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).

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Bacteria and Rickettsia

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

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

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Toxins

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

BC: Blood culture bottle C: Citrated blood (3-ml)	E: EDTA (3-ml) H: Heparin (3-ml)	TT: Tiger-top (5 - 10 ml) RT: Red top if no TT

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Viruses

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