Please note: This procedure may be revised as needed.

The National Animal Health Laboratory Network (NAHLN) is a partnership of state and federal diagnostic laboratories that provide investigational and surveillance testing for high consequence agricultural pathogens. The 2015 highly pathogenic avian influenza (HPAI) outbreak provided valuable insight into the diagnostic testing capacity of the network. BSL-3 containment is ideal but includes restrictions that create undue burden and must be weighed against the need for rapid, highly efficient processing of samples to identify infected premises and allow depopulation. The 2015 outbreak demonstrated that BSL-2 enhanced space adequately served as the primary safety level for diagnostic testing. Because most of the Biosafety in Microbiological and Biomedical Laboratories (BMBL) is written for human research and diagnostic testing, we recognize a need for guidelines describing testing conditions for outbreaks of agricultural select agents. To this end, the following BSL-2 enhanced guidelines are presented as an adaption of the BMBL 6th Edition https://www.cdc.gov/labs/BMBL.html.

A. General considerations:

- Personnel should receive appropriate training and demonstrate competency in enhanced BSL-2 practices.
- Personnel should be offered seasonal influenza vaccination and should be enrolled in a medical surveillance program.
- Reporting and follow-up procedures should be in place for emergencies and accidents.
- Personnel directly involved in processing and testing of these samples should not be in close contact with susceptible species or associated materials (feed, bedding, etc.) for a minimum of 5 days after their last potential exposure.

B. Sample handling

Sample processing prior to inactivation by lysis represents the greatest risk for generation of aerosol. Procedures should be done in rooms with controlled access. Actual sample transfer and lysis should be done in a biosafety cabinet (BSC). After chemical inactivation, BSL-2 precautions are sufficient.

1. Sample receiving and accessioning

- It is strongly recommended that samples from a suspect premise be submitted to a different receiving area than the normal diagnostic sample stream.
  i. Alternatively, the laboratory must establish procedures to minimize the risk of cross-contamination from samples received from suspect infected premises.
- It is strongly recommended for sample receiving staff to change out of street clothes into scrubs or don a full body suit so that street clothes are completely covered.
- Personnel must wear lab coats (unless donning a body suit), eye protection, double gloves, hair bonnets, and booties.
- Decontaminate all waste daily before it leaves the sample receiving area.
- Decontaminate all reusable personal protective equipment (PPE) (scrubs, lab coats, goggles) daily.
• Decontaminate laboratory equipment and work surfaces routinely, and after spills, splashes, or other potential contamination.

2. Sample transfer and inactivation safety guidance
   • Follow enhanced BSL-2 practices.
   • Personnel must change out of street clothes into scrubs or don a full body suit so that street clothes are completely covered.
   • Wear solid-front or wraparound gowns (over scrubs but not necessary with a body suit), eye protection, double gloves, hair bonnets, and booties.
   • Perform all work with live agents in a certified Class II BSC.
   • Avoid use of sharps (provide sampling material in plastic tubes).
   • Decontaminate all materials before removing them from the BSC.
   • Decontaminate all waste daily before it leaves the facility.
   • Decontaminate all reusable PPE (scrubs, lab coats, goggles) daily.
   • Decontaminate laboratory equipment and work surfaces routinely, and after spills, splashes, or other potential contamination.

3. Removal of inactivated materials to standard BSL-2
   • Once inactivated, the plate with samples should be covered, surface decontaminated and removed from the BSC.
   • Samples may be included in standard BSL-2 workflow from this point forward.
   • All automated extraction instruments, thermocyclers, and liquid handlers can remain in standard BSL-2.

4. BSL-3 activities
   • Agent isolation for detection of live agent from samples identified as non-negative by rRT-PCR must be done in BSL-3 space.