

Inspection Checklist for Biosafety: ACL-3 (9 CFR 121, 42 CFR 73, BMBL 6th Edition, Arthropod Containment Guidelines version 3.2)**Entity Name:****Inspection Date:****Building/Rooms:****Inspectors:**

When information is entered in this form, the form is to be considered "Sensitive Select Agent Information."

Section	Regulation Text	Observation	Status	Comments
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Furniture and incubators containing arthropods are located to prevent accidental contact and release by laboratorians, custodians, and service persons. Arthropods are located in dedicated ACL-3 rooms, wings, or suites, preferably in incubators to serve as an additional layer of containment. Less mobile vector arthropods (e.g., ticks) may be held in vials contained in desiccator cabinets or escape-proof secondary or tertiary housing. Site specific risk assessments address other practices.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	All equipment is appropriately decontaminated and disinfested before transfer between rooms in the insectary. Equipment and supplies not required for ongoing ACL-3 work are removed from the insectary after appropriate decontamination and disinfestation or stored in a designated area and not on open shelves.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	In addition to measures for general arthropod elimination within the insectary, materials used to wipe or mop are autoclaved before disposal. Only persons trained to work with arthropods and BSL-3 agents and equipped with appropriate PPE, clean up spills.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Primary containers are disinfested without creating aerosols. The outer surfaces of containers are decontaminated before moving the material. All primary containers and arthropod waste materials are autoclaved or incinerated for disposal, or as specified by the site-specific risk assessment. Living, infected arthropods are killed by freezing or other appropriate methods, followed by autoclaving or incineration.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Cages used to hold arthropods are non-breakable and screened with mesh of a size to prevent escape. Containers are autoclavable or disposable. Openings are designed to prevent escape during removal and introduction of arthropods.	☐ No ☐ Yes ☐ N/A	

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12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Arthropods are identified with descriptive labels to include the species, strain/origin, date of collection, responsible investigator, and number of arthropods in the containment container and records are maintained to account for all arthropods from the time of transfer to the ACL-3 insectary to the time of termination. Labels are firmly attached to the container and cover, if removable. Vessels containing stages with limited mobility (e.g., eggs, pupae, hibernating adult) are likewise labeled and housed or stored to prevent progression to, and escape of, mobile life stage. All viable potentially infected arthropod life stages must not leave the laboratory.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	PPE is doffed as specified by the risk assessment and IBC protocol. Hands are washed on exit and showering out is based on risk assessment. No material is disposed through the sewer unless appropriately decontaminated (e.g., treatment by 10% hypochlorite, heat, autoclaving, or incineration).	☐ No ☐ Yes ☐ N/A	
12(a)	An individual or entity required to register under this part must develop and implement a written biosafety plan that is commensurate with the risk of the select agent or toxin, given its intended use. The biosafety plan must contain sufficient information and documentation to describe the biosafety and containment procedures for the select agent or toxin, including any animals (including arthropods) or plants intentionally or accidentally exposed to or infected with a select agent. The current biosafety plan must be submitted for initial registration, renewal of registration, or when requested.	Appropriate biosafety and containment procedures have been developed and implemented to assess the effectiveness of the arthropod surveillance program and these are documented. "Count-in, count-out" procedures are recommended and/or additional barriers to escape implemented, as provided by local risk assessment.	☐ No ☐ Yes ☐ N/A	
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12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Harborage and breeding areas are eliminated. Furniture and racks are minimized and can be easily moved to permit cleaning and location of escaped arthropods. Equipment in which water is stored or might accumulate (e.g., humidifiers) is screened to prevent arthropod access, or contains chemicals to prevent arthropod survival.	☐ No ☐ Yes ☐ N/A	
12(a)	An individual or entity required to register under this part must develop and implement a written biosafety plan that is commensurate with the risk of the select agent or toxin, given its intended use. The biosafety plan must contain sufficient information and documentation to describe the biosafety and containment procedures for the select agent or toxin, including any animals (including arthropods) or plants intentionally or accidentally exposed to or infected with a select agent. The current biosafety plan must be submitted for initial registration, renewal of registration, or when requested.	Persons entering the area are made aware of the presence of BSL-3 agents in arthropod vectors. If infected material is present, a biohazard sign is posted on the entrance to the insectary, listing all species handled within and is updated whenever new species are introduced or pathogenic infectious agents are present. The hazard warning sign identifies the arthropod species, agent(s) known or suspected to be present, the name and telephone number of the responsible person(s), and indicates any requirements for entering the insectary (e.g., immunizations, PPE, or respirators).	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	All work is done within a primary barrier. Appropriate biological safety cabinets, other physical containment devices, and/or personal protective equipment are used when conducting procedures to infect arthropods with BSL-3 agents, or when handling arthropods. Manipulation of arthropods and rearing of transovarially infected immature stages are performed in a designated area.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	All infectious and potentially infectious samples are collected, labeled, transported, and processed in a manner that contains and prevents transmission of the agent(s). Transfer of arthropods between manipulation and holding areas is in non-breakable secure containers.	☐ No ☐ Yes ☐ N/A	

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12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Other animals are not accessible to the arthropods. Animals used as hosts or blood sources generally are not housed with arthropods. If present, they are adequately protected from access by escaped arthropods.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	White laboratory coats, gowns, or jumpsuits are worn at all times by personnel entering the insectary. Wraparound or solid-front gowns are worn over this clothing and front button coats alone are unsuitable. Gowns are removed and left in the insectary. Laboratory PPE (e.g., scrubs) are based on local risk assessment, are removed and appropriately contained, and decontaminated before laundering or disposal.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Access to the facility is limited to trained, approved personnel through a self-closing and self-locking door. The external insectary entry doors are controlled by a key lock, card key, or proximity reader. Entry into the insectary is via a double-door entry that includes a change room and shower(s). An appropriately plumbed shower is available in the insectary suite. Shower is plumbed to prevent arthropod escape. An additional double-door access (air lock) or double-door autoclave may be provided for movement of supplies and wastes into and out of the facility, respectively. The two contiguous doors must never be opened simultaneously. Internal doors may open outward or be sliding, but are self-closing, and are kept closed when arthropods are present.	☐ No ☐ Yes ☐ N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	The insectary is designed, constructed, and maintained to facilitate cleaning and housekeeping. The interior walls are light colored so a loose arthropod can be easily located, recaptured, or killed. Light-colored floors are smooth and uncovered. Ceilings are as low as possible to simplify detection and capture of flying insects. Spaces around doors are sealed to facilitate decontamination. Any windows present are resistant to breakage (e.g., double paned or wire reinforced) and sealed. Penetrations of walls, floors, and ceilings are sealed/caulked.	☐ No ☐ Yes ☐ N/A	

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12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	If a central vacuum system is installed, each service outlet is fitted with suitable barriers/filters to prevent arthropod escape. Filters are installed to permit decontamination and servicing. Other vacuum devices are appropriately filtered to prevent transfer and exhausting of arthropods.	o No o Yes o N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	If floor drains are present, traps must be filled with an appropriate treatment to prevent survival of any arthropod stage (e.g., mosquito larvae). Ideally, all drains are plumbed to a holding tank with heat or chemical treatment to kill all stages of arthropods before disposal into the waste system.	o No o Yes o N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Ventilation is appropriate for arthropod maintenance, but does not compromise containment. Exhaust air is discharged to the outside without being re-circulated to other rooms. Exhaust must be dispersed away from occupied areas and air intakes, or the exhaust must be high-efficiency particulate air (HEPA)-filtered. Supply vents protected by HEPA filtration are required if working with BSL-3 veterinary agents. Appropriate filter/barriers are installed to prevent escape of arthropods. The direction of airflow in the insectary is inward. A progressively negative pressure gradient is maintained as distance from the main entrance increases. A visual monitoring device/meter is used to confirm directional inward airflow. Audible alarms alert personnel to system failure.	o No o Yes o N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	Class II biological safety cabinets are HEPA-fitted and certified annually. If Class III cabinets are used, they must be installed appropriately and certified annually. Static glove boxes for arthropod manipulation (e.g., Plexiglass) do not require negative pressure, although accommodation for interior air pressure may be needed based on entity risk assessment (e.g., 0.22 micron filter vent).	o No o Yes o N/A	
12(b)	The biosafety and containment procedures must be sufficient to contain the select agent or toxin (e.g., physical structure and features of the entity, and operational and procedural safeguards).	The insectary must be tested for verification that the design and operational parameters have been met before operation. ACL-3 insectaries are reverified by the PI and by the institutional biosafety representatives where applicable at least annually against these procedures.	o No o Yes o N/A	