BSL-3 Laboratories

Architectural and Mechanical Design Considerations

The potential threat of bio-terrorism and concern over the possible spread of other naturally occurring infectious diseases, such as SARS, has initiated government funding programs for biological laboratories and research programs associated with infectious diseases protection. As a result, many new biological laboratories are being planned and will be constructed in the coming years. To qualify for government funding, biological laboratories must be designed, constructed, and operated in accordance with strict safety requirements.

This report addresses the physical design factors for a Biological Safety Level 3 (BSL-3) laboratory, including ventilation requirements, in accord with a consensus of current safety standards and best design practices for ensuring occupant health and safety.

Classification of Biological Laboratories

Biological laboratories are normally classified according to the hazard posed by the organisms intended to be present in the laboratory. The classes, called Biosafety Levels, are established and described along with the specific safety requirements in the standards and guidelines listed below. Architects and mechanical system designers need to focus on the aspects of biosafety that lie within their scope of responsibility.

The four main classes help the biosafety community communicate requirements and practices. They do not capture the full range of variation among facilities. When the laboratory director (or another responsible individual) establishes the policies for a particular laboratory, it is with reference to a risk assessment conducted specifically for that lab. The result may combine features that cross the boundaries of biosafety levels. Consequently, there is no all-encompassing description of all BSL-3 laboratories. An individual BSL-3 facility may include features typical of other biosafety levels.

Primary Biological Laboratory Safety Standards and Guidelines:

- **Biosafety in Microbiological and Biomedical Laboratories (BMBL)**
  Published by the U.S. Department of Health and Human Services (U.S. Centers for Disease Control)

  Published by U.S. Department of Agriculture

- **The Laboratory Biosafety Guidelines**
  Published by Health Canada

- **Laboratory Biosafety Manual**
  Published by the World Health Organization

Biological laboratory classifications include:

- **Biosafety Level 1 (BSL-1)**—This is the lowest biological laboratory hazard classification and applies to laboratories involving only low risk activities. Biosafety Level 1 activities may also be conducted within a general chemical laboratory.\(^1\) In actuality, few biological laboratories are designed, constructed or intended to be used as a BSL-1 laboratory.

- **Biosafety Level 2 (BSL-2)**—This classification applies to laboratories handling organisms and agents with an elevated risk in comparison to BSL-1. It is the minimum hazard level that a hospital, clinical or public health laboratory is required to meet and currently covers the largest percentage of existing biological laboratories. Although some substances present in category BSL-2 are infectious, work can be done on tables or benches if the potential for airborne transfer of infectious pathogens (germs) is very

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\(^1\) Where biological operations are conducted in a chemical laboratory, the room design and associated building mechanical systems need to address the safety requirements for a chemical laboratory.
low. However, BSL-2 laboratories increasingly utilize biological safety cabinets for greater worker protection.

All room surfaces and fixtures (HVAC grills, lights, wall switches, etc.) in a BSL-2 laboratory room must be able to be thoroughly cleaned and sanitized. Provisions must also be made for sterilization of all room apparatus and the decontamination of all biological waste material.

- **Biosafety Level 3 (BSL-3)**—This classification applies to a containment laboratory working with highly infectious agents that can cause serious or lethal disease, often transmissible by air and can pose a significant health risk to the general population. (This includes agents that might be used in a biological attack.)

- **Biosafety Level 4 (BSL-4)**—This is a maximum containment laboratory for work with highly infectious, exotic and extremely lethal agents. Typically, there is no known antidote for exposure to the biological agents. Standards pose extensive safety requirements for this type of laboratory that often involves the use of full body airtight worker protective suits with a respiration connection. High security must also be maintained to prevent unauthorized access. In actuality, comparatively few laboratories are designed to meet BSL-4 criteria.

- **Biosafety Level 3 Agriculture (BSL-3AG)**—This type of laboratory classification is a designation by the USDA Animal Research Service (ARS) and applies to a laboratory that presents a high risk of infection from agents that normally affect larger animals and plant foodstuffs. Examples include Foot and Mouth Disease, Mad Cow Disease, and Q Fever. BSL-3AG laboratories need to follow most of the design criteria of a BSL-4 laboratory with the main exception being no requirement for worker full body protection suits.

**Architectural Requirements for BSL-3 Laboratories**

The following compilation of physical design requirements for BSL-3 laboratories is taken from the previously listed standards; augmented by good engineering considerations:

1. The most preferable arrangement is where the BSL-3 laboratory and associated support rooms are in an entirely separate building. However, if a BSL-3 laboratory is within a larger building, the actual laboratory rooms and associated support rooms should be separated from the other areas of the building. Separation methods can include locating the BSL-3 laboratory unit or suite in a remote part of the building that is inaccessible to non-authorized personnel. Another possibility is locating it at the blind end of a building corridor.

2. When the laboratory room entry is not from within a laboratory suite, the entry provision should incorporate two self-closing doors. Electrical door access control systems should be supplemented by mechanical locks so that entry can be prevented during abnormal situations such as a gaseous room or biosafety cabinet decontamination. Doors should be of a sufficient width to allow movement of equipment—typically 36 inches wide. However, in some situations wider doors may be required. For safety, all doors should have vision panels and the necessary fire rating. The entry arrangement should also prevent entrance of vermin and insects. An emergency exit from a laboratory or suite may be necessary in case the primary exit becomes unusable. Applicable fire codes affect this requirement.

3. Appropriate signs should be at the laboratory entry as well as other locations, as may be appropriate. Signs should identify the potential hazards and state that only specific personnel are allowed in the area. Additional information should include the name(s) and contact information regarding regular laboratory operations and also what to do and whom to call in emergencies.

4. Laboratory rooms within the BSL-3 unit should be designed with ample working space and sufficient space for the required equipment. The room layout should facilitate cleaning, decontaminating and sanitizing. The temperature and humidity generally needs to be

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2. A BSL-3 biological laboratory typically involves more than a single room. Typically, a suite of several rooms adjacent to the laboratory room itself is necessary to fulfill the health and safety requirements as well as enable the overall laboratory functions to be carried out.

3. A laboratory suite may encompass several laboratory rooms and associated support rooms. Entry into the laboratory suite requires passage through clothes changing areas that are negatively pressurized with respect to the outer corridor and the non-laboratory areas of a building. Once inside the laboratory suite, entry into individual BSL-3 laboratory and support rooms does not then require special double door entry provisions.
controlled as required by the substances present and provide for worker comfort.4

5. Walls, ceilings, floors, and fixtures should be smooth, easy to clean, impermeable to liquids and resistant to chemicals and disinfectants normally used. Ceilings constructed of gypsum board are preferable to conventional lay-in ceiling tiles. A continuous cove floor that curves up at the wall is desirable. Exposed piping and ductwork should be avoided. Floors should be seamless (for example, epoxy coated), slip-resistant and only have drains where needed such as under emergency showers or for hand washing. Drains may not be directly connected to a sanitary sewer and must have backflow prevention and first lead to tanks where liquid waste will undergo decontamination.

6. Penetrations made in walls, floors, and ceilings for installing electrical, plumbing, HVAC, and other services must be properly sealed to facilitate maintaining negative room pressurization, prevent entry of insects or vermin, and allow effective gaseous decontamination of the room. Electrical outlets are preferably surface mounted to reduce wall penetrations.

7. If present, windows should be non-operable and break-resistant.

8. Bench tops that extend to a wall should be sealed at the wall to prevent liquids from entering crevices at the wall. Bench tops and work surfaces should be impervious to liquids and resistant to disinfectants, acids, alkalis, solvents, and moderate heat.

9. Biological safety cabinets and fume hoods5 should not be located near the normal laboratory room entry area or along regular walk routes.

10. Room illumination should be adequate and designed to minimize glare in the glass panels of biological safety cabinets and fume hoods.

11. Open spaces between and under furniture should allow ready access for cleaning.

12. Sufficient storage space for laboratory supplies should be provided so that such items are not stored on bench tops, biosafety cabinets, or in other functional space within the room.

13. Provision for storing outer garments and personal items should be provided in a separate room (locker room/gowning room) outside of the laboratory room, but within the laboratory suite. Ideally, passage through these rooms should be a part of the normal laboratory suite entry procedure. When warranted, shower, and clothes changing provisions should be available.

14. A hand-wash sink that is operated by a foot pedal, elbow lever, or automatic proximity sensor should be provided near each laboratory room exit door. At least one emergency eyewash sink must also be present in each laboratory room.

15. Provision for eating and drinking should be outside of, but convenient to, the laboratory rooms.

16. There should be no cross-connections between sources of laboratory water and the drinkable water supply. Anti-backflow devices must protect the public water system.

17. Stand-by power is desirable for critical laboratory equipment, such as incubators, freezers, certain biological safety cabinets, and for ventilating animal cages and animal holding rooms. Emergency lighting should ensure safe exit from the laboratory rooms and suite in the event of a power failure.

18. Reliable and adequate gas service should be available for laboratory room equipment.

19. Provision for waste disposal should include:
   - Autoclaves6 or incinerators for solid waste treatment
   - Emission controls on incinerators7
   - Wastewater decontamination tanks

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6. An autoclave uses steam to sterilize equipment and destroy living organisms. Autoclaves work by first creating a near vacuum within the interior space and then allowing steam at 15 psi (103 kPa) to enter. The internal autoclave temperature becomes approximately 485°F (120°C) and is then maintained for at least 15 minutes. Pass-Through autoclaves are designed to mount in a barrier wall and thus enable the loading and removal operations to be conducted from separate rooms.

7. Incinerators are generally only necessary when larger masses of biological matter must be disinfected, as is the case when large animals are utilized in laboratories. Since incinerators are subject to very stringent federal and local codes it is best to only provide them if necessary.
20. A facility monitoring and control system should announce and automatically summon an appropriate response to emergencies such as a fire, hazardous spill, injury, and the activation of a room emergency shower or eyewash station. A first-aid area or a room suitably equipped and readily accessible for immediate treatment should be in proximity to the laboratory suite.

21. Laboratory security should be maintained with adequate room access controls and intrusion detection systems. CCTV monitoring of laboratory room interiors is usually prudent.

22. Laboratory rooms should have a telephone or intercom for communication between the laboratory room and outside locations. (Personal cell phones and portable computers should not be brought in and out of the laboratory room since there is no practical way to ensure adequate decontamination of such devices before removing them from the room.) An adequate number of clocks should be in the laboratory suite and individual rooms so that wristwatches are not needed.

23. Means of transferring data from the interior of laboratory rooms (FAX, e-mail, electronic media, etc.) is highly recommended in lieu of physically transporting paperwork, record books, etc.

24. Before occupancy, BSL-3 laboratories must be tested and certified to ensure that all systems function properly and required room pressurization is attained.

**Biological Laboratory Layout**

As stated, biological laboratories are usually comprised of suites of several rooms based upon the laboratory’s purpose and the support services required. There may be no typical BSL-3 laboratory suite. Each one is designed according to specific requirements and constraints. The following discussion uses a fictitious BSL-3 suite to illustrate some desirable design features. This layout is more elaborate than many actual BSL-3 labs.

The suite consists of multiple laboratory rooms along with additional rooms for refrigerated storage, specimen reception, incubation, sterilization, supply storage, and many other purposes.

Before entering the laboratory room or suite, provision must be made for the workers to enter, change clothes, and put on protective outer garments (gown-up). Upon departing, the workers must have provision to remove outer garments (de-gown), shower, and put on their regular clothes. BSL-3 laboratory suites should be equipped with their own washrooms to minimize the necessity for workers to leave the laboratory area. Individual BSL-3 laboratory facility configurations will differ based upon an evaluation of the specific needs of the facility and a risk evaluation.

