

Review

Research and Prospects of Airtightness of Biological Laboratory Enclosures: Influencing Factors and Evaluation Methods

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Abstract

The airtightness of enclosures in biological laboratories is paramount for effective isolation between internal and external environments, ensuring containment of hazardous pathogens and mitigating accidental release risks. Consequently, studying and enhancing the airtightness of these enclosures significantly contributes to maintaining laboratory safety, safeguarding personnel health, and preventing disease. This paper examines the critical role of the enclosure structure in biological laboratories and how airtightness impacts the laboratory environment. It analyzes the current state of research on the airtightness of high-level biological laboratory enclosures, drawing on domestic and international sources. This includes exploring issues and challenges related to material properties, process methods, and evaluation methods for the airtightness of concrete structural materials as well as steel structures. The paper also studies the airtightness of enclosure structures of biological laboratories, highlighting the shortcomings in this important field of research and its future prospects.

Keywords: high-level biosafety laboratory; enclosure structure; material properties; airtightness evaluation methods



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1. Introduction

The COVID-19 pandemic, caused by the novel coronavirus SARS-CoV-2, has highlighted the critical role of biosafety laboratories in global public health. These facilities are essential for researching highly pathogenic microorganisms, developing diagnostics, and formulating vaccines. Among various biosafety levels, Biosafety Level 4 (BSL-4, or P4) laboratories represent the highest containment level, designed to handle pathogens that pose a serious threat to human health and the environment. The airtightness of these laboratory enclosures is paramount, as it ensures the isolation of hazardous pathogens from the external environment, thereby preventing accidental releases and safeguarding both personnel and the public [1].

The development of biosafety laboratories originated in the mid-20th century, pioneered by the United States. Over the decades, advancements in laboratory design and construction have been driven by the need to address emerging infectious diseases, such

as SARS, MERS, and Ebola. Globally, there are currently 62 operational or planned BSL-4 laboratories across 24 countries, with Europe and North America leading in both quantity and technological maturity [2,3]. These facilities are predominantly government-owned, reflecting their strategic importance to national and global health security.

In China, the construction of high-level biosafety laboratories has accelerated in recent years, spurred by the “Plan for the Construction of High-Level Biosafety Laboratory System (2016–2025)” issued by the National Development and Reform Commission and the Ministry of Science and Technology. This plan aims to establish 5–7 BSL-4 laboratories and at least one BSL-3 laboratory in each province by 2025 [4]. Despite this progress, challenges remain, including uneven geographical distribution, gaps in management systems, and a shortage of qualified professionals [5].

The airtightness of laboratory enclosures is a critical factor in their design and operation. It involves the integration of material science, structural engineering, and advanced construction techniques to create impermeable barriers. Concrete and stainless steel are the primary materials used, each with distinct properties and challenges. For concrete structures, factors such as mixing ratios, material additives, and construction processes significantly influence airtightness [6,7]. Steel structures, particularly those employing stainless steel seam welding, offer superior airtightness and durability but require meticulous welding techniques and material selection [8].

Despite progress, gaps persist in the research on airtightness evaluation methods and standards. While international standards such as ISO 9972:2015 [9] and ASTM E779-19 [10] provide robust frameworks, China’s national standards, such as GB 19489-2008 and GB 50346-2011, still require refinement to enhance accuracy and applicability [11]. Furthermore, the interactions between material properties, structural design, and long-term operational reliability remain underexplored.

This paper aims to address these gaps through a comprehensive review of the airtightness of biological laboratory enclosures. It examines the current state of global BSL-4 laboratory construction, analyzes the material properties and construction techniques that influence airtightness, and evaluates existing testing methods and standards. By synthesizing domestic and international research, this study seeks to contribute to the development of more reliable and efficient enclosure systems, ultimately enhancing the safety and functionality of high-level biosafety laboratories.

The significance of this research extends beyond the academic realm, offering practical insights for policymakers, engineers, and laboratory operators tasked with designing, constructing, and maintaining these critical facilities. As the world faces increasing biological threats, the lessons learned from this study will be instrumental in fortifying global biosafety infrastructure.

