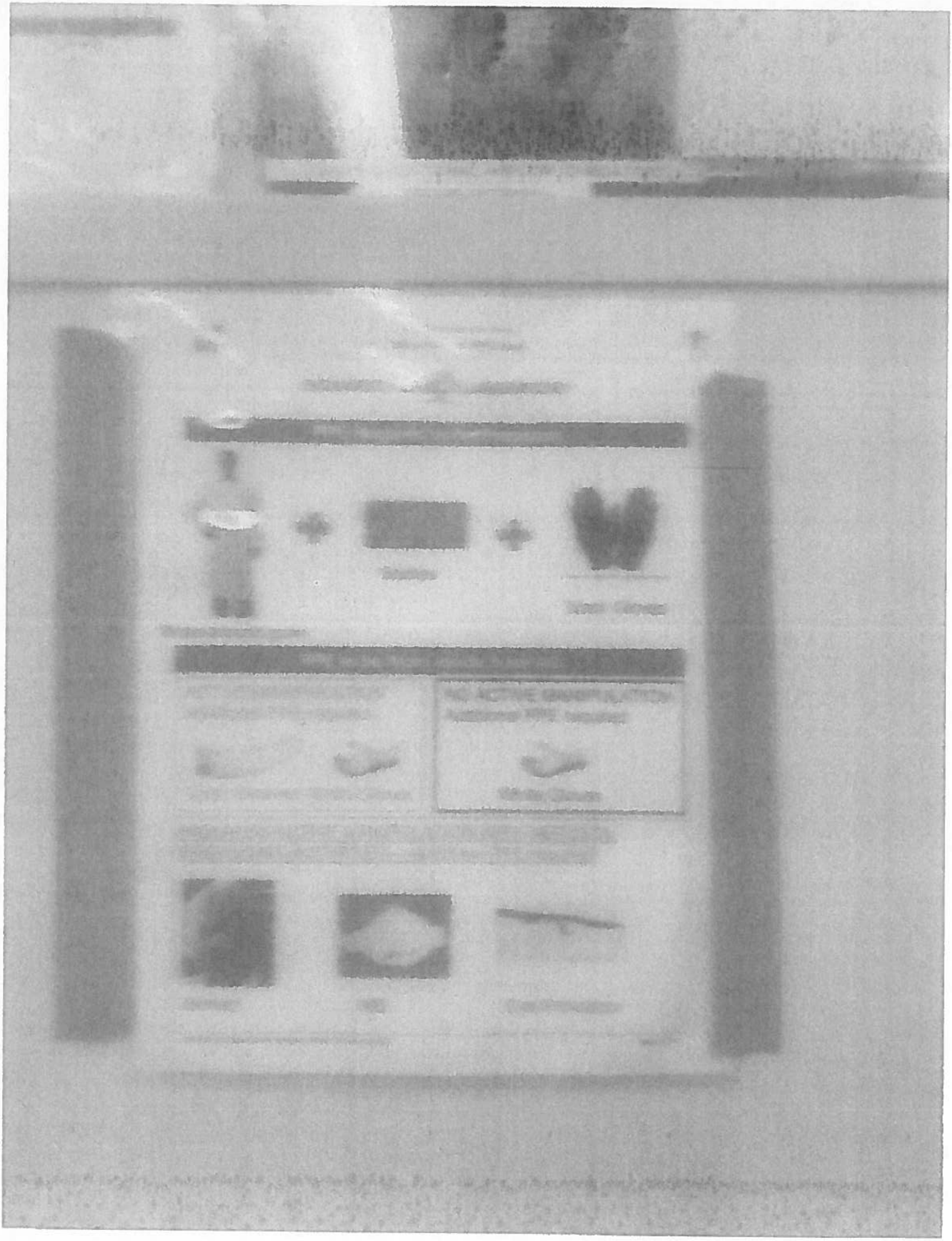
HIGH-RISK ACTIVE MANIPULATION WITH AEROSOL PRODUCING ACTIVITIES -- Additional PPE required

Bonnet

N95



Eye Protection



7.0 Decontamination

All materials or equipment that are contaminated or potentially contaminated with etiologic agents must be rendered nonhazardous (i.e., autoclaved or chemically inactivated) before disposal or washing/storage.

- Non-metal sharps (pipets and pipet tips) – rendered non-infectious by contact with disinfectant, generally 10% bleach for at least 15 – 30 minutes. Rinsed to remove excess bleach, and put in a closeable, leak proof container and placed in a biohazard bag for sterilization.

- Metal sharps (scalpels) – placed in ridge, plastic, closable container and sterilized. Complete EHSO turn-in forms.

- Plastic ware (T-flasks) – placed in a red biohazard bag for sterilization.

- Infected agar plates – placed in two red biohazard bags, both loosely closed, and into a larger red biohazard bag for sterilization.

- Liquid waste – chemically neutralized with freshly made 10% bleach.

- Equipment – surface decontaminate with agent appropriate decontamination solution (Cavicide, 70% ethanol).

Decontamination Method – Autoclave

One of the most effective physical decontamination controls is steam sterilization (autoclave) which generates moisture and high temperature pressurized steam within a sealed chamber.

