HOW TO SAFELY WORK WITH INFECTIOUS VETERINARY DIAGNOSTIC SAMPLES
DISCLAIMER

The findings and conclusions in this presentation are those of the author(s) and do not necessarily represent the views of the Centers for Disease Control and Prevention and Federal Select Agent Program.
AGENDA

• Risk groups
• Zoonotic pathogens
• Biosafety program
• Containment
• Risk assessment
• Facility practices and procedures
• Working with diagnostic samples
• Disinfection
**RISK GROUP CLASSIFICATIONS**

**BASED ON NIH AND WHO GUIDELINES**

**RG 1**
Not associated with disease in healthy adult humans
No or low individual and community risk

**RG 2**
Associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available
Moderate individual risk, low community risk

**RG 3**
Associated with serious or lethal human disease for which preventive or therapeutic interventions may be available
High individual risk, low community risk

**RG 4**
Likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available
High individual and community risk
### HHS Select Agents and Toxins

1. *Akbiria* [8]
4. *Botulinum neurotoxin* [21]
5. *Clostridium perfringens* [24]
6. *Coxiella burnetii* [14]
8. *Diabetesis* [22]
9. *Eastern Equine Encephalitis virus* [47]
10. *Ebole virus* [1]
12. *Lassa fever virus* [13]
13. *Lyssa virus* [17]
14. *Marburg virus* [14]
17. *Rickettsia prowazekii* [18]
18. *SARS-associated coronavirus* (SARS-CoV) [21]
19. *Saxitoxin* [16]

### South American Haemorrhagic Fever viruses:

- Chapare
- Guanarito
- Jarrin
- Machupo
- Señora

### Tick-borne encephalitis complex (flavivirus)

- Far Eastern subtype [22]
- Siberian subtype [25]
- Kyasanur forest disease virus [25]
- Omkham hemorrhagic fever virus [25]
- Vector major virus (Sindbis virus) [11]
- Vector minor virus (Nasarum) [11]

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### USDA Select Agents and Toxins

37. *Bacillus anthracis* Pasteur strain
38. *Brucella abortus*
39. *Brucella melitensis*
40. *Brucella suis*
42. *Burkholderia pseudomallei* [11]
43. *Hendra virus*
44. *Nipah virus*
45. * Rift Valley fever virus*
46. *Venezuelan equine encephalitis virus* [47]

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### USDA Select Agents and Toxins

47. *African horse sickness virus*
48. *African swine fever virus*
49. *Asian influenza virus* [47]
50. *Classical swine fever virus* [8]
51. *Foot-and-mouth disease virus* [14]
52. *Goat pox virus*
53. *Lumpy skin disease virus*
54. *Mycoplasma capricolum*
55. *Mycoplasma mycoides*
56. *Newcastle disease virus* [26]
57. *Peste des petits ruminants virus*
58. *Rinderpest virus* [13]
59. *Sheep pox virus*
60. *Swine vesicular disease virus* [5]

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### USDA Plant Protection And Quarantine (PPQ) Select Agents and Toxins

61. *Colletotrichum gloeosporioides* (formerly *Phoma* and *Fusarium oxysporum*; *Fusarium oxysporum*
62. *Peromyscus nuttalli* 
63. *Rasson* *olivacearum* [11]
64. *Rhabdovirus* 
66. *Synchytrium endobioticum*
67. *Xylella fastidiosa*

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[https://www.selectagents.gov/sat/list.htm](https://www.selectagents.gov/sat/list.htm)
NON-SELECT AGENT ZOONOTIC PATHOGENS

Sheep and goats:
- Orf (contagious ecthyma)

Cows:
- *Mycobacterium bovis*

NHPs:
- Herpesvirus B
- Hepatitis B
- *Mycobacterium tuberculosis*

Birds:
- *Chlamydia psittaci*

Rodents:
- LCMV (lymphocytic choriomeningitis virus)

All species:
- Rabies

And so many more...
BIOSAFETY PROGRAM

A fundamental objective of any biosafety program is the containment of potentially hazardous biological agents and toxins.

