

## Appendix E: Medical Sample Collection for Biological Threat Agents

This guide helps determine which clinical samples to collect from individuals exposed to aerosolized biological threat agents. Proper collection of specimens is dependent on the time-frame following exposure. Sample collection is described for “Early post-exposure”, “Clinical”, and “Convalescent/ Terminal/ Postmortem” time-frames. These time-frames are not rigid and will vary according to the concentration of the agent used, the agent strain, and predisposing health factors of the patient.

- Early post-exposure: when it is known that an individual has been exposed to a bioagent aerosol; aggressively attempt to obtain samples as indicated
- Clinical: samples from those individuals presenting with clinical symptoms
- Convalescent/Terminal/Postmortem: samples taken during convalescence, the terminal stages of infection or toxicosis or postmortem during autopsy

Shipping Samples: Most specimens sent rapidly (less than 24 h) to analytical labs require only blue or wet ice or refrigeration at 2 to 8°C. However, if the time span increases beyond 24 h, contact the USAMRIID “Hot-Line” (1-888-USA-RIID) for other shipping requirements such as shipment on dry-ice or in liquid nitrogen.

Blood samples: Several choices are offered based on availability of the blood collection tubes. Do not send blood in all the tubes listed, but merely choose one. Tiger-top tubes that have been centrifuged are preferred over red-top clot tubes with serum removed from the clot, but the latter will suffice. Blood culture bottles are also preferred over citrated blood for bacterial cultures.

Pathology samples: routinely include liver, lung, spleen, and regional or mesenteric lymph nodes. Additional samples requested are as follows: brain tissue for encephalomyelitis cases (mortality is rare) and the adrenal gland for Ebola (nice to have but not absolutely required).

**Appendix E: Medical Sample Collection for Biological Threat Agents  
Bacteria and Rickettsia**

<b>Early post-exposure</b>	<b>Clinical</b>	<b>Convalescent/ Terminal/Postmortem</b>
<b>Anthrax</b>		
<i>Bacillus anthracis</i>		
<u>0 – 24 h</u> Nasal and throat swabs, induced respiratory secretions for culture, FA, and PCR	<u>24 to 72 h</u> Serum (TT, RT) for toxin assays Blood (E, C, H) for PCR. Blood (BC, C) for culture	<u>3 to 10 days</u> Serum (TT, RT) for toxin assays Blood (BC, C) for culture. Pathology samples
<b>Plague</b>		
<i>Yersinia pestis</i>		
<u>0 – 24 h</u> Nasal swabs, sputum, induced respiratory secretions for culture, FA, and PCR	<u>24 – 72 h</u> Blood (BC, C) and bloody sputum for culture and FA (C), F-1 Antigen assays (TT, RT), PCR (E, C, H)	<u>&gt;6 days</u> Serum (TT, RT) for IgM later for IgG . Pathology samples
<b>Tularemia</b>		
<i>Francisella tularensis</i>		
<u>0 – 24 h</u> Nasal swabs, sputum, induced respiratory secretions for culture, FA and PCR	<u>24 – 72 h</u> Blood (BC, C) for culture Blood (E, C, H) for PCR Sputum for FA & PCR	<u>&gt;6 days</u> Serum (TT, RT) for IgM and later IgG, agglutination titers. Pathology Samples
<hr/> BC: Blood culture bottle C: Citrated blood (3-ml)	<hr/> E: EDTA (3-ml) H: Heparin (3-ml)	<hr/> TT: Tiger-top (5 – 10 ml) RT: Red top if no TT

## Appendix E: Medical Sample Collection for Biological Threat Agents *Bacteria and Rickettsia*

Early post-exposure	Clinical	Convalescent/ Terminal/Postmortem
<b>Glanders</b>		
<i>Burkholderia mallei</i>		
<u>0 – 24 h</u> Nasal swabs, sputum, induced respiratory secretions for culture and PCR.	<u>24 – 72 h</u> Blood (BC, C) for culture Blood (E, C, H) for PCR Sputum & drainage from skin lesions for PCR & culture.	<u>&gt;6 days</u> Blood (BC, C) and tissues for culture. Serum (TT, RT) for immunoassays. Pathology samples.
<b>Brucellosis</b>		
<i>Brucella abortus, suis, &amp; melitensis</i>		
<u>0 – 24 h</u> Nasal swabs, sputum, induced respiratory secretions for culture and PCR.	<u>24 – 72 h</u> Blood (BC, C) for culture. Blood (E, C, H) for PCR.	<u>&gt;6 days</u> Blood (BC, C) and tissues for culture. Serum (TT, RT) for immunoassays. Pathology samples.
<b>Q-Fever</b>		
<i>Coxiella burnetii</i>		
<u>0 – 24 h</u> Nasal swabs, sputum, induced respiratory secretions for culture and PCR.	<u>2 to 5 days</u> Blood (BC, C) for culture in eggs or mouse inoculation Blood (E, C, H) for PCR.	<u>&gt;6 days</u> Blood (BC, C) for culture in eggs or mouse inoculation Pathology samples.
<hr/> BC: Blood culture bottle C: Citrated blood (3-ml)	<hr/> E: EDTA (3-ml) H: Heparin (3-ml)	<hr/> TT: Tiger-top (5 - 10 ml) RT: Red top if no TT