After the autoclave bag is placed in the autoclave, the operator will ensure that the mouth of each autoclave bag is sufficiently open to allow complete and effective penetration of steam throughout its contents. Autoclaved ("treated") infectious waste are removed from the designated pathogen laboratory and disposed of as trash, only after infectious waste have been certified decontaminated. The primary means of verifying routine sterilization are through the use of chemical indicators --- autoclave tape and labels --- placed on the waste bags. A Biological Indicator, such as a commercial spore test are run in the autoclave and verified as effective by Kaka'ako EHSO on a monthly basis.

Autoclave Certification

Each autoclave run is recorded, and a permanent record of time and temperature of the operation taken.

The performance of the autoclaves in the designated pathogen laboratories are checked by commercial spore test monthly. Autoclave bags containing autoclaved ("treated") infectious waste are held in the pathogen laboratory until the results of the performance tests on that particular run are known.

Commerical Biological Indicators are ampoules containing suspensions of Bacillus stearothermophilus spores in culture medium. These ampoules are placed within normal load, in a position that is difficult for steam to penetrate. After autoclaving, the test ampoule is incubated according to the manufacturer's directions. If the color of medium in the autoclaved ampoule remains purple, sterilization was effective. If color of the medium turns yellow, sterilization was NOT EFFECTIVE and should be repeated.

Liquid Disinfectants

Sodium hypochlorite is a universal disinfectant that is active against all microorganisms, and is commercially available as Clorox™. It is a strong oxidizing agent and extremely corrosive to metals such as stainless steel containers and sinks. Dilute hypochlorite solutions gradually lose strength, necessitating frequent preparation of fresh solutions. A general all-purpose laboratory disinfectant solution should have a concentration of 1 g/liter (1000 ppm) as available chlorine. A stronger solution containing 10 g/liter (10,000 ppm) of available chlorine is recommended for disinfection involving blood spillage and the presence of gross organic matter. Typically, a 5-10% liquid solution is made fresh daily, with a few drops of liquid detergent to assure proper wetting.

The characteristics of chlorine and iodine are similar. Iodophor compounds with 1,600 ppm free available iodine provide a relatively rapid inactivation of all microorganisms, including some bacterial spores. A commonly available iodophor is Wescodyne. The manufacturer of Wescodyne recommends a range of dilution from 1-3 ounces per 5 gallons of water, giving a solution containing from 25-75 ppm of free iodine. At these concentrations, available iodine may be rapidly taken up by any extraneous protein present and will not be an effective sporicide. A solution providing 1,600 ppm iodine is recommended for hand washing or for use as a sporicide.

Disposal

No infectious waste is disposed of until verified safe. Inactivation is the first step in the disposal of etiologic agents or materials that are potentially contaminated. All contaminated or potentially contaminated material must be effectively disinfected or sterilized by an approved procedure. Solid waste is placed in cans, sturdy bags or boxes. Rigid, puncture-resistant, sealable containers are used for packaging "sharps." When wet materials are packaged for disposal, the materials are placed in a leak-proof container. Heavy waste is placed in rigid containers ensuring that the burst strength of the container is not exceeded.

11.0 Incident Reporting

An accident can be defined as "an undesirable event that results in harm to people, damage to property, or loss to process." This includes injuries, occupational disease, damage to University equipment, damage to property, environmental pollution, release of hazardous material or disruption to services.

All suspected and confirmed incidents in the BSL-2 facilities must be reported (file an EHSO Incident Report). Immediate action may be necessary depending on the incident (e.g. fire, medical emergency); use judgment. A completed EHSO Incident Report Form and any additional forms as required (See Forms) should be submitted within 48 hours of the incident. Users are expected to report all abnormal occurrences within the laboratory, even if the incident is thought to be minor.

Submit the completed report to your PI. The Incident Report will be reviewed by the PI and then submitted to the Chair T3MP. The Chair T3MP will forward the report to EHSO and any other individuals as he sees appropriate. All reviewers shall review the incident and provide input on corrective actions and remediation. The AVS Manager will report the incident to the UH IACUC and animal welfare compliance office, as applicable.

JABSOM INCIDENT REPORT

Date(s) of Occurrence: _____ **Time:** _____ **Location:** _____

Type of Incident (check all that apply):

- ☐ Near-miss
- ☐ Spill/Release
  - ○ Minor
  - ○ Major
- ☐ Exposure
- ☐ Inhalation
- ☐ Ingestion
- ☐ Needlestick
- ☐ Animal Bite
- ☐ Animal Escape
- ☐ PPE Failure
- ☐ Equipment Failure OR Alarm
- ☐ Facility Abnormality
- ☐ Injury
- ☐ Medical Emergency
- ☐ Theft
- ☐ Leak OR Flood
- ☐ Fire OR Fire Alarm
- ☐ Pest (non-lab insect or rodent) Complaint
- ☐ Smells/Odors
- ☐ Security Breach
- ☐ Unauthorized Entry
- ☐ Misconduct
- ☐ SOP Violation
- ☐ Other:_____

Description of Incident (use as much space as necessary):

_____

Corrective Actions Taken (use as much space as necessary):

_____

Was medical attention required: _____ Yes _____ No
Name of Attending Health Professional:_____

Report Completed By (Title/Name/Signature):

_____

Date: _____ Phone: _____

Witnesses (Title/Name/Signature):

_____

Primary Investigator (Name/Signature/Date):

_____

JABSOM EHSO (Name/Signature/Date):

_____

- JABSOM Security will verify Visitor with government issued photo identification and verification from home institution or office.

