

Inspection Checklist for Arthropod Containment Level 3 (ACL-3)

Principal Investigator: _____
Building & Room Number: _____
Phone Number: _____
Department: _____
Inspection Date: _____

		Yes	No	N/A	Comments
Arthropod Containment Specialized Rooms					
Location	Furniture and incubators containing arthropods are located in such a way that accidental contact and release by laboratorians, custodians, and service persons does not occur. This is usually achieved by locating arthropods in dedicated rooms, wings or suites in incubators located out of the traffic flow in areas of the building dedicated to BSL-3 activities.				
Supply Storage	Equipment and supplies not absolutely required for ongoing ACL-3 work are removed from the insectary after appropriate decontamination. Those present are located in a designated area and not on open shelves. It is recommended that a closed storage room, cabinets with tight-fitting doors or drawers be used. Doors and drawers are open only during access.				
General Arthropod Elimination	In addition to measures for general arthropod elimination within the insectary, materials used to wipe or mop are autoclaved before disposal. Only persons trained and equipped to work with arthropods and BSL-3 agents clean up spills.				
Primary Container Cleaning and Disinfestation	Care is taken to disinfest primary containers in a manner that does not create aerosols. All primary containers are autoclaved or incinerated.				

		Yes	No	N/A	Comments
Primary Container Construction	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. Disposable containers are recommended.				
Disposal of Arthropods	Living arthropods are not to be disposed of. All wastes from the insectary (including arthropod carcasses, and rearing medium) are transported from the insectary in leak-proof, sealed containers for appropriate disposal in compliance with applicable institutional or local requirements. All stages of arthropods are killed before disposal. Autoclaving or incineration of material infected with a non-pathogen is recommended. Material may be killed with hot water or freezing before flushing down drains. Autoclaving or incineration of arthropod materials is recommended. The outer surfaces of containers are decontaminated before moving the material. All arthropod waste materials are autoclaved or incinerated.				
Isolation of Uninfected Arthropods	Where possible, only arthropods requiring ACL-3 procedures are housed in the ACL-3 insectary. If it is necessary to house ACL-2 or lower arthropods in the ACL-3 insectary, all procedures and practices must meet the ACL-3 standards.				
Primary Container Identification and Labeling	Arthropods are identified adequately. Labels giving species, strain/origin, date of collection, responsible investigator, and so on are firmly attached to the container (and cover if removable). Vessels containing stages with limited mobility (e.g. eggs, pupae, hibernating adults) are securely stored.				
Prevention of Accidental Dispersal on Persons or via Sewer	Before leaving the insectary and after handling cultures and infected arthropods, personnel wash their hands, taking care not to disperse viable life stages into the drainage system. No material is disposed of through the sewer. Non-infected material may be destroyed by heat or freezing if followed by autoclaving or incineration.				
Pest Exclusion Program	A program to prevent the entrance of wild arthropods (e.g. houseflies, cockroaches, spiders) and rodents effectively precludes predation, contamination, and possible inadvertent infection.				

		Yes	No	N/A	Comments
Escaped Arthropod Monitoring	Additional measures are taken to measure the effectiveness of the arthropod trapping program and these are documented. As part of the IBC review and commissioning process of a new facility, the physical integrity and security practices might be tested by a simple release-recapture study. A known number of non-infected arthropods would be released and then these would be recaptured to assess the physical integrity of security barriers. Exterior and within-building monitoring is considered. Records of exterior captures are maintained.				
Source and Harborage Reduction	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 chemicals to prevent arthropod survival.				
Microbiological and Medical Sharps	Sharps are stringently limited and use is justified only when alternatives are not available.				
Arthropod Sharps	In addition to minimizing arthropod handling sharps, these are restricted for use in the insectary regardless of infection status of the material handled.				
Routine Decontamination	Equipment and work surfaces in the insectary are routinely decontaminated with an effective chemical or by radiation (e.g. heat) after actual or potential contact with an infectious agent, and especially after overt spills and splashes of viable materials (including soil or water that might contain infectious agents or eggs.)				
Notification and Signage	Persons entering the area are aware of the presence of arthropod vectors. If infected material is present, a BSL-3 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, lists the name and telephone number of the responsible person(s) and indicates any special requirements for entering the laboratory (e.g. the need for immunizations or respirators).				
Procedure Design	All procedures are carefully performed to prevent arthropod escape, and the creation of aerosols or splatters. Protocols are practiced with non-infected arthropods / animals and modified before implementation.				

