## B Virus testing related instructions and useful information

In case of a B Virus exposure the following are ZE LABS recommendations/guidelines for sample submission for herpes B virus testing. It is helpful to get familiar with these recommendations and please refer to this when submitting specimens to ZE LABS.

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| **Day Zero or Base Line Samples** | Human Baseline Serum (2.0 mL – 3.0 mL) collected at the Time of Injury or as soon as possible after injury |
| | Primate Baseline Serum (1.0 mL – 3.0 mL) collected at the Time of Injury or as soon as possible after injury |
| | Human Day Zero swab sample from the buccal cavity and ship the sample in Viral Transport Media tube. |
| | Primate Day Zero swab samples of the buccal cavity, eye or genitalia. Separate swab samples should be collected for each site and sent in separate Viral Transport Media tubes. |
| **Post Seroconversion/ Follow-up Samples (2-3 Weeks Post Exposure)** | Human Follow-up Serum (2.0 mL – 3.0 mL) collected 2-3 weeks after Injury |
| | Primate Follow-up Serum (1.0 mL – 3.0 mL) collected 2-3 weeks after Injury |
| | Human Follow up swab samples of the wound/lesion (if applicable) or buccal cavity collected 2-3 weeks after Injury and sent in separate Viral Transport Media tubes. |
| | Primate Follow up swab samples of the buccal cavity, eye or genitalia, collected for each site 2-3 weeks after incident and sent in separate media tubes. |
| | |
| **ATTENTION:**<br>✓ Please do not use cotton swabs or swabs with wooden or metal shafts as they may contain PCR inhibitors. Only sterile swabs with dacron tips and nylon shafts should be | |

used.

- ✓ If the NHP is not available for follow up testing, a second follow-up serum sample (60 -90 days post exposure) from the exposed individual should be collected and shipped for testing.

## Sample Collection:

### Serum Samples:

- Following standard precautions for collecting biological specimen, collect 5 to 7 ml whole blood in a properly labelled (with patient or animal ID & date) blood collection tube.
- Separate serum following standard serum separation methods.
- Transfer 2.0 – 3.0 ml of serum to a cryovial labelled with patient or animal ID and date of sample collection.
- Either ship immediately following shipping instructions provided in this document or store at appropriate storage temperatures.
- Serum samples can be stored refrigerated (2.0 – 6.0° C) up to one week.
- Serum samples can be stored frozen (≤ -20.0 ° C) for extended periods of time.
- Ship serum sample with appropriate amount of dry ice (5-8 pounds) and use additional dry ice for weekend or holiday shipping. Keep paperwork separate and pack to avoid sample leaks.
- Serum sample can also be shipped with frozen cold packs and use additional frozen cold packs for weekend or holiday shipping.

### Swab Samples:

- Following standard precautions for collecting biological specimen, using a sterile swab, swab each collection site (if applicable) with a new swab and place in an accurately labeled (patient or primate ID, date and swab site) viral transport media sample tube.
- Break the shafts if longer than the tube and seal the tube properly to prevent leaks and possible exposure to B virus.
- Either ship immediately following shipping instructions provided in this document or store at appropriate storage temperatures.
- Swab samples can be stored refrigerated (2.0 – 6.0° C) up to one week.
- Swab samples can be stored frozen (≤ -70.0 ° C) for extended periods of time.
- Ship swab sample with appropriate amount of dry ice (5-8 pounds) and use additional dry

ice for weekend or holiday shipping. Keep paperwork separate and pack to avoid sample leaks.

## Other Sample Types:

## Cerebral Spinal Fluid (CSF):

- Follow standard precautions for collecting biological specimens.
- Accurately label (patient or primate ID, and date) the CSF sample tube.
- Collect 2.0-3.0 mL clean CSF sample using appropriate CSF collection methods. A sample with traces of blood may negatively impact test results. The CSF can be transferred to a sterile CSF storage tube. Do NOT centrifuge CSF.
- Either ship immediately following shipping instructions provided in this document or store at appropriate storage temperatures.
- CSF samples can be stored refrigerated (2.0 – 6.0° C) up to one week.
- CSF samples can be stored frozen (≤ -70.0 ° C) for extended periods of time.
- Ship CSF sample with appropriate amount of dry ice (5-8 pounds) and use additional dry ice for weekend or holiday shipping. Keep paperwork separate and pack to avoid sample leaks.

## Biopsy and Necropsy Tissue:

- Follow standard precautions for collecting biological specimens.
- Accurately label (patient or primate ID, and date) the Viral Transport Media tube.
- Collect an appropriate size tissue sample (for example 4mmx5mm).
- Use separate Viral Transport Media tube for each tissue site and place tissue in respective Viral Transport Media tube.
- Either ship immediately following shipping instructions provided in this document or store at appropriate storage temperatures.
- Tissue samples can be stored refrigerated (2.0 – 6.0° C) up to one week.
- Tissue samples can be stored frozen (≤ -70.0 ° C) for extended periods of time.
- Ship tissue samples with appropriate amount of dry ice (5-8 pounds) and use additional dry ice for weekend or holiday shipping. Keep paperwork separate and pack to avoid sample leaks.

## Cell Lines:

- Follow standard precautions for collecting biological specimens.

- Accurately label (patient or primate ID, and date) the sample tube.
- Collect and pellet down 1-5x$10^7$ cells in an appropriate sample tube, remove the supernatant and freeze the cell pallet.
- Either ship immediately following shipping instructions provided in this document or store at appropriate storage temperatures.
- Cell line samples should be stored frozen (≤ -70.0 ° C) for any periods of time.
- Ship cell line with appropriate amount of dry ice (5-8 pounds) and use additional dry ice for weekend or holiday shipping. Keep paperwork separate and pack to avoid sample leaks.

**Note:** **Only appropriately trained individuals should collect the specimens.**

**Ship the Samples to:** Zoonotic Exposure Laboratory (ZE-LAB)
7540 Louis Pasteur Drive
STE # 200
San Antonio, TX 78229.

## Before shipping samples to ZE-LAB, please ensure:

- Paperwork is complete, accurate, and paperwork matches the specimen being shipped.
- Follow appropriate packaging guidelines, use suitable primary and secondary shipping containers with adequate absorbent material, proper labels on the containers (reference Federal Register 42 CFR Part 72).
- The package has sufficient dry ice or frozen cold packs to maintain appropriate temperature during transit (reference instructions, “Sample Packing”).
- Shipping Address is legible and marked clearly (reference instructions, “Sample Shipping”).
- Package does not contain any glass specimen tubes (reference instructions, “Sample Collection & Handling”). – provided a contact name and phone number in case of emergency (reference instructions, “Sample Packing”).

## Important Considerations:

- For reliable diagnosis, paired specimen (Day Zero/exposure day and Follow-up) testing is required. Therefore, failure to submit a baseline (Day Zero/exposure day) or follow-up (post seroconversion) serum sample will result in incomplete exposure testing, leading to

potentially unreliable diagnosis.

- Specimen should be labeled with name or ID, date of collection, swab collection site. Specimen with incorrect labels may lead to incomplete results. Any unlabeled specimens may not be tested.
- Please do not write PI's name, experiment number, study number or cage number etc on sample tubes. Such info is not required and may interfere with sample identification.

| Test Description | Order Code | Pricing | Methodology | Sample Type |
|---|---|---|---|---|
| Human – B Virus Routine Surveillance | HS01 | $175.00 | ELISA | Serum/ Plasma |
| | HS02 | $200.00 | PCR | Buccal Swab/ Wound Swab/Lesion Swab |
| Human – B Virus Exposure – Baseline | HB01 | $175.00 | ELISA | Serum / Plasma |
| | HB02 | $200.00 | PCR | Buccal Swab/ Eye Swab |
| Human – B Virus Exposure – Follow up | HF01 | $175.00 | ELISA | Serum / Plasma |
| | HF02 | $200.00 | PCR | Buccal Swab/ Lesion Swab/ Eye Swab |
| NHP – B Virus Incident – Baseline | NB01 | $115.00 | ELISA | Serum / Plasma |
| | NB02 | $200.00 | PCR | Buccal Swab/ Wound Swab/Lesion Swab/ Genital Swab |
| NHP – B Virus Incident – Follow up | NF01 | $115.00 | ELISA | Serum / Plasma |
| | NF02 | $200.00 | PCR | Buccal Swab/ Wound Swab/Lesion Swab/ Genital Swab |
| Additional Confirmation – Upon | BWB | $175.00 | Western Blot | Serum / Plasma |

## Sample Shipment:

The samples should be shipped as "Clinical Samples, Biological Substance Category B (UN3373)" in appropriately labeled packages ("Biological Substance – Category B" label, UN 3373 label, and dry ice label -UN 1845) if applicable.

## More info on regulated shipping can be found here:

- Title 42 Code of Federal Regulations, Part 72. Interstate shipment of etiologic agents (Department of Health and Human Services) http://www.cdc.gov/od/ohs/biosfty/shipregs.htm
- Guidelines for the Safe Transport of Infectious Substances and Diagnostic Specimens https://www.who.int/publications/i/item/9789240019720
- The IATA Dangerous Goods Regulations, (International Air Transport Association) http://www.iata.org/ps/publications/dgr/Pages/index.aspx
- Title 49 Code of Federal Regulations, Parts 100-185. Hazardous Materials regulations (Department of Transportation) http://www.phmsa.dot.gov/hazmat/regs
- Title 42 Code of Federal Regulations, Part 72.6. Additional requirements for facilities transferring or receiving select agents (Department of Health and Human Services) http://www.selectagents.gov/
- Biosafety in Microbiological and Biological and Biomedical Laboratories (CDC/NIH) http://www.cdc.gov/od/ohs/biosfty/bmbl/bmbl-1.htm
- International shipments must adhere to CDC, USFWS, and CITES regulations.

## Sample collection related frequently asked questions (FAQ's):

**Q. Why are 2 serum samples (Day Zero and Follow-up) needed from an individual who potentially got exposed to B virus in an incident?**

Paired (two) samples are required because the first sample (**Day Zero**) represents the serum antibody status at the time of potential exposure/injury. Since B virus exposure is not common in humans, antibodies against B virus will be absent in most individuals at the time of incident. The second (**follow-up**) sample is taken after certain time required for antibody generation if exposure happened. The Day Zero sample sets the baseline, and the second sample provides the difference in antibody levels or presence of B virus DNA if exposure occurred. If no difference is observed between the two samples, most likely exposure did not happen.

**Q. Can I collect the swab samples from the exposure/injury associated monkey 1-2 days or more post the incident?**

Swab samples collected right after the incident provide information about the infection status of the monkey at the time of exposure/injury and inform if the monkey was shedding virus at the time of incident. If samples are collected 1-2 or more days later it may impact the accuracy of the results and result in misinformation, as it is possible for the monkey to shed the virus intermittently. It is of paramount importance to determine if viruses are being shed at the time of exposure. We encourage that the swab samples be collected as soon as possible post exposure.

**Q. How to store samples collected on exposure that occurred on a weekend or a holiday, until they are shipped?**

The serum samples can be stored refrigerated (2.0 – 6.0° C) up to one week and stored frozen (≤ -20.0 ° C) for extended periods of time. Similarly, the Swab samples can be stored refrigerated (2.0 – 6.0° C) up to one week and stored frozen (≤ -70.0 ° C) for extended periods of time.

**Q. Does immediate placement of exposed individual on prophylaxis treatment affect the recommendations for sample collection?**

**A.** Yes. In this case a second follow-up serum sample collection is recommended, 14 to 21 days after the start of medication. Thus, in total 3 serum samples are required from individuals who are on treatment. 1– baseline sample (Day Zero); 2– first follow-up (**2-3 Weeks Post Exposure**); 3- second follow-up sample (**2-3 Weeks Post start of treatment)**.