3.8. Access for Emergency Response Personnel: Emergency Crews and Fire Personnel may respond to emergencies in the area but may not enter unless the JBF Director authorize entry. Refer to the specific procedures in the JBF Incident Response Plan.

3.9. Recordkeeping: The JBF Director, JBF Associate Director, and the JBF Supervisor shall keep all access documents/forms/records on file for a minimum of three (3) years. All training and certifications will be reviewed at least annually, and a current record of authorized individuals maintained.

SECTION 4: JBF Occupational Health Program / Medical Surveillance Plan

4.1. Occupational Health Program: All JBF users (participate in active manipulation of RG3 agents) must be enrolled in the UH/JABSOM Occupational Health Program in order to access the JBF. They must complete the health history questionnaire and be medically cleared by a health professional familiar with Occupational Health and receive documented training on the occupational health provided by UH Biosafety and EHSO.
The purpose of the Occupational Health program is to:

- Provide risk assessment and hazard communication – guide for selecting appropriate medical preventions and countermeasures, and defining health hazards associated with the RG3 agent and disease associated symptoms, and appropriate treatment
- Provide medical evaluations and screening for personnel who will work in the facility -- ensure individual employees are physically fit for the nature and extent of the work to be undertaken.
- Perform periodic medical reassessment of employees (at least once every three years; or if the individual's medical condition and/or work conditions change) to determine if medical conditions associated with employment are prevalent and, if so, to undertake definitive measures to alleviate them.
- Provide a medical surveillance program for all personnel working in the facility
- Respond and conduct follow up treatment to personnel exposure

4.1.1. Risk assessment and Hazard Communication

- Complete the JBF Risk Assessment and Hazard Communication Form
- Read and understand the JBF Risk Assessment and Hazard Communication Supplement describing the research infectious agents.

4.1.2 Enroll in the AVS Occupational Health and Safety Program -- https://researchcompliance.hawaii.edu/programs/animal-veterinary-services/occupational-health-safety-program/

- Complete Health History Questionnaire – form I
- Complete Health Professional Medical Evaluation to Principal Investigator – form B
- Complete Registration Date Sheet – form D
- Complete Authorization for Release of Medical Information – form E
- Send in forms to Straub Occupational Health Services at email, Jennifer.oldershaw@straub.net AND dora.sakata@straub.net, Fax (808)-529-4950, or mail to:

Straub Occupational Health Services
800 S. King Street, 3rd Floor
Honolulu, HI 96813

· Send proof of enrollment and suitability to work in the JBF to Eileen at etnakano@hawaii.edu

- Complete 3M respiratory evaluation survey
  - Email Eileen at etnakano@hawaii.edu to receive an access number and instructions to take survey
  - Send Eileen proof of completed survey and suitability to wear an N95 or PAPR.

4.1.2. Renewals In OHSP through Straub

You are required to be reassessed after three years or if there have been changes in health status and/or work assignment since your first enrollment. You must complete the following forms:

· Complete Health History Questionnaire – form I
· Complete Health Professional Medical Evaluation to Principal Investigator – form B
· Complete Registration Date Sheet – form D
· Complete Authorization for Release of Medical Information – form E
· Send in forms to Straub Occupational Health Services at email, Jennifer.oldershaw@straub.net AND dora.sakata@straub.net, Fax (808)-529-4950, or mail to:

Straub Occupational Health Services
800 S. King Street, 3rd Floor
Honolulu, HI 96813

4.1.3. If you choose to decline medical evaluation and/or immunizations recommended by the Health Professional, complete the following forms:

· Complete Declination Form C
· Complete Health Professional Medical Evaluation to Principal Investigator – form B
· Complete Registration Date Sheet – form D
· Complete Authorization for Release of Medical Information – form E
· Send in forms to Straub Occupational Health Services at email, Jennifer.oldershaw@straub.net AND dora.sakata@straub.net, Fax (808)-529-4950, or mail to:

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Honolulu, HI 96813

4.1.3. Additional considerations

4.1.3.1. Pregnancy

Staff, who want to start families, are encouraged to inform their supervisors or principal investigators and Employee Health. Employees are urged to discuss exposure issues with their supervisors or principal investigators regarding associated risks of research being conducted. Women who are pregnant or become pregnant will be given advice about precautions that may be necessary. It is recognized that exposure to certain infectious agents may adversely affect a fetus during pregnancy if the mother is infected with the agent. Therefore, if pregnancy is possible while you are working in an infectious disease laboratory or laboratory engaged in work with infectious agents you should consult your health professional

through the process described for medical assessments. The Employee Health is also available for questions regarding the potential harm from the biological agents present within your laboratory.

Employee Health is a resource for all staff to discuss questions or concerns they may have about risks in their work environment. Employee Health may also act as a liaison between pregnant employees and their respective supervisors or principal investigators.

4.1.3.2. Other restrictions

Restrictions or recommendations will be made on an individual basis after discussion with the Employee Health Physician and the employee's personal physician. Examples of conditions that may warrant special precautions are HIV infection, immunosuppressive conditions or drug therapy that suppresses the immune system. Therefore, if you have any of the above conditions, you must inform your physician and Employee Health about the situation.