Figure 1 shows the overall suite layout and Figure 2 shows the normal entry and exit path for laboratory workers. The suite in Figure 1 has separate men’s and women’s entry and exit provisions including locker rooms, gowning, de-gowning rooms, and washrooms. Shower rooms are also present.

For safety reasons, a separate SPECIMEN ENTRY is provided to enable biological agents to be brought into the laboratory apart from the worker entry areas. Specimens must be brought into the laboratory in sealed protective containers and deposited in the SPECIMEN ENTRY area. The specimens are then retrieved from the SPECIMEN ENTRY room by trained and knowledgeable laboratory workers. Specimens are normally placed in a biological safety cabinet before being removed from their protective container.

After biological agents have been subjected to the intended research process and are no longer needed, they are put into another protective container and taken to the AUTOCLAVE ROOM where they are decontaminated. The decontaminated waste is removed from the AUTOCLAVE through a separate outlet in the adjoining room and properly disposed. Laboratory supplies and other non-biological items can be brought to the laboratories via the service corridor. This corridor can also be used as an emergency exit for the laboratory workers in case the NORMAL ENTRY/EXIT is not usable or accessible.

The laboratory suite has the necessary SUPPORT ROOMS. Depending upon the nature of the work being done, multiple support rooms might be needed especially when laboratory animals are used.

Although the room ventilation systems that serve the laboratory suite need not be in a location that is separated from the systems serving non-laboratory areas, a separate MECHANICAL EQUIPMENT ROOM for the laboratory suite is shown. This provides greater operational security and helps to

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8. Shower provisions are not specifically required in all instances. The inclusion of a shower provision is typically based upon a risk assessment of the specific laboratory hazard. Normally showers would be required when the lab work involves larger animals.
facilitate proper preventive maintenance and regular testing.

Figure 2 shows the normal entry and exit (NEE) routes for laboratory workers. Entry consists of passing through the normal entry and exit and proceeding to their respective locker room. Workers then remove their outer garments and secure personnel items that are difficult to decontaminate. This includes watches, purses, wallets, keys, jewelry, contact lenses cell phones, etc. They then proceed to the gowns room and put on the prescribed outer protective garments that would normally include a lab coat and gloves. In some instances, safety glasses, respirators, hair covering, and other items may also be required. They can then enter the inner portion of the laboratory suite that includes the laboratory and support rooms, break room and washrooms.

Figure 1. BSL-3 Example of a BSL-3 Biological Laboratory Suite.
Figure 2. BSL-3 Biological Laboratory–Normal worker Entry and Exit Routes.

Figure 3. BSL-3 Laboratory Workers with Respirators Working at a Biological Safety Cabinet.
To leave the laboratory suite, a worker first enters the washroom and proceeds to the de-gowning room where they remove their outer protective garments and place them into a container for disposal or sanitizing as appropriate. Then, they must disinfect any personal items that were with them inside of a laboratory room (eye-glasses, hearing aids, etc.) Finally, depending upon the individual circumstances and individual laboratory practices, they may need to shower before proceeding to their locker room to obtain their regular outer clothing and other personal items. Exit is then through the normal entry and exit double door arrangement.

**Ventilation Requirements for BSL-3 Laboratories**

Figure 4 shows a diagram of a ventilation system arrangement for a BSL-3 laboratory room.

**Ventilation Airflow**

The room supply air in Figure 4 consists of 100% outside air and the airflow quantity and final discharge temperature is controlled at the ROOM SUPPLY AIR TERMINAL. The air is gently diffused into the room by one or more perforated ceiling diffusers. Room supply air should always be gently dispersed into the room at the entry area to establish airflow from the room entry area towards the higher risk area without creating high velocity air currents. It is important not to have undesirable high velocity cross currents at the biological safety cabinets or fume hoods.

Biosafety cabinet exhaust is connected to the laboratory exhaust system by hard duct connections as required for B1 or B2 Biosafety cabinets. A continual amount of room air is also exhausted from the ceiling as well as near the floor level to remove chemical fumes that are lighter or heavier than the room air.

**Room Ventilation Rate**

BSL-3 room ventilation rates should be 4 to 8 air changes per hour (ACH). The higher rate applies during occupied periods and 4 ACH is acceptable when the room is unoccupied. Animal holding rooms within laboratory suites require a constant 15 ACH.

**Room Pressurization**

The ventilation system must establish a reliable airflow through the suite in the direction of increasing contamination. This means the laboratory room should be maintained at a negative pressure; typically -0.10 Inch WC (-25 Pa) or more below non-laboratory areas of the facility. A greater negative pressure may be utilized to ensure the airflow direction is always into the area of greatest risk especially when various support facilities (incubation rooms, autoclave rooms, etc.) are a part of the laboratory suite. BSL-3 laboratory room air handling system ductwork should be designed to withstand the pressure levels resulting from any anticipated mode of operation or failure. For example, if the supply system fails or is shut down while the exhaust system remains operational, the pressure in the supply and exhaust ducts may reach the full suction capability of the exhaust fans. Supply systems should be designed so that supply dampers fail open if the exhaust systems remain operating to minimize the negative impact on the laboratory.

Figure 5 shows differential pressure relationships between the laboratory suite rooms as well as the area outside of the laboratory suite. Three different negative pressure levels are utilized in this example. The areas of highest risk such as the Laboratory Rooms and the Autoclave Room are the most negative as indicated by three negative signs (- - -). Support rooms and rooms used for normal entry and exit procedures are maintained at a lesser negative pressure level indicated by two negative signs (- - ).

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9. A full and complete body shower is normally required when animals are present due to the increased likelihood of airborne pathogens that could adhere to the exposed skin or hair of the workers.

10. If type A biosafety cabinets are used they are not required to be hard ducted to the exhaust system but may return a portion of their HEPA filtered exhaust into the room. Such biosafety cabinets usually use a thimble connection that only allows the exhaust system to remove a portion of the cabinet exhaust.

11. Some chemical fumes associated with biological laboratories are heavier than air and therefore a floor level room exhaust provision is sometimes recommended.

12. This is the NFPA 45 recommendation. However, specific requirements may call for different ACH rates or not allow reduction of the ACH rate during unoccupied periods.

13. The 0.10 Inch WC value is based on double door entry system with a reliable pressure difference of 0.05 Inches WC across each door. If there are more pressurized boundaries, the lab room pressure is likely to be even more negative.
Double door interiors are maintained at a lower negative pressurization level as indicated by a single negative sign (-). Although not specifically required, it is desirable to use electrically interlocking doors so that only one door may be open at a time. This helps maintain containment even when a door is opened.

**Differential Pressure Monitors**

Differential Pressure Monitors are located at doorways where lab users can confirm proper pressurization. These monitors indicate the static pressure difference and warn of a loss of required differential pressure at that location. While a door is open, the differential pressure level across it drops until the door is again fully closed. Therefore, differential pressure alarms must have an adjustable alarm delay to prevent nuisance alarms during passage through doors.

![Room Ventilation Diagram for BSL-3 Laboratory Room](image)

**Figure 4. Room Ventilation for a BSL-3 Laboratory Room.**
Air Filtration

BSL-3 laboratory supply air is not required to be equipped with HEPA (High Efficiency Particulate Air) filters. However, the USDA requires HEPA filters on supply air serving animal rooms to ensure containment upon system failure.  

BSL-3 laboratory exhaust air is required to be HEPA filtered except when the laboratory will only be manipulating non-airborne transmitted organisms. When filtering is required, it must include the room’s ceiling and/or floor exhausts and any other exhausts from the room such as canopy exhausts and specialty exhausts. Even though Class II Biosafety cabinets are equipped with HEPA exhaust air filters, good laboratory design practice is to have all of the combined exhaust from the entire laboratory room pass through a final HEPA filter before connecting to the facility’s exhaust system.

HEPA filter(s) located in the exhaust system ductwork should be in bag in/bag out housings. The filter housing should be isolated by bubble tight isolation dampers to enable decontamination of the filter before removal. To ensure continued operation of the laboratory while replacing filters, dual filter exhaust housings in parallel are recommended. Filters should be accessible from outside of the laboratory rooms, preferably in the mechanical equipment room or in a service area.

14. A HEPA filter will effectively block airborne bacteria and most viruses, but will not stop prions or chemical fumes.

15. Bubble Tight isolation dampers are normally manually operated and do not require a remote operation provision.

16. Mechanical equipment rooms serving biological laboratories should preferably be located close to or adjacent to the laboratory suite and be negatively pressurized if exhaust filters are accessible from within the room.
corridor. Exhaust air from the laboratory suite, laboratory rooms, fume hoods and biological safety cabinets must not be re-circulated to other rooms or parts of the facility, even when HEPA filtered. As with chemical fume exhaust, the air exhausted from biological laboratories must be discharged outside of the building and dispersed in a manner that does not pose a hazard to persons, other buildings, or allows entrainment into building fresh air intakes.

Isolation Dampers

Biosafety cabinets are periodically decontaminated using formaldehyde or vaporized hydrogen peroxide while the laboratory room is unoccupied. If a spill or release of some biological agent occurs within the laboratory, the room itself may also need to be decontaminated. The BMBL requires that BSL-3 laboratories be designed to accommodate these procedures. Therefore, a bubble tight isolation damper may be located between the room supply air terminal and room supply air diffuser(s). Exhaust air bubble tight isolation dampers should be located after the juncture of all separate room exhaust provisions and the final HEPA filters associated with the room exhaust. If multiple laboratories have their exhausts combined into a single manifold type exhaust system, each individual laboratory room exhaust must be capable of being isolated from the other room exhausts by a bubble tight damper. Access to such dampers, as well as all duct components, should preferably be from outside of the laboratory suite.

Maintenance and Housekeeping

Perhaps one of the most overlooked aspects of safe and efficient biological laboratory operations is that of maintenance. As with any type of facility, maintenance tasks must be routinely performed. This includes servicing laboratory equipment as well as elements of the facility itself such as HVAC components, plumbing fixtures, electrical light fixtures, etc. Those performing such tasks must receive proper training regarding the potential hazards of the laboratory environment along with the proper entry and exit procedures.

Laboratory suites should be designed to ensure that maintenance personnel can only enter and exit by the normal process. This includes clothes changing, gowning, etc. In the case of maintenance workers, additional problems can exist with respect to the need to have tools and other equipment such as ladders, portable lights, etc. If these items are brought into a BSL-3 laboratory or support room, there must be provision to ensure adequate decontamination before being removed from the laboratory suite. Since this may not be easily accomplished, it is advantageous to provide a storage provision within the laboratory suite for such items so that workers do not have to bring in commonly needed tools and equipment every time a maintenance task needs to be done.

Housekeeping tasks are normally performed by laboratory assistant personnel who are aware of the potential hazards and safety procedures necessary to protect against accidental contact with biological agents, aerosols, and especially how to prevent the potential migration of such agents outside of the laboratory. It is recommended that adequate housekeeping storage also be provided within the laboratory suite for items normally needed for housekeeping purposes. Items include cleaning supplies, mops, janitorial sinks, disinfectants, wipes, tissues, etc. Note that Figure 1 and Figure 5 show Maintenance and Janitorial Closets within the laboratory suite for this purpose.

In addition, there must be a procedure for the safe removal and disposal of regularly discarded items from within the laboratory suite, such as waste paper, laboratory wipes, broken glassware, and other refuse.