## Appendix E: Medical Sample Collection for Biological Threat Agents *Toxins*

<u>Early post-exposure</u>	<u>Clinical</u>	<u>Convalescent/ Terminal/Postmortem</u>
<b>Botulism</b>		
Botulinum toxin from <i>Clostridium botulinum</i>		
<u>0 – 24 h</u>	<u>24 to 72 h</u>	<u>&gt;6 days</u>
Nasal swabs, induced respiratory secretions for PCR (contaminating bacterial DNA) and toxin assays. Serum (TT, RT) for toxin assays	Nasal swabs, respiratory secretions for PCR (contaminating bacterial DNA) and toxin assays.	Usually no IgM or IgG Pathology samples (liver and spleen for toxin detection)
<b>Ricin Intoxication</b>		
Ricin toxin from Castor beans		
<u>0 – 24 h</u>	<u>36 to 48 h</u>	<u>&gt;6 days</u>
Nasal swabs, induced respiratory secretions for PCR (contaminating castor bean DNA) and toxin assays. Serum (TT) for toxin assays	Serum (TT, RT) for toxin assay Tissues for immunohistological stain in pathology samples.	Serum (TT, RT) for IgM and IgG in survivors
<b>Staph enterotoxigenesis</b>		
<i>Staphylococcus</i> Enterotoxin B		
<u>0 – 3 h</u>	<u>2 - 6 h</u>	<u>&gt;6 days</u>
Nasal swabs, induced respiratory secretions for PCR (contaminating bacterial DNA) and toxin assays. Serum (TT, RT) for toxin assays	Urine for immunoassays Nasal swabs, induced respiratory secretions for PCR (contaminating bacterial DNA) and toxin assays. Serum (TT, RT) for toxin assays	Serum for IgM and IgG
<b>T-2 toxicosis</b>		
<u>0 – 24 h postexposure</u>	<u>1 to 5 days</u>	<u>&gt;6 days postexposure</u>
Nasal & throat swabs, induced respiratory secretions for immunoassays, HPLC/ mass spectrometry (HPLC/MS).	Serum (TT, RT), tissue for toxin detection	Urine for detection of toxin metabolites

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BC: Blood culture bottle  
C: Citrated blood (3-ml)

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E: EDTA (3-ml)  
H: Heparin (3-ml)

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TT: Tiger-top (5 - 10 ml)  
RT: Red top if no TT

## Appendix E: Medical Sample Collection for Biological Threat Agents Viruses

Early post-exposure	Clinical	Convalescent/ Terminal/Postmortem
<p><b>Equine Encephalomyelitis</b> VEE, EEE and WEE viruses <u>0 – 24 h</u> Nasal swabs &amp; induced respiratory secretions for RT-PCR and viral culture</p>	<p><u>24 to 72 h</u> Serum &amp; Throat swabs for culture (TT, RT), RT-PCR (E, C, H, TT, RT) and Antigen ELISA (TT, RT), CSF, Throat swabs up to 5 days</p>	<p><u>&gt;6 days</u> Serum (TT, RT) for IgM Pathology samples plus brain</p>
<p><b>Ebola</b> <u>0 – 24 h</u> Nasal swabs &amp; induced respiratory secretions for RT-PCR and viral culture</p>	<p><u>2 to 5 days</u> Serum (TT, RT) for viral culture</p>	<p><u>&gt;6 days</u> Serum (TT, RT) for viral culture. Pathology samples plus adrenal gland.</p>
<p><b>Pox (Small pox, monkey pox)</b> <i>Orthopoxvirus</i> <u>0 – 24 h</u> Nasal swabs &amp; induced respiratory secretions for PCR and viral culture</p>	<p><u>2 to 5 days</u> Serum (TT, RT) for viral culture</p>	<p><u>&gt;6 days</u> Serum (TT, RT) for viral culture. Drainage from skin lesions/ scrapings for microscopy, EM, viral culture, PCR. Pathology samples</p>
<hr/> <p>BC: Blood culture bottle C: Citratd blood (3-ml)</p>	<hr/> <p>E: EDTA (3-ml) H: Heparin (3-ml)</p>	<hr/> <p>TT: Tiger-top (5 - 10 ml) RT: Red top if no TT</p>