4.2. Medical Surveillance Plan: Personnel working in ABSL-3 and BSL-3 research facilities face occupational health risks due to biological agents they may possibly be exposed to during the course of their job duties. Participation in JBF's Medical Surveillance Program and vigilant adherence to appropriate Exposure Control Plans in the laboratory are required to protect the health and safety of these individuals.

JBF Medical Consultants:

Dr. Cecilia Shikuma MD: 692-1328 (O); 781-1832 (C)

Dr. Bruce Shiramizu, MD: 282-6759 (C)

The Medical Surveillance Program assesses the exposure risk and health status of BSL-3/ABSL-3 personnel to prevent occupational injury or illness.

It may include:

- Medical assessment of each participant's ability to safely work in the BSL-3/ABSL-3 facility
- Medical recommendations for appropriate medical screening, immunizations, and protective measures
- Periodic medical assessment and monitoring to confirm safety practices are effectively preventing occupational injury or illness

4.2.1. BSL-3/ABSL-3 Risks: Personnel working in the BSL-3/ABSL-3 facilities face occupational health risks due to work with agents that:

- Have a known potential for aerosol transmission
- May cause serious or potentially lethal infections
- Are indigenous or exotic in origin

Primary hazards to personnel working with BSL-3/ABSL-3 agents relate to:

- Autoinoculation (intact/broken skin or mucous membranes)
- Ingestion
- Exposure to infectious aerosols

Additional requirements may include the following:

- Tuberculosis (TB) screening (if entering a facility where TB work is conducted)
- Respirator Medical Questionnaire (if respirator use is required)
- Latex Allergy Questionnaire (if history of latex allergy)
- Immunizations (based upon specific agents in use)
- Medical referral (if indicated based upon initial surveillance review)

4.2.2. Vaccinations: All laboratory personnel will be offered vaccination against specific medically important microbial agents. For example, vaccination against Japanese Encephalitis virus or Yellow Fever virus will be offered to those working with such agents or related flaviviruses. Individuals who refuse vaccination will be required to provide documentation of prior vaccination or to sign a declination stating that they are declining the offer of vaccination.

Vaccinations, if available, are provided as necessary at no cost to the employee. Communications to the employee must include: identification of the agents, vaccination availability, justification for offering (or not offering) the vaccination, and means to receive the vaccination.

At minimum, all individuals must have had or declined to have Hepatitis B virus vaccination.

4.2.3. Respiratory Protection and Medical Monitoring: All personnel must be able to be medically qualified to wear respiratory protection. A respiratory program has been established for the JBF which consists of wearing an N95 or Powered Air Purifying Respirator (PAPR) in the BSL-3 or ABSL-3 depending upon agent and risk assessment. At a minimum, an N95 respirator is required when conducting activities which are considered high risk due to the possibility of generating aerosols (i.e., tissue homogenization, high speed centrifugation, and processing of tissues or cultures which have high, concentrated viral/bacterial content). In the ABSL-3, respiratory protection is required in areas which house infected animals.

4.2.4. Antibody testing: Antibody testing maybe required. Clearly defined and conveyed: when antibody testing is required, clearing defined who is tested, who collects samples, tests, and reports; what are the conditions/schedule for testing and reporting, and what is the reponse to negative or positive results.

4.2.5. High Risk Personnel: Persons, who are at increased risk of acquiring infection or for whom infection may have serious consequences (i.e., pregnant women, immuno-compromised health status, etc.) are encouraged to self-identify and shall not be permitted entry into the facility without proper approval. All individuals at increased risk must seek advice of an occupational health professional to help them evaluate their medical risks and the option of other non-Level 3 work will be provided to the worker.

4.2.6. Procedure for Reporting Illness in Laboratory Workers: All laboratory employees working with infectious agents MUST report any illness to the Laboratory Director. Included in the report will be:

- Symptoms
- Any laboratory incident or unusual circumstanced preceding the illness
- Laboratory activities preceding the illness
- Infectious agents used
- If the illness could be work related or occurs on a weekend, personnel should contact the JBF Director on cell phone or at home. In the employee is unable to contact the Laboratory Director, then contact the Laboratory Supervisor.

9.0. Occupational Health/Medical Surveillance Program

Medical surveillance of laboratory personnel can help ensure that workers who are at risk of occupational exposure to infectious agents and who develop symptoms of illness receive time and appropriate medical evaluation and treatment. This benefits the laboratory worker but also helps to prevent further transmission, alerts medical personnel to the potential risks and ensures prompt attention to the infection.

Do not enter the BSL2 laboratory or Vivarium facility if you have a fever or are showing symptoms of an illness.

Medical surveillance has several components:

- Every person who works in the BSB Trop Med facilities with an infectious disease agent must be familiar with the signs and symptoms of infection with all agents being handled in the laboratory and should monitor themselves for these symptoms. Principal Investigator is responsible to communicate this hazard to all staff with potential exposure.

- Trop Med personnel working with infectious agents should be offered agent specific vaccine or prophylactics if available.

- Breach in laboratory procedures must be immediately reported and evaluated.

    - In the event of a breach in procedures, the laboratory worker should immediately implement the applicable laboratory procedures for emergency management and notify the supervisor. The supervisor and other appropriate personnel will evaluate the breach in procedure to determine if an exposure has occurred and to plan appropriate follow-up, including any further diagnostic evaluation such as collection of an acute-phase serum ample.

    - If it is determined that an exposure has occurred, the laboratory work should be instructed to be vigilant for the development of relevant symptoms (this includes self quarantine, temperature monitoring, etc.). The worker should immediately notify the supervisor if symptoms develop.

    - Workers with an exposure and no symptoms of illness should discuss the need for activity restriction with the appropriate medical advisor.

    - Any exposed laboratory worker who develops symptoms should immediately inform the appropriate person and report to the designated location for clinical evaluation.

    - Laboratory worker who develop symptoms and who have no recognized exposure should immediately contact the supervisor. The supervisor will notify the appropriate occupational health personnel

There are certain medical conditions for which exposure to certain infectious agents may adversely affect them.

- Pregnancy – exposure to a fetus during pregnancy. If pregnancy is possible while working in an infectious disease laboratory or laboratory engaged in work with infectious agents, researcher should consult their Principal Investigator or supervisor.
- Immuno-compromised – increased risk of infection, possibility of more serious illness. Researcher is encouraged to self-identify to their Principal Investigator or supervisor.

5.0 Apparel - PPE

5.1. Standard apparel

- Long pants and skirts are worn in the laboratory area. Shorts, kilts, and mini-skirts are not considered appropriate attire for working in the laboratories and they do not protect the legs against splashing or spill hazards.
- Closed toe shoes are worn; no open-toe shoes, sandals or slippers are permitted in the work area.

5.2. PPE

PPE is specialized clothing or equipment worn by employees for protection against a hazard. PPE is considered "appropriate" only if it does not permit potentially infectious material to pass through or reach the employee's clothes, undergarments, skin, eyes, mouth, or other mucous membranes under normal conditions of use and for the duration of time which the protective equipment will be used. The following items are required to be worn while in within certified BSL2 laboratory space:

- **Splash-resistant lab coats** (gowns, aprons) should be used by laboratory staff members while working with biological materials/waste, hazardous chemical materials/waste. Lab coats should have a closed front or an overlapping front. Lab coats must not be worn or used outside of the laboratory.

- **Gloves** (disposable, single use) provide a barrier between infectious agents and the skin. They should worn at all times while doing active work in the laboratory. Disposable gloves should be replaced when contaminated, torn or punctured, and between patient contacts. Disposable gloves may not be washed or decontaminated for reuse; these are single use only. Gloves are discarded into a labeled biohazard container. Gloves must be removed before leaving the laboratory and before entering any common areas, including elevator lobbies, stairwells, office suites, break rooms, bathrooms, and before contacting communal items such as phones, computers, door handles, elevator buttons, etc. Note: The laboratory also provide specific gloves for handling hot or cold items. These are marked and left in the laboratory; they are not a single use item.

- **Eye protection**, such as safety glasses or face shields, are available in the laboratory for any procedures where splashing or aerosol generation is a hazard; whenever eye, nose, or mouth contamination can be reasonably anticipated (e.g., retrieving cells from the liquid nitrogen cryo-storage tanks, working with infectious agents outside of a BSC).

- Survey the Manipulation Suite before leaving. Verify that equipment, samples, and reagents are stored at the appropriate conditions.
- Inspect gown for any signs of contamination. If contamination is suspected, immediately doff gown and replace with clean gown (extra supplies are kept in a drawer within the Manipulation Suite).
- If a transport carrier was used, clean the empty carrier with agent appropriate decontamination solution (e.g., Accel TB, 70% ethanol, Cavicide) and remember to transport back into the Prep Room when exiting the Manipulation Suite so it is ready for the next use.
- Any samples containers that need to be transferred to the Prep Room freezers are sprayed with disinfectant, placed in secondary containment, and the secondary container sprayed with agent appropriate decontamination solution.
- Remove secondary gloves (and sleeves if worn) and place in biohazard waste bin.
- Remove single-use, secondary gowns and dispose of as required by your agent specific SOP.
- Spray down front and arms of re-usable gowns with agent appropriate decontamination solution.
- If PAPRs are used, thoroughly wipe down PAPRs (helmet, belt, hose, battery pack, exposed portion of shroud, etc.) with agent appropriate decontamination solution prior to leaving the suite.
- If N95 respirator is used, remove and dispose in biohazard waste bin.
- If wearing a bonnet, remove and dispose in biohazard waste bin.
- Remove eye protection and wipe with agent appropriate decontamination solution.
- Thoroughly spray until wet booties and primary gloves with agent appropriate decontamination solution (e.g., Accel TB, 70% ethanol, Cavicide).
- Remove Active Manipulation Sign from door.
- Turn off the light.
- Exit into Prep Room.

6.1.5. Exit Procedures for the BSL-3 Prep Room

- Remove the Active Manipulation sign from the Suite door.
- If necessary, transfer samples into the freezer from your secondary container. Thoroughly disinfect the secondary container and store on the shelf in the Prep Room.
- Doff PAPRs and plug in PAPR battery packs. If necessary, wipe down PAPRs a second time, according to your agent specific Risk Assessment.
- Doff eye protection and store appropriately. .
- Survey equipment used located in the Preparation Room before leaving.
- Fax out the appropriate laboratory notes to code 113 (JBF Director).

- If no longer needed, doff re-usable gowns and dispose of in biohazard waste bin.
- If you are leaving late in the day, prepare 50 mL drain disinfectant for the sink traps and record this on the Sink Log.
- If work is completed for the day, proceed to the biohazard waste bin near the hand washing sink and remove booties with gloved hands.
- Doff primary gloves and dispose of in the biohazard waste bin.
- Wash hands with soap thoroughly.

Vesticular stomatitis virus (VSV)

Some of the following information was taken from the CDC and other government websites: Vesticular stomatitis virus (VSV) is a bullet-shaped, enveloped virus, approximately 70 nm in diameter and 170 nm in length, and has a single-stranded, negative-sense RNA genome. It is a member of the *Rhabdoviridae. VSV has eight main serotypes: Indiana, New Jersey, Cocal, Alagoas, Isfahan, Chandipura, Maraba, and Piry.*

VSV is the most common vesicular disease of livestock in the Americas and was first isolated in 1925, although VSV has been reported since the 1800s. Mexico, Central America, and northern South America continue to experience endemic cycles of VSV (VSV-NJ and IND-1), while infections are reported less frequently in northern Mexico and the United States.

This viral disease primarily infects horses and cattle, while occasionally infecting swine, sheep, goats, llamas, alpacas, and people who handle infected affected animals. In infected livestock, VSV causes blister-like lesions to form in the mouth and on the dental pad, tongue, lips, nostrils, hooves, and teats. These blisters swell and break, leaving raw tissue that is so painful that infected animals generally refuse to eat and drink and show signs of lameness. Severe weight loss usually follows, and in dairy cows a severe drop in milk production commonly occurs. Infected dairy cattle can appear to be normal and will continue to eat about half of their feed intake.

Most human infections with Indiana and New Jersey VSV serotypes appear to be subclinical. In patients that show clinical manifestations, the initial symptom is high fever that is often biphasic. Subsequent symptoms are "flu-like" including severe malaise, headaches, myalgia, arthralgia, retrosternal pain, eye aches, and nausea. Vesicle formation on the oral mucosa, lips, and nose is possible, but these are rare symptoms of vesicular stomatitis (VS). Human infections with Cocal virus have not been documented, whereas Alagoas virus infections in humans have only been reported in Brazil, with flu-like symptoms that resolved within 3-4 days. Chandipura virus has only been reported in India, where it mostly infects children. Symptoms include fever, sensory disorders, convulsions, vomiting, diarrhoea, and encephalitis leading to coma and death. Reports on the pathogenicity of Piry virus in humans are inconsistent and virtually absent from the primary literature; however, Piry virus is closely related to Chandipura virus, based on glycoprotein sequence analysis. The pathogenicity of Maraba virus in humans is also not known. Isfahan virus was associated with human infections in Iran; however, the virus has not been definitively linked to human illness.

Occupational Infections

Infections with VSV were seemingly common among laboratory workers and animal handlers before the advent of modern biological safety procedures and equipment (before 1980). In one study, 54 of 74 laboratory workers or animal handlers had antibodies to these viruses. Humans contract vesicular stomatitis by coming into contact with lesions, saliva, or nasal secretions from infected animals. In laboratories, aerosol transmission and after accidental inoculation (needlestick injuries) have resulted in vesticular stomatitis. This is in contrast to a later study which reported only 17 out of 133 people exposed to the virus became infected. This suggests with modern safety precautions clinical cases are rare. However, others point out that human infections may be underreported as they can easily be misdiagnosed as influenza. In the study above, 31 of the 54 seropositive laboratory workers or animal handlers reported having had a mild, acute illness consistent with vesicular stomatitis. In most cases, clinical cases have had no serious consequences. Laboratory infections have resolved without complications, and no deaths have been reported.

Natural Modes of Infection

The transmission of vesicular stomatitis is incompletely understood. The relative importance of the various transmission routes in each situation is also sometimes unclear. Insect vectors are thought to introduce VSV into populations of domesticated animals. Sand flies (*Lutzomyia* sp.), blackflies (family *Simuliidae*) and *Culicoides* midges can act as biological vectors. Sand flies seem to be important vectors in endemic areas, but have a limited flight range and are not thought to spread these viruses long distances. Blackflies are believed to be particularly important vectors in parts of the western U.S. Where the virus originates, before entering livestock populations, is still uncertain. Once animals develop lesions, however, insects may become infected by feeding on viruses in these lesions or contaminated secretions. In addition, infected blackflies can transmit VSV to other blackflies feeding at the same time on a host, even if the host is not infected. Transovarial transmission has been demonstrated in sandflies and blackflies in the laboratory, and may be possible in Culicoides. It might contribute to virus overwintering in cold climates.

People can be infected by contact with lesions or secretions from infected animals, particularly vesicular fluid and saliva, or when manipulating VSV in the laboratory. Some people are probably infected through insect bites, as antibodies to these viruses are common in endemic regions. In healthy human cells the virus cannot reproduce, likely because of the interferon response, which allows the cells to adequately respond to viral infection

Laboratory Safety and Containment Recommendations

VSV serotypes, Indiana, Cocal, Alagoas, New Jersey, Isfahan and Maraba viruses are classified as Risk Group 2 human pathogens, requiring BSL2 containment.

With respect to infected animals, VSV is shed in vesicle material. Viruses from lesions in the mouth and on the muzzle can contaminate saliva, and to a lesser extent, nasal secretions. However, VSV has also been detected in the saliva of some experimentally infected horses that did not have oral lesions. Vesicular stomatitis viruses are not considered to be shed in feces, urine or milk, although they have been detected occasionally in the feces of symptomatic, experimentally infected swine. Livestock can be infected experimentally by aerosols in the laboratory, but this route did not result in skin lesions in most species. VSV does not appear to cross the placenta or cause fetal seroconversion.

VSV in saliva was reported to survive for 3-4 days on milking pails, mangers and hay. Viruses dried onto glass, plastic or stainless steel in the laboratory lost a great deal of infectivity within the first 1-6 days at 22°C, although some infectious virus was still recovered after 2-8 days. However, survival in liquid medium that contained organic material (i.e., cell culture medium with 5% fetal bovine serum) was prolonged, especially at cold temperatures. These suspensions did not lose significant infectivity for at least 4 weeks at 4°C. Approximately 90% of infectious virus disappeared during the first 8 days in such suspensions incubated at 28°C, but some viable viruses were still present after 4 weeks. At 37°C, 90% of infectivity had been lost by 3 days, and no live viruses could be detected after 21 days. Only 10% of the viruses suspended in cell culture medium without serum were still viable by 4-12 days at 4°C.

Special Issues

In addition to live VSV Indiana (laboratory adapted), the laboratory also works with recombinant VSV. The biosafety level of a viral vector defaults to the Risk Group of the wild type viral stain from which the vector is derived. This biosafety level is applied during preparation, during use in cell culture systems

Agent Appropriate Decontamination Solutions

Resistance to physical and chemical action Temperature: Inactivated by 58°C for 30 minutes pH: Stable between pH 4.0 and 10.0 Chemicals/Disinfectants: Sensitive to formaldehyde, ether and other organic

solvents; chlorine dioxide, formalin (1%), 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde, 2% sodium carbonate, 4% sodium hydroxide, and 2% iodophore disinfectants, all effective disinfectants.
Survival: Inactivated by sunlight; survives for long periods at low temperatures

is recommended for processing field collected mosquito pools whereas BSL-3 and ABSL-3 practices, containment equipment, and facilities are recommended, for all manipulations of WNV cultures and for experimental animal and vector studies, respectively.

Dissection of field collected dead birds for histopathology and culture is recommended at BSL-3 containment due to the potentially high levels of virus found in such samples. Non-invasive procedures performed on dead birds (such as oropharyngeal or cloacal swabs) can be conducted at BSL-2.

Agent Appropriate Decontamination Solutions

WNV is susceptible to many disinfectants: 3-8% formaldehyde, 2% glutaraldehyde, 2-3% hydrogen peroxide, 500 to 5000 ppm available chlorine (5-10% bleach), 70% alcohol, 1% iodine, 5% quaternary ammonium compound, phenol iodophors, and other organic solvents and detergents.

Ebola Virus

Some of the following information was taken from CDC and other US government websites. Ebola virus is a single-stranded negative-sense RNA, enveloped virus, belonging to the Filoviridae family. Within the genus Ebolavirus there are 6 species, Ebola virus (EBV), Sudan virus, Taï Forest virus, Bundibugyo virus. Reston virus, and Bombali virus. Filovirus particles form long, sometimes branched, filaments of varying shapes, as well as shorter filaments, and may measure up to 14,000 nanometers in length with a diameter of 80 nanometers.

Ebola virus was first discovered in 1976 near the Ebola River in what is now the Democratic Republic of Congo. Since its discovery, the virus has caused a number of small and large outbreaks. During the last 2014 outbreak in West Africa, approximately 20,200 cases were reported with close to 8,000 deaths. Currently there is a new EBV outbreak in the Democratic Republic of the Congo.

The virus spreads to people initially through direct contact with the blood, body fluids and tissues of animals infected with EBV. Ebola virus then spreads to other people through direct contact with body fluids of a person who is sick with or has died from Ebola Virus Disease (EVD). This can occur when a person touches these infected body fluids (or objects that are contaminated with them), and the virus gets in through broken skin or mucous membranes in the eyes, nose, or mouth. People can get the virus through sexual contact with someone who is sick with EVD, and also after recovery from EVD. The virus can persist in certain body fluids, like semen, after recovery from the illness.

Treatment for EVD is mainly supportive care - rehydration with oral or intravenous fluids. There are no FDA approved treatment, although many are in preclinical or clinical trials. An EBV vaccine has been approved by the FDA, but only administered under special circumstances. Additional EBV vaccines are in development, including one developed by the Lehrer laboratory in Tropical Medicine, Medical Microbiology, & Pharmacology.

Occupational Infections

Health care workers, those in direct contact with EBV infected patients are at significantly higher compared to laboratory staff for contracting EVD. Completion data from all EBV outbreaks over the past 50 years suggest a 30% difference between those with direct contact of EBV patients and laboratory works (128 vs 19; Selvaraj, IDSA, 2018; 218(S5):S679–89). The CDC recommends laboratory workers follow all OSHA's Bloodborne Pathogens Standard and wear appropriate PPE. Generation of aerosols should be kept to a minimum.

Natural Modes of Infection

Some fruit bats are considered to be the natural host of Ebola virus. In Africa, infection has been documented through the handling of infected chimpanzees, gorillas, fruit bats, monkeys, forest antelope and porcupines found ill or dead in the rainforest. Subsequently, the virus spreads from person to person,

through direct contact (such as through broken skin or mucous membranes in the eyes, nose, or mouth) with:

- Blood or body fluids (urine, saliva, sweat, feces, vomit, breast milk, and semen) of a person who is sick with or has died from Ebola virus disease (EVD).
- Objects (such as clothes, bedding, needles, and medical equipment) contaminated with body fluids from a person who is sick with or has died from EVD.
- Infected fruit bats or nonhuman primates (such as apes and monkeys).
- Semen from a man who recovered from EVD (through oral, vaginal, or anal sex). The virus can remain in certain body fluids (including semen) of a patient who has recovered from EVD, even if they no longer have symptoms of severe illness. There is no evidence that Ebola can be spread through sex or other contact with vaginal fluids from a woman who has had Ebola.

Symptoms may appear anywhere from 2 to 21 days after contact with the virus, with an average of 8 to 10 days. Primary signs and symptoms of Ebola often include some or several of the following:

- Unexplained acute fever (greater than 38.6° C or 101.5° F)
- Aches and pains, such as severe headache, muscle and joint pain, and abdominal (stomach) pain
- Weakness and fatigue
- Gastrointestinal symptoms including diarrhea and vomiting
- Abdominal pain
- Unexplained hemorrhaging, bleeding or bruising
- Other symptoms may include red eyes, skin rash, and hiccups (late stage).

Many common illnesses can have the same symptoms as EVD, including influenza (flu), malaria, or typhoid fever.

The virus can remain in areas of the body that are immunologically privileged sites after acute infection. These areas include the testes, interior of the eyes, placenta, and central nervous system, particularly the cerebrospinal fluid. Whether the virus is present in these body parts and for how long varies by survivor.

Ebola virus can survive on dry surfaces, like doorknobs and countertops for several hours; in body fluids like blood, the virus can survive up to several days at room temperature. Cleaning and disinfection should be performed using a hospital-grade disinfectant.

Laboratory Safety and Containment Recommendations

Filoviruses are highly infectious agents and strict precautions must be applied when handling specimens for diagnosis. Laboratory tests on the non-inactivated virus present an extreme biological risk. Proper precautions and engineering control (i.e. facility and equipment) must be observed at all times, in accordance with the issues identified in the risk assessment for each procedure. Biosafety recommendations for laboratories conducting diagnostic testing for EVD with appropriate biosafety BSL3/BSL4 facilities. Use appropriate Personal Protective Equipment (PPE) when handling the specimens before inactivation: gloves, fit-tested masks such as N95 Respirators and Filtering Face Piece (FFP) 3, Powered Air Purifying Respirators (PAPR) if fit-testing fails, full face shields, and disposable impermeable gowns.

- Virus isolation should be done only in a maximum containment BSL4 laboratory. Ensure safe and secure handling and storage of the virus isolates and other specimens from accidental or deliberate release. The inactivation of specimens, depending on the detection protocol used, should be performed under BSL3 conditions.

- For non-inactivated samples, RT PCR and enzyme-linked immunosorbent assay (ELISA) testing can be performed at a BSL3 laboratory. Specimens for either PCR or ELISA testing should be processed inside a Class III biosafety cabinet (glovebox) with current certification in a separate laboratory area.

- If samples have been inactivated (i.e. cell lysis) RT PCR and ELISA testing can be performed at a BSL2 laboratory.

Agent Appropriate Decontamination Solutions

- EBVis susceptible to many disinfectants: 3-8% formaldehyde, 2% glutaraldehyde, 2-3% hydrogen peroxide, 500 to 5000 ppm available chlorine (5-10% bleach), 70% alcohol, 1% iodine, 5% quaternary ammonium compound, phenol iodophors, and other organic solvents and detergents. For a list of EPA's Registered Antimicrobial Products that Meet the CDC Criteria for Use Against the Ebola Virus see: https://www.epa.gov/sites/production/files/2018-01/documents/2018.10.01 lstl pdf

Special Issues

It should be noted that all serum samples involved in this project will be obtained from afebrile, healthy patients. Pooled samples will be PCR screened for the presence of EBV prior to transit to the US. No PCR positive sample from the pool will be sent to Hawaii. It is therefore felt, with respect to EBV, it is highly unlikely these samples will contain infectious material.

If a sample sent to JABSOM is found to be EBV positive, immediately do the following:

- Put the sample in a separate box, and label the box, "suspected EBV positive". Return the sample to the -80 ° C freezer.
- Inform the JBF Supervisor and the JBF Director.
- JBF Director will inform the BioSafety Officer, and the Federal Select Agent Program Office
  - 404-718-2000 or 404-488-7100 (after hours)
  - Select Agent Program Office will advise.
    - Hold
    - Destroy
    - Transfer
    - Fill out APHIS/CDC Form 4 (APHIS/CDC Form 4A)
- Begin Medical surveillance