2. Current Status of Research Worldwide and in China

2.1. Development of Biosafety Laboratories Worldwide

The establishment of biosafety laboratories dates back to the 1950s and 1960s in the United States. Subsequently, in the 1980s, China collaborated with international partners to establish its first high-level biosafety laboratory. By the 1990s, China had developed the capability to design and construct such laboratories independently. As of now, there are 62 operational and planned BSL-4 laboratories situated in 24 countries globally. Approximately 60% of these facilities are government-owned, 20% are affiliated with universities, and 20% are military establishments [2]. Table 1 summarizes the global distribution of BSL-4 laboratories, with particular emphasis on regional variations in airtightness techniques and operational challenges. A comprehensive overview of operational and planned BSL-4 laboratories worldwide, including their construction years and areas, is provided in Appendix A

(Table A1). These data reveal significant disparities between developed and developing regions, informing subsequent discussions on material innovations (Sections 3 and 4) and testing standards (Section 6).

Table 1. Global Distribution and Airtightness Practices of BSL-4 Laboratories.

Region	Country	No.	Representative Facilities	Year	Area (m ²)	Airtightness Standards /Techniques	Key Challenges
Europe	UK	7	Public Health England (PHE)	1974	105	ISO 9972:2015, EN 13829:2000; Stainless steel welding, Epoxy coatings	Common: High initial costs (materials and welding); Specific: Aging infrastructure requires specialized retrofitting.
	Germany	4	Bernhard Nocht Institute (BNI)	2014	400	Airtight concrete additives, Dynamic pressure control systems	Common: High operational costs; Specific: Stringent energy efficiency requirements increase the complexity of HVAC systems.
North America	USA	11	NIH Integrated Research Facility (IRF)	2008	1145	ASTM E779-19; Composite materials, HEPA filtration systems	Common: Regulatory compliance burdens; Specific: Large facility size amplifies joint leakage risks.
Asia	China	4	Wuhan Institute of Virology	2016	200	GB 19489-2008; Steel/concrete hybrid structures	Common: Technician training deficiencies; Specific: Thermal expansion mismatch in hybrid structures.
	India	4	Microbial Containment Complex (MCC)	2012	848	ISO 10648-2; Gasket-sealed doors	Common: Funding gaps; Specific: Tropical climate accelerates gasket degradation.
Africa	South Africa	1	NICD Special Pathogens Unit	1979	N/A	Limited data; Reliance on international collaborations	Common: Resource constraints; Specific: Outdated facilities require donor-funded upgrades.
Oceania	Australia	4	Australian Centre for Disease Preparedness	1985	567	Laser welding technology, Smart pressure monitoring	Common: High operational costs; Specific: Geographical isolation increases parts replacement time.

Notes: “N/A” indicates unavailable data Data current as of 2024; some facilities are under construction. Regional totals include both operational and planned facilities.

The comparison of airtightness techniques in Table 1 reveals regional variations in material selection and their consequences. For instance, the steel/concrete hybrid structure adopted by the Wuhan Institute of Virology combines the cost-effectiveness of concrete with localized steel reinforcement at joints, achieving low leakage rates under laser welding. However, this design may face long-term micro-crack formation in concrete under cyclic pressure loads, requiring periodic sealant maintenance. In contrast, PHE’s all-stainless-steel enclosure with epoxy coatings offers superior corrosion resistance and seamless weld integrity, but at 2–3 times the material cost of hybrid designs (Section 5.1). These differences reflect the trade-off between initial investment and long-term maintenance.

In a regional context, Europe boasts the highest concentration of P4 laboratory facilities and is the earliest developed, followed by North America. In contrast, laboratories in other regions are sporadic and have been established at a later stage. Regarding individual countries, the United States has the largest number of P4 laboratories and has demonstrated maturity in constructing and operating high-level laboratories [3,12]. This distribution is illustrated in Figures 1 and 2.

The global disparities in biosafety laboratory development arise from varying technical capabilities, policy frameworks, economic resources, and historical contexts. Developed countries such as the US and Europe have benefited from early technological advantages, robust regulatory systems, and substantial funding, enabling them to establish advanced facilities decades ago. Meanwhile, developing regions face challenges including technological gaps, inconsistent policies, and limited budgets, often relying on international collaborations for capacity building. The high cost of BSL-4 laboratories further limits their deployment in resource-constrained environments. Notably, defense-related research has historically driven biosafety investments in Western countries, while emerging economies like China and India are now prioritizing laboratory networks in response to recent pan-

demics and growing biosecurity needs. These systemic differences highlight the importance of context-specific approaches to global biosafety infrastructure development.