- The term “containment” is used to describe safe methods for managing infectious materials in the laboratory environment where they are being handled or maintained.
- The purpose of containment is to reduce or eliminate exposure of laboratory workers, other persons, and the outside environment to potentially hazardous agents.
Primary containment, the protection of personnel and the immediate laboratory environment from exposure to infectious agents, is provided by both good microbiological technique and the use of appropriate safety equipment.

- The BSC is the standard device used to provide containment of hazardous biological agents and toxins when conducting microbiological activities.
- Additional primary containment devices may include:
  - Sealed rotors and centrifuge safety cups which prevent aerosols, droplets, and leakage of hazardous biological agents and toxins that may result during centrifugation.
  - Sealed containers provide containment for transfers between laboratories.
Personal protective equipment (PPE) helps protect the user’s body from injury from a variety of sources (e.g., physical, electrical, heat, noise, chemical) or potential exposure to biological hazards and airborne particulate matter.

- Examples: gloves, coats, gowns, shoe covers, closed-toe laboratory footwear, respirators, face shields, safety glasses, goggles, or ear plugs.

PPE is usually used in combination with other biosafety controls (e.g., BSCs, centrifuge safety cups, and small animal caging systems) that contain the hazardous biological agents and toxins, animals, or materials being handled.

In situations where a BSC cannot be used, **PPE may become the primary barrier between personnel and the hazardous biological agents and toxins.**

- Examples include fieldwork, resource-limited settings, certain animal studies, animal necropsy, and activities relating to operations, maintenance, service, or support of the laboratory facility.
SECONDARY CONTAINMENT

Secondary containment, the protection of the environment external to the laboratory from exposure to infectious materials, is provided by a combination of facility design and operational practices. Therefore, the three elements of containment include laboratory practice and technique, safety equipment, and facility design.

Such design features may include, but are not limited to the following:

- Ventilation strategies to ensure containment of the hazards;
- Effluent decontamination systems; and
- Specialized building/suite/laboratory configurations, including: controlled access zones to support the separation of the laboratory from office and public spaces;
- Anterooms; and
- Airlocks.
# Table 1. Summary of Laboratory Biosafety Levels (BSLs)

<table>
<thead>
<tr>
<th>BSL</th>
<th>Agents</th>
<th>Special Practices*</th>
<th>Primary Barrier and Personal Protective Equipment*</th>
<th>Facilities (Secondary Barriers)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Well-characterized agents not known to consistently cause disease in immunocompetent adult humans and present minimal potential hazard to laboratory personnel and the environment.</td>
<td>Standard microbiological practices</td>
<td>No primary barriers required; protective laboratory clothing; protective face, eyewear, as needed</td>
<td>Laboratory doors; sink for handwashing; laboratory bench; windows fitted with screens; lighting adequate for all activities</td>
</tr>
<tr>
<td>2</td>
<td>Agents associated with human disease and pose moderate hazards to personnel and the environment</td>
<td>Limited access; occupational medical services including medical evaluation, surveillance, and treatment, as appropriate; all procedures that may generate an aerosol or splash conducted in a BSC; decontamination process needed for laboratory equipment</td>
<td>BSCs or other primary containment device used for manipulations of agents that may cause splashes or aerosols; protective laboratory clothing; other PPE, including respiratory protection, as needed</td>
<td>Self-closing doors; sink located near exit; windows sealed or fitted with screens; autoclave available</td>
</tr>
<tr>
<td>3</td>
<td>Indigenous or exotic agents; may cause serious or potentially lethal disease through the inhalation route of exposure</td>
<td>Access limited to those with need to enter; viable material removed from laboratory in primary and secondary containers; opened only in BSL-3 or ABSL-3 laboratories; all procedures with infectious materials performed in a BSC</td>
<td>BSCs for all procedures with viable agents; solid front gowns, scrubs, or coveralls; two pairs of gloves, when appropriate; protective eyewear, respiratory protection, as needed</td>
<td>Physical separation from access corridors; access through two consecutive self-closing doors; hands-free sink near exit; windows are sealed; ducted air ventilation system with negative airflow into laboratory; autoclave available, preferably in laboratory</td>
</tr>
<tr>
<td>4</td>
<td>Dangerous and exotic agents that pose high individual risk of aerosol-transmitted laboratory infections and life-threatening disease that are frequently fatal, for which there are no vaccines or treatments; and related agents with unknown risk of transmission</td>
<td>Clothing change before entry; daily inspections of essential containment and life support systems; all wastes decontaminated prior to removal from laboratory; shower on exit</td>
<td>BSCs for all procedures with viable agents; solid front gowns, scrubs, or coveralls; gloves; full-body, air-supplied, positive-pressure suit</td>
<td>Entry sequence; entry through airlock with airtight doors; walls, floors, ceilings form sealed internal shell; dedicated, non-recirculating ventilation system required; double-door pass-through autoclave required</td>
</tr>
</tbody>
</table>

* Each successive BSL contains the recommendations of the preceding level(s) and the criteria in the cell.

a: Applies to Cabinet Laboratory
b: Applies to Suit Laboratory
Clinical laboratories routinely work with unknown specimens and specimens that have the potential to be infected with multiple pathogens; as such, the occupational risks in a clinical laboratory environment differ from those of a research or teaching laboratory. Most public and animal health clinical laboratories use BSL-2 facility, engineering, and biosafety practices.

Clinical diagnostic laboratory personnel may not know what infectious agent or other hazard(s) exist in the specimen they handle and process.
Final determination on the combination of containment measures required to address the relevant biosafety risk present at a facility should be based on a comprehensive biosafety risk assessment.

<table>
<thead>
<tr>
<th>Probability of the event occurring</th>
<th>Frequent</th>
<th>Likely</th>
<th>Occasional</th>
<th>Seldom</th>
<th>Unlikely</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severity of the outcome</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Catastrophic</td>
<td>Extremely high</td>
<td>Extremely high</td>
<td>High</td>
<td>High</td>
<td>Moderate</td>
</tr>
<tr>
<td>Critical</td>
<td>Extremely high</td>
<td>High</td>
<td>High</td>
<td>Moderate</td>
<td>Low</td>
</tr>
<tr>
<td>Marginal</td>
<td>High</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Low</td>
<td>Low</td>
</tr>
<tr>
<td>Negligible</td>
<td>Moderate</td>
<td>Low</td>
<td>Low</td>
<td>Low</td>
<td>Low</td>
</tr>
</tbody>
</table>

Fox, James G. *Laboratory animal medicine*. Elsevier, 2015. pg 1299
CONDUCTING RISK ASSESSMENTS IN A CLINICAL LABORATORY ENVIRONMENT

The assessment team should determine what hazards may exist and the risks associated with those hazards.

- When the agent hazards are unknown, it may be helpful for clinical laboratories to monitor current disease outbreaks and compile lists of commonly encountered pathogens for a population, region, or specimen type.

To help structure biological risk assessments, clinical laboratories should consider what procedures or activities will be performed, where the work will be performed, who will perform the work, and what undesirable events could occur.

It is also essential to evaluate the potential routes of transmission of the suspected infectious agent (i.e., inhalation of aerosols, ingestion, percutaneous inoculation from sharps or non-intact skin, and direct mucous membrane contact from splashes or droplets).
# The Standard Risk Assessment Formula

Severity x Likelihood = Risk

## Criteria for Probability Classifications (per OADLSS SOP)

<table>
<thead>
<tr>
<th>Probability</th>
<th>Classification</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>FREQUENT</td>
<td>&gt; 50% OF TESTING</td>
<td></td>
</tr>
<tr>
<td>PROBABLE</td>
<td>11 - 50% OF TESTING</td>
<td></td>
</tr>
<tr>
<td>OCCASIONAL</td>
<td>1 - 10% OF TESTING</td>
<td></td>
</tr>
<tr>
<td>REMOTE</td>
<td>&lt; 1% OF TESTING</td>
<td></td>
</tr>
<tr>
<td>IMPROBABLE</td>
<td>STATISTICALLY INSIGNIFICANT</td>
<td></td>
</tr>
</tbody>
</table>

## Criteria for Severity Classification (per OLSS SOP)

<table>
<thead>
<tr>
<th>Severity</th>
<th>Potential for appreciable material cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catastrophic</td>
<td>National or international impact</td>
</tr>
<tr>
<td></td>
<td>Work with risk group 4 pathogens</td>
</tr>
<tr>
<td>Critical</td>
<td>Potential for appreciable material cost</td>
</tr>
<tr>
<td></td>
<td>Scope of impact reaching outside CDC</td>
</tr>
<tr>
<td></td>
<td>Work with risk group 3 pathogens</td>
</tr>
<tr>
<td>Serious</td>
<td>Potential for appreciable material cost</td>
</tr>
<tr>
<td></td>
<td>Scope of impact reaching outside the branch</td>
</tr>
<tr>
<td></td>
<td>Work with risk group 2 pathogens</td>
</tr>
<tr>
<td>Minor</td>
<td>Potential for minor material cost</td>
</tr>
<tr>
<td></td>
<td>Scope of impact limited to the team or branch</td>
</tr>
<tr>
<td></td>
<td>Work with risk group 1 organisms</td>
</tr>
<tr>
<td>Negligible</td>
<td>Potential for minor material cost</td>
</tr>
<tr>
<td></td>
<td>Scope of impact limited to the team or branch</td>
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<tr>
<td></td>
<td>Work with nucleic acid extracts or other inactivated material</td>
</tr>
</tbody>
</table>
## Likelihood/Probability

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</tr>
<tr>
<td><strong>Remote</strong></td>
<td>&lt; 1% of testing</td>
</tr>
<tr>
<td><strong>Improbable</strong></td>
<td>Statistically insignificantly</td>
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## Likelihood/Probability

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<td>Statistically insignificant</td>
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## CONSEQUENCE SEVERITY

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</tr>
<tr>
<td>Work with risk group 4 pathogens</td>
</tr>
<tr>
<td><strong>Critical</strong></td>
</tr>
<tr>
<td>Potential for appreciable material cost</td>
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<tr>
<td>Scope of impact reaching outside CDC</td>
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<td>Work with risk group 3 pathogens</td>
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<tr>
<td><strong>Serious</strong></td>
</tr>
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<td>Potential for appreciable material cost</td>
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<td>Scope of impact reaching outside the branch</td>
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<td><strong>Minor</strong></td>
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WHY DO A RISK ASSESSMENT?

Occupational risk assessment: systematic process of evaluating risks from identified occupational hazards associated with a procedure/process in order to mitigate such risks as well as determining if a risk is acceptable or not.

When to do a risk assessment:
- New procedure
- In response to an incident
- Annually

Diagram:
- Identify
- Review
- Evaluate
- Mitigate
- Trial runs
- Constant Communication and Consultation
Elimination: remove the hazard

Substitution: replace the hazard

Engineering controls: isolate people from the hazard

Administrative controls: change the way people work

PPE: protect the person
Established facility-specific best practices and procedures are essential to support the implementation and sustainability of a successful biosafety program.

Persons working in facilities that handle and store hazardous biological agents and toxins must be able to properly identify all potential hazards and be trained and proficient in necessary safe practices and procedures.

Strict adherence to documented laboratory best practices and procedures is an essential element of a robust biosafety program.

All facilities should develop and implement a biosafety program that identifies the hazards and specifies risk mitigation strategies to eliminate or reduce the likelihood of exposures and unintentional releases of hazardous materials.
HANDLING OF TISSUES WITHIN PRIMARY CONTAINMENT

Biosafety cabinet – Class II BSCs are the primary containment devices that protect the worker, product, and environment from exposure to microbiological agents.

Down draft table – Ventilated workstation that pulls air down and away from personnel.

Centrifuge cups
Sealed containers

PPE is still important!!!!
According the BMBL, when a procedure cannot be performed within a primary barrier, a combination of personal protective equipment and other containment devices must be used.

PPE choice depends on risk/possible exposure:
- Gloves (1 vs 2 layers) – Protects skin
- Scrubs/Gown/Disposable coveralls – Protects skin and clothing
- Face shield - Protects against splashes
- Safety glasses/Safety goggles – Protects against splashes
- Respirator vs PAPR – Protects against aerosols
- Dedicated shoes/Shoe covers

Other containment devices:
- Isolated zone
- Sealed containers
APPROPRIATE PPE WHEN PERFORMING NECROPSIES, SURGICAL PROCEDURES, AND DIAGNOSTIC TESTING

Depends on risk assessment:

- Possible exposure to infectious agents?
  - What agent?
    - Select agent
    - Zoonotic agent
  - Severity of disease?
    - No disease
    - Low risk of disease – Therapeutic available
    - Severe disease – Therapeutic available
    - Severe disease – No treatment
  - Route of transmission?
    - Percutaneous
    - Mucous membrane
    - Inhalation
    - Ingestion
    - Environmental
WHAT PPE/CONTAINMENT PRACTICES SHOULD YOU USE?

You receive a placenta from a sheep that recently aborted….

- Possible exposure to infectious agents?
  - What agent?
    - *Coxiella burnetii* (Q fever)
  - Severity of disease?
    - Range from subclinical to severe – Therapeutics available
  - Route of transmission?
    - Inhalation***
    - Others…….

According to BMBL pg 239:

Q fever is the second most commonly reported Laboratory-associated infection (LAI) in Pike’s compilation with outbreaks involving 15 or more persons recorded in several institutions. Infectious aerosols are the most likely route of LAI.

BSL-3 practices and facilities are recommended for activities involving the inoculation, incubation, and harvesting of *C. burnetii*, the necropsy of infected animals, and the manipulation of infected tissues. BSL-2 practices and facilities are recommended for nonpropagative laboratory procedures, including serological examinations and staining of impression smears.

https://www.cdc.gov/vhf/rvf/exposure/index.html
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<tr>
<td>2</td>
<td>Agents associated with human disease and pose moderate hazards to personnel and the environment</td>
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<td>3</td>
<td>Indigenous or exotic agents; may cause serious or potentially lethal disease through the inhalation route of exposure</td>
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DISINFECTION

Always clean and sanitize your workspace and any instruments used with an APPROPRIATE disinfectant for the APPROPRIATE contact time.
TABLE. Chemical compounds used for disinfection, effectiveness of chemical disinfectants and selected products against certain organisms, and selected properties of chemical disinfectants that should be considered when used for cleaning and disinfection

<table>
<thead>
<tr>
<th>Chemical compounds</th>
<th>Chlorine*</th>
<th>Iodine isophor</th>
<th>Chlorhexidine</th>
<th>Alcohol†</th>
<th>Oxidizing agents</th>
<th>Phenol</th>
<th>Quaternary ammonium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Selected products</td>
<td>0.01%–5%</td>
<td>0.5%–5%</td>
<td>0.05%–0.5%</td>
<td>70%</td>
<td>0.2%–3%</td>
<td>0.2%–3%</td>
<td>0.1%–2%</td>
</tr>
<tr>
<td><strong>Bactericidal</strong></td>
<td>Good</td>
<td>Good</td>
<td>Good</td>
<td>Good</td>
<td>Good</td>
<td>Good</td>
<td>Good</td>
</tr>
<tr>
<td><strong>Bacterial spores</strong></td>
<td>Good</td>
<td>Poor</td>
<td>Poor†</td>
<td>Poor†</td>
<td>Fair to good</td>
<td>Poor**</td>
<td>Poor</td>
</tr>
<tr>
<td><strong>Virucidal</strong></td>
<td>Good</td>
<td>Good</td>
<td>Poor</td>
<td>Fair</td>
<td>Good</td>
<td>Poor**</td>
<td>Poor</td>
</tr>
<tr>
<td><strong>Envelope viruses</strong></td>
<td>Yes</td>
<td>Yes</td>
<td>Limited</td>
<td>Yes</td>
<td>Yes</td>
<td>Limited</td>
<td>Limited</td>
</tr>
<tr>
<td><strong>Nonenvelope viruses</strong></td>
<td>Yes</td>
<td>Limited</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td><strong>Fungicidal</strong></td>
<td>Good</td>
<td>Fair</td>
<td>Fair to good</td>
<td>Good</td>
<td>Fair</td>
<td>Fair</td>
<td>Fair</td>
</tr>
<tr>
<td><strong>Protozoal parasite</strong></td>
<td>Fair</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor (ammonia)</td>
</tr>
</tbody>
</table>

**Effectiveness of chemical disinfectants against certain organisms**
- Good = effective; fair = moderate effect; and poor = inferior effect.
- Effectiveness in eliminating envelope and nonenvelope viruses: yes = effective; limited = moderate effect; and no = not effective.
- Effectiveness in organic matter: yes = effective; no = not effective.
- Inactivated by soap: yes = effective; no = not effective.
- Effective in hard water: yes = effective; no = not effective.
- Residual activity: yes = effective; no = not effective.

**Properties of chemical disinfectants**

- Chlorine*: Bleach should be mixed fresh daily and replaced whenever contaminated with organic matter (1:32 dilution of 5.75% solution provides >1,500 ppm chlorine).
- Rubbing alcohol is flammable.
- Alcohol synergistically potentiates the sporidal effect of hypochlorites (chlorine). Mix 5.75% solution of hypochlorite 1:1 with 50% ethyl alcohol/water. Mix fresh at the time of use and provide contact time of >30 minutes.
- The effectiveness of 2-phenylphenol (ortho-phenylphenol) is fair.

**Source:** Adapted from the Nebraska Cooperative Extension and the U.S. Department of Agriculture, 2003.
IN CONCLUSION

Determine exposure risk
Establish SOPs
Use appropriate containment devices and PPE

Report any possible exposures to your immediate supervisor and Responsible Official.
You receive a blood sample from a macaque....

1. Possible exposure to infectious agents?
   A. Yes
   B. No

2. What agent should you be most concerned about?
   A. *Mycobacterium tuberculosis*
   B. *Chlamydia psittaci*
   C. *Francisella tularensis*
   D. Herpesvirus B

3. What is the primary route of transmission?
   A. Percutaneous
   B. Inhalation
   C. Mucous membrane
   D. A and C

4. What biosafety level should the blood sample be handled at?
   A. ABSL 1
   B. ABSL 2
   C. ABSL 3
   D. ABSL 4

https://www.cdc.gov/herpesbvirus/index.html
Exposure via mucous membranes or skin breaks provides this agent access to a new host, whether the virus is being shed from a macaque or human, or is present in or on contaminated cells, tissues, or surfaces. B virus is not generally found in serum or blood, but these products obtained through venipuncture should be handled carefully because contamination of needles via skin can occur.

BSL-3 practices are recommended for handling diagnostic materials with possible B virus. BSL-2 practices and facilities are suitable for all activities involving the use or manipulation of tissues, cells, blood, or serum from macaques with appropriate personal protective equipment. pg 255

All macaques regardless of their origin should be considered potentially infected. Animals with no detectable antibody are not necessarily B virus-free. Macaques should be handled with strict barrier precaution protocols and injuries should be tended immediately according to the recommendations of the B Virus Working Group led by NIH and CDC.

Table 1. Summary of Laboratory Biosafety Levels (BSLs)

<table>
<thead>
<tr>
<th>BSL</th>
<th>Agents</th>
<th>Special Practices*</th>
<th>Primary Barrier and Personal Protective Equipment*</th>
<th>Facilities (Secondary Barriers)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Agents associated with human disease and pose moderate hazards to personnel and the environment</td>
<td>Limited access; occupational medical services including medical evaluation, surveillance, and treatment, as appropriate; all procedures that may generate an aerosol or splash conducted in a BSC; decontamination process needed for laboratory equipment</td>
<td>BSCs or other primary containment device used for manipulations of agents that may cause splashes or aerosols; protective laboratory clothing; other PPE, including respiratory protection, as needed</td>
<td>Self-closing doors; sink located near exit; windows sealed or fitted with screens; autoclave available</td>
</tr>
</tbody>
</table>

Additional information:
https://www.cdc.gov/herpesbvirus/index.html
A rabbit was found dead and submitted for necropsy.

1. Possible exposure to infectious agents?
   A. Yes
   B. No

2. What agent should you be most concerned about?
   A. *Mycobacterium tuberculosis*
   B. *Chlamydia psittaci*
   C. *Francisella tularensis*
   D. Herpesvirus B

3. What is the primary route of transmission?
   A. Ingestion
   B. Inhalation
   C. Percutaneous
   D. All the above

4. What biosafety level should the necropsy be conducted at?
   A. ABSL 1
   B. ABSL 2
   C. ABSL 3
   D. ABSL 4

https://www.cdc.gov/tularemia/index.html
ACCORDING TO THE BMBL 6TH ED.

The agent may be present in lesion exudates, respiratory secretions, CSF, blood or lymph node aspirates from patients, tissues from infected animals, fluids from infected animals, and fluids from infected arthropods. Direct contact of skin or mucous membranes with infectious materials, accidental parenteral inoculation, ingestion, and exposure to aerosols and infectious droplets have resulted in infection.

BSL-3 and ABSL-3 practices, containment equipment, and facilities are recommended for all manipulations of suspect cultures, animal necropsies, and for experimental animal studies. pg 166

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<table>
<thead>
<tr>
<th>BSL</th>
<th>Agents</th>
<th>Special Practices*</th>
<th>Primary Barrier and Personal Protective Equipment*</th>
<th>Facilities (Secondary Barriers)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>Indigenous or exotic agents: may cause serious or potentially lethal disease through the inhalation route of exposure</td>
<td>Access limited to those with need to enter; viable material removed from laboratory in primary and secondary containers; opened only in BSL-3 or ABSL-3 laboratories; all procedures with infectious materials performed in a BSC</td>
<td>BSCs for all procedures with viable agents; solid front gowns, scrubs, or coveralls; two pairs of gloves, when appropriate; protective eyewear, respiratory protection, as needed</td>
<td>Physical separation from access corridors; access through two consecutive self-closing doors; hands-free sink near exit; windows are sealed; ducted air ventilation system with negative airflow into laboratory; autoclave available, preferably in laboratory</td>
</tr>
</tbody>
</table>

Additional information:

https://www.cdc.gov/tularemia/index.html
TRANSPORTATION OF INFECTIOUS MATERIAL

Category A Infectious Substance
UN 2814 (infectious toward humans or both animals and humans)
UN 2900 (infectious towards Animals only)

- Capable of causing permanent disability or life-threatening or fatal disease to otherwise healthy humans or animals.
- Triple packaging system:
  1. Watertight primary receptacle
     - Absorbent material
  2. Watertight secondary packaging
  3. Rigid outer packaging of adequate strength for its capacity, mass, and intended use
- Each surface of the external dimension of the packaging must be 100 mm (3.9in) or more.
- Drop test 1.2m (3.9ft), water-spray test, pressure change of 95 kPa (0.95bar, 14psi), temperatures in the range of -40°C to +55°C.

Figure 1. A Category A UN Standard Triple Packaging
TRANSPORTATION OF INFECTIOUS MATERIAL

Figure 2. A Category B Non-specification Triple Packaging

Biological specimen, Category B UN 3373 (non-infectious)

- A Category B infectious substance does not cause permanent disability or life-threatening or fatal disease in otherwise healthy humans or animals when exposure to it occurs.
- Triple packaging system:
  1. Leak proof primary receptacle
     - Absorbent material
  2. Leak proof secondary packaging
  3. Rigid outer packaging
- At least one surface of the outer packaging must have a minimum dimension of 100mm by 100mm (3.9in).
- Internal pressure differential of 95 kPa.
- Capable of passing a 1.2m (3.9ft) drop test.