

Biosafety Level 2 – Animals (BL2-N)
***NIH Guidelines for Research Involving Recombinant or
Synthetic Nucleic Acid Molecules
(Appendix M-II-B) – April 2024***



Biosafety Level 2 - Animals (BL2-N)

If recombinant or synthetic nucleic acid is derived from a Class 2 organism requiring BL2 containment, personnel shall be required to have specific training in handling pathogenic agents and directed by knowledgeable scientists.

Appendix M-II-B-1. Standard Practices (BL2-N)

a. Animal Facility Access (BL2-N)

- 1) The containment area shall be locked.
- 2) The containment area shall be patrolled or monitored at frequent intervals.
- 3) The containment building shall be controlled and have a locking access.
- 4) The Animal Facility Director shall establish policies and procedures whereby only persons who have been advised of the potential hazard and who meet any specific entry requirements (e.g., vaccination) may enter the laboratory or animal rooms.
- 5) Animals of the same or different species, which are not involved in the work being performed, shall not be permitted in the animal area.

b. Decontamination and Inactivation (BL2-N)

- 1) Contaminated materials that are decontaminated at a site away from the laboratory shall be placed in a closed durable leak-proof container prior to removal from the laboratory.
- 2) Needles and syringes shall be promptly placed in a puncture-resistant container and decontaminated, preferably by autoclaving, before discard or reuse.

c. Signs (BL2-N)

- 1) When the animal research requires special provisions for entry (e.g., vaccination), a warning sign incorporating the universal biosafety symbol shall be posted on all access doors to the animal work area. The sign shall indicate: (i) the agent, (ii) the animal species, (iii) the name and telephone number of the Animal Facility Director or other responsible individual, and (iv) any special requirements for entering the laboratory.

d. Protective Clothing (BL2-N)

- 1) Laboratory coats, gowns, smocks, or uniforms shall be worn while in the animal

area or attached laboratory. Before entering non-laboratory areas (e.g., cafeteria, library, administrative offices), protective clothing shall be removed and kept in the work entrance area.

- 2) Special care shall be taken to avoid skin contamination with microorganisms containing recombinant or synthetic nucleic acid molecules. Impervious and/or protective gloves shall be worn when handling experimental animals and when skin contact with an infectious agent is unavoidable.

e. Records (BL2-N)

- 1) Any incident involving spills and accidents that result in environmental release or exposures of animals or laboratory workers to organisms containing recombinant or synthetic nucleic acid molecules shall be reported immediately to the Animal Facility Director, Institutional Biosafety Committee, NIH OSP, and other appropriate authorities (if applicable). Reports to the NIH OSP shall be sent to the Office of Science Policy, National Institutes of Health, preferably by e-mail to: NIHGuidelines@od.nih.gov (Contact information: 6705 Rockledge Drive, Suite 750, MSC 7985, Bethesda, MD 20892-7985 (20817 for non-USPS mail), 301-496-9838, 301-496-9839 (fax)). Contact information is also available on the OSP website (www.osp.od.nih.gov). Medical evaluation, surveillance, and treatment shall be provided as appropriate and written records maintained. If necessary, the area shall be appropriately decontaminated.
- 2) When appropriate and giving consideration to the agent handled, baseline serum samples shall be collected and stored for animal care and other at-risk personnel. Additional serum specimens may be collected periodically depending on the agent handled and the function of the animal facility.

f. Transfer of Materials (BL2-N)

- 1) Biological materials removed from the animal containment area in a viable or intact state shall be transferred to a non-breakable sealed primary container and then enclosed in a non-breakable sealed secondary container. All containers, primary and secondary, shall be disinfected before removal from the animal facility. Advance approval for transfer of material shall be obtained from the Animal Facility Director. Packages containing viable agents may only be opened in a facility having an equivalent or higher level of physical containment unless the agent is biologically inactivated or incapable of reproduction.

g. Other (BL2-N)

- 1) All genetically engineered neonates shall be permanently marked within 72 hours after birth, if their size permits. If their size does not permit marking, their containers should be marked. In addition, transgenic animals should contain distinct and biochemically assayable DNA sequences that allow identification of transgenic animals from among non-transgenic animals.
- 2) Needles and syringes shall be used only for parenteral injection and aspiration of fluids from laboratory animals and diaphragm bottles. Only needle-locking syringes or disposable syringe- needle units (i.e., needle is integral to the syringe) shall be used for the injection or aspiration of fluids containing organisms that contain recombinant or synthetic nucleic acid molecules. Extreme caution shall be used

when handling needles and syringes to avoid autoinoculation and the generation of aerosols during use and disposal. Following use, needles shall not be bent, sheared, replaced in the needle sheath or guard, or removed from the syringe. Needles and syringes shall be promptly placed in a puncture-resistant container and decontaminated, preferably by autoclaving, before discard or reuse.

- 3) Appropriate steps should be taken to prevent horizontal transmission or exposure of laboratory personnel. If the agent used as a vector is known to be transmitted by a particular route (e.g., arthropods), special attention should be given to preventing spread by that route. In the absence of specific knowledge of a particular route of transmission, all potential means of horizontal transmission (e.g., arthropods, contaminated bedding, or animal waste, etc.) should be prevented.
- 4) Eating, drinking, smoking, and applying cosmetics shall not be permitted in the work area.
- 5) Individuals who handle materials and animals containing recombinant or synthetic nucleic acid molecules shall be required to wash their hands before exiting the containment area.
- 6) A double barrier shall be provided to separate male and female animals unless reproductive studies are part of the experiment or other measures are taken to avoid reproductive transmission. Reproductive incapacitation may be used.
- 7) The containment area shall be in accordance with state and Federal laws and animal care requirements.
- 8) A biosafety manual shall be prepared or adopted. Personnel shall be advised of special hazards and required to read and follow instructions on practices and procedures.

Appendix M-II-B-2. Animal Facilities (BL2-N)

- a. Animals shall be contained within an enclosed structure (animal room or equivalent) to minimize the possibility of theft or unintentional release and to avoid arthropod access. The special provision to avoid the entry or escape of arthropods from the animal areas may be waived if the agent in use is not known to be transmitted by arthropods.
- b. Surfaces shall be impervious to water and resistant to acids, alkalis, organic solvents, and moderate heat.
- c. The animal containment area shall be designed so that it can be easily cleaned.
- d. Windows that open shall be fitted with fly screens.
- e. An autoclave shall be available for decontamination of laboratory wastes.
- f. If arthropods are used in the experiment or the agent under study can be transmitted by an arthropod, interior work areas shall be appropriately screened (52 mesh). All perimeter joints and openings shall be sealed and additional arthropod control mechanisms used to minimize arthropod entry and propagation, including appropriate screening of access doors or the equivalent.