		Yes	No	N/A	Comments
Safety Manual	A safety manual is prepared, approved by the IBC, and adopted. The manual contains emergency procedures, standard operating procedures, waste disposal and other information necessary to inform personnel of the methods for safe maintenance and operation of the insectary.				
Training	The training required for laboratory personnel under ACL-3 is more detailed and extensive, and BSL-3 certification is required if infected materials are handled.				
Medical Surveillance	An appropriate medical surveillance program is in place. All personnel receive appropriate immunizations or tests for the agents handled or likely to be present. When appropriate, a serum surveillance system is implemented (see BMBL for guidance). Personnel are aware of the symptoms of infection and the procedure to follow in reporting these. In general, persons who may be at increased risk of acquiring infection, or for whom infection may be unusually hazardous (e.g. immunocompromised), are not allowed in the insectary unless special personal protection procedures are in place to eliminate extra risk. An assessment is made by the occupational health physician for persons who may be at unusual risk.				
Access Restrictions	The insectary director limits access to the insectary to the fewest number of persons possible. Personnel who must enter the insectary for program or service purposes when work is in progress are accompanied by trained laboratorians and are advised of the potential hazards to themselves, co-workers, and the potential consequences of arthropod release. Because of the increased risk to non-trained personnel, laboratory staff should perform general cleaning activities that would otherwise be performed by custodial staff.				
Special Arthropod Handling Containers and Areas	All work is done within a primary barrier. Appropriate biological safety cabinets, other physical containment devices, and/or personal protective equipment are used whenever conducting procedures to infect arthropods with BSL3 agents, or when handling arthropods. Appropriate designs will consider the life history and behavior of the arthropod and may differ from that required by the agent alone. Such modifications should be made in consultation with biosafety experts. Manipulation of arthropods and, for example, rearing of transovarially infected immature stages, are performed in a designated area. SALS (5) suggests a separate room or double screened area that is separated from the main insectary by rooms having two screened or solid door that open inward and close automatically.				

		Yes	No	N/A	Comments
Safe Transport in the Laboratory	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.				
IACUC and IBC Approval	IBC approval is required and IACUC if vertebrates are used as hosts				
Housing of Non-Arthropod Animals	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, and protocols are approved by the IBC and IACUC.				
Containment During Blood-Feeding	Arthropods fed on host animals are prevented from accidental transfer to host cages. When handling/removing animals after exposure to arthropods, precautions must be taken to prevent arthropod escape through screens, covers, and by flying. Host animals are inspected closely (e.g. concealment in fur, ears, crevices), and the primary container is sufficiently robust to prevent escape during feeding. ACL-3 containment of arthropods during blood-feeding are strictly assured by special practices and container design.				
Blood Source	The blood source is considered as a source of inadvertent arthropod infection and transmission. Measures are implemented to prevent such an event. Use of sterile blood or blood from sources known to be pathogen-free is recommended. In contrast, use of blood from animals or humans whose disease status is uncertain is to be avoided.				
Escaped Arthropod Handling	Loose arthropods must be killed and disposed of, or recaptured and returned to the container from which they escaped. Infected arthropods are not killed with hands, and must be transferred using filtered mechanical or vacuum aspirators. Only personnel properly trained and equipped to work with designated arthropods and BSL-3 infectious agents are to recover and/or kill escaped arthropods.				
Accidental Release reporting	A release procedure is developed and posted. This includes contacts and immediate mitigating actions. Accidents that result in release of infected arthropods from primary containment vessels, or that result in overt exposure to infectious material must be reported immediately to the insectary director who is responsible for ensuring that appropriate and documented action is taken to mitigate the release. Location, number, and type of material are prominently posted until the source is eliminated. Follow-up medical evaluation, surveillance, and treatment are provided as appropriate, and written records are maintained.				

		Yes	No	N/A	Comments
Movement of Equipment	All equipment must be appropriately decontaminated and disinfested before transfer between rooms within the insectary, and before removal from the insectary.				
Inventory of Arthropods	In addition to appropriate primary containment cages, when possible, the number of arthropods must be included on the label, and records maintained to account for all arthropods from the time of transfer to the ACL-3 insectary to the time of termination. Vessels containing low mobility stages (e.g., eggs, pupae, hibernating adults) should not be stored within the ACL-3 insectary unless they meet the ACL-3 criteria.				
Eye and Face Protection	Appropriate face/eye and respiratory protection are worn by all personnel entering the insectary.				
Gloves	Personnel wear gloves when handling infected arthropods, or host animals and associated equipment. Gloves are removed aseptically.				
Torso Apparel	White laboratory coats, gowns, and/or uniforms in the insectary are worn at all times. Wrap-around or solid-front gowns are worn over this clothing. Front-button laboratory coats alone are unsuitable. The gowns are removed and left in the insectary. Before leaving the insectary, scrub suits and uniforms are removed and appropriately contained and decontaminated before laundering or disposal.				
Foot Apparel	Boot, shoe covers, or other protective footwear, and disinfectant foot baths (with appropriate anti-arthropod measures) are available and used where indicated.				
Personal Clothing	Clothing should minimize the area of exposed skin (e.g. skirts, shorts, open-toed shoes, sandals, tee shirts are inadvisable), since this can increase the risk of attracting and being bitten by a loose arthropod.				
Arthropod-Specific Personal Protective Equipment	Personal protective equipment is worn as appropriate e.g. respirators for arthropod-associated allergies, particle masks, head covers. Personal protective equipment is used for all activities involving manipulations of infected or potentially infected arthropods.				
Pesticide	Pesticide for emergency use is available in areas in which escape of arthropods is likely.				

		Yes	No	N/A	Comments
Location of Insectary	The insectary is strictly separated from areas that are open to unauthorized, untrained personnel within the building by locked door				
Insectary Doors	Access to the facility is limited to trained, approved personnel by a self-closing and self-locking door. The external insectary doors are controlled by a key lock, card key, or proximity reader. Entry into the insectary is via double-door entry that includes a change room and shower(s). Showers are 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 outwards or be sliding, but are self-closing, and are kept closed when arthropods are present. Additional barriers (e.g. hanging curtains) are recommended.				
Insectary Windows	Windows are not recommended. Any windows present are resistant to breakage (e.g., double-paned or wire enforced) and well-sealed. If present, fixed light windows are recommended.				
Vacuum Systems	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.				
Interior Surfaces	The insectary is designed, constructed, and maintained to facilitate cleaning and housekeeping. The interior walls are light-colored so that a loose arthropod can be easily located, recaptured, or killed. Gloss finishes, ideally resistant to chemical disinfectants and fumigants, are recommended. Floors are light colored, smooth and uncovered. Ceilings are low as possible to simplify detection and capture of flying insects. Spaces around doors are sealed to facilitate decontamination or troughs surrounding door frames can be installed and filled with sticky or greasy material that will trap crawling arthropods.				
Floor Drains	Floor drains are not recommended. If 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 to facilitate heat or chemical treatment to kill all stages of arthropod prior to disposal into the waste system.				

		Yes	No	N/A	Comments
Plumbing and Electrical Fixtures	Internal facility appurtenances (e.g., light fixtures, pipes and ducting) are minimal since these provide hiding places for loose arthropods. Penetrations of walls, floors, and ceilings are minimal and sealed/caulked. Ideally, light fixtures are flush with the ceiling, sealed, and accessed from above.				
HVAC	Ventilation is appropriate for arthropod maintenance, but does not compromise containment. Exhaust air is discharged to the outside without being recirculated to other rooms. Exhaust must be dispersed away from occupied areas and air intakes, or the exhaust must be HEPA-filtered. 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. Personnel must verify that the direction of the airflow is proper (a visual monitoring device/meter is recommended to confirm directional airflow). Audible alarms alert personnel to system failure.				
Sterilization Equipment	An autoclave is available within the suite of rooms containing arthropods.				
Sink and Shower	The facility has a hand-washing sink with hot water and with suitable plumbing to prevent arthropod escape. An appropriately plumbed shower is available within the insectary suite.				
Illumination	Illumination is appropriate for arthropod maintenance but does not compromise arthropod containment, impede vision, or adversely influence the safety of procedures within the insectary. Lighted (or dark) openings that attract escaped arthropods are avoided.				
Facility Compliance Monitoring	The completed ACL-3 insectary design and operational procedures must be documented by the PI and reviewed by the IBC. The insectary must be tested for verification that the design and operational parameters have been met prior to operation. ACL-3 insectaries are re-verified at least annually against these procedures as modified by operational experience.				

Inspector summary and comments:

Lead inspector signature: _____

Date: _____