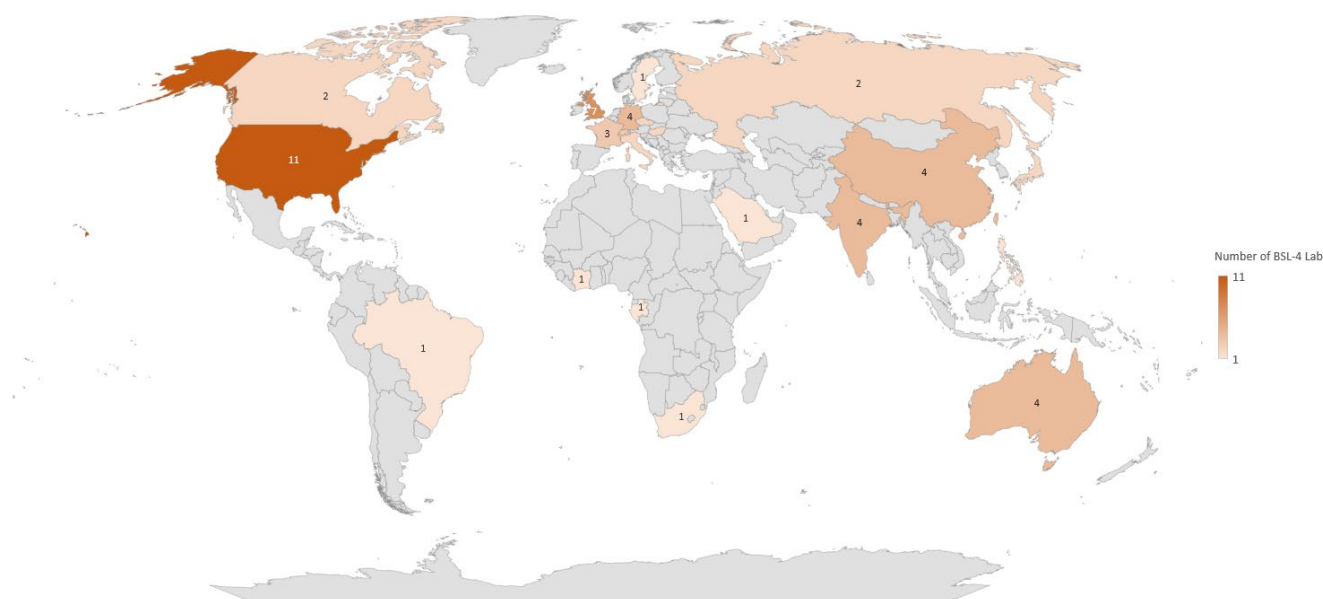


Figure 1. Number of BSL-4 labs by country.

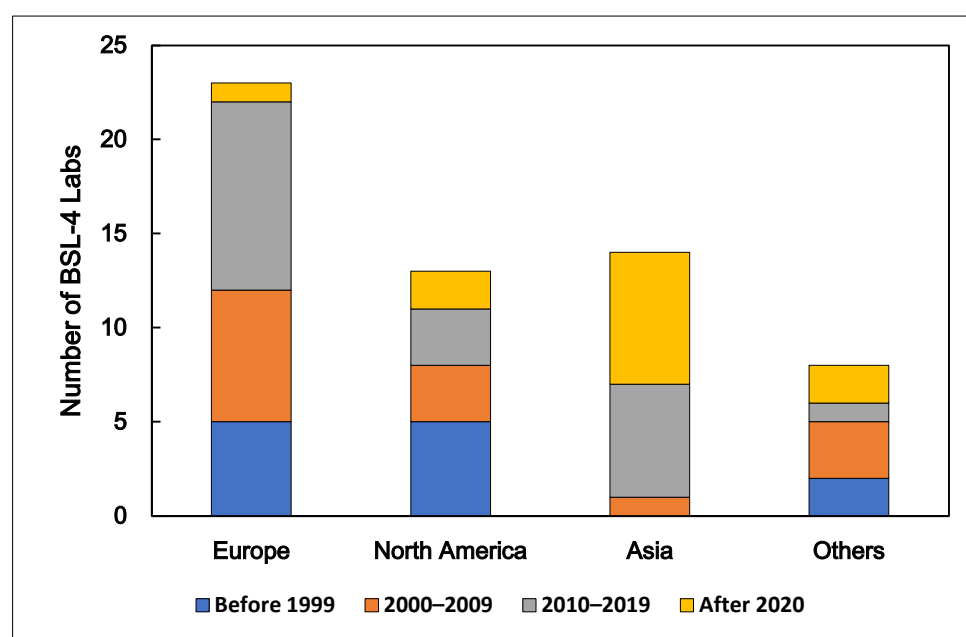


Figure 2. Number of BSL-4 labs in each region by time of construction.

2.2. Research Status in China

China has achieved significant progress in biosafety laboratory development despite its later initiation compared to its global counterparts. The establishment of a key regulatory framework began with the promulgation of the “General Guidelines for Biosafety in Microbiological and Biomedical Laboratories” in 2002, marking China’s first steps toward standardized biosafety practices. A major milestone was achieved in 2018 with the operational launch of two critical facilities: the Wuhan National Biosafety (Level 4) Laboratory and the Harbin National High-Level Biosafety Laboratory for Animal Disease Control [6,7]. These achievements demonstrate China’s growing capabilities in high-containment laboratory construction and management.

The Wuhan National Biosafety Laboratory (Level 4) represents China's most advanced high-containment facility, demonstrating exceptional airtightness performance through its stainless-steel welded enclosure system. Pressure decay tests conducted under standard operating conditions (500 Pa test pressure) showed a pressure drop of only 180 Pa over 20 min, significantly exceeding the maximum decay requirement of 250 Pa specified in GB 19489-2008 [13]. This outstanding performance is attributed to advanced laser welding techniques and stringent quality control measures for the steel/concrete hybrid structure. However, material consistency challenges, particularly in thermal expansion differentials between steel and concrete, require ongoing monitoring and adjustment in regional BSL-4 implementations. Independent evaluations, including WHO expert assessments, have confirmed the facility's robust isolation capabilities. Biological aerosol tests using *Serratia marcescens* and phage Φ X174 demonstrated that the isolation efficiency at all penetration points exceeded 99.99%. However, insufficient technical staff training remains a common challenge for China's biosafety network. The laboratory's design incorporates multiple features, including mechanical compression doors with ultrasonic leak detection and HEPA-filtered ventilation systems, making it a benchmark for modern BSL-4 containment architecture [14,15]. These technical achievements meet international standards (ISO 9972:2015, ASTM E779-19) while also fulfilling China's specific biosafety requirements for handling Category IV pathogens.

Since then, China has significantly expanded its biosafety infrastructure, now boasting numerous BSL-3 facilities and an increasing number of BSL-4 laboratories. This expansion has provided valuable experience in laboratory design, construction, and testing methods. Chinese national standards such as GB 19489-2008 and GB 50346-2011 have provided important guidance for laboratory construction, particularly in defining protection zones and enclosure airtightness requirements [13,16]. However, gaps remain in airtightness testing standards and research, with current studies predominantly focusing on concrete material formulations, steel structure welding techniques, and component-level performance, while paying less attention to integrated concrete/steel systems. Establishing P4 laboratories in China still faces some challenges. Notably, there are significant geographical disparities in the distribution of these laboratories, with most located in economically developed regions. Additionally, the biosafety management systems in place are far from comprehensive, and their enforcement needs to be strengthened. Financial constraints also hinder the effective operation of these laboratories. Furthermore, there is a significant shortage of qualified professionals [4].

Recent studies in China on laboratory enclosures have emphasized innovative design and efficiency assessment. Chen et al. (2022) [17] proposed a novel design for a nucleic acid testing laboratory utilizing air/membrane structures. These structures feature high airtightness and swift installation capabilities, and have been successfully applied across multiple countries, enhancing testing capacity. Wang et al. (2024) [18] suggested that intelligent building enclosures are becoming increasingly prevalent. These structures can autonomously adjust to environmental changes, thereby enhancing indoor comfort and improving energy efficiency. Concurrently, numerous scholars have rigorously assessed the performance of current building enclosures and offered viable suggestions for their enhancement.

Disparities in the development of biosafety laboratories across regions stem from the complex interplay of technological, policy, economic, and geopolitical factors. Developed countries such as the United States and European countries have leveraged their long-standing expertise in biosafety technology to establish and maintain high-level laboratories early on, supported by stringent regulatory frameworks and substantial government investments in public health infrastructure. In contrast, regions like Africa and parts of Asia

face significant technological gaps and rely heavily on international collaborations to build capacity, compounded by inconsistent policies and limited funding that hinder progress. The high construction and operational costs of BSL-4 laboratories further restrict their proliferation in economically weaker regions, while developed nations prioritize biosafety as a cornerstone of national security, allocating robust budgets to sustain advanced facilities. Historically, defense-related biosafety research in the US and Europe has driven early advancements, whereas emerging economies like China and India are now accelerating their efforts in response to recent pandemics and escalating biosecurity demands. These multifaceted disparities underscore China's need for tailored strategies to address regional imbalances in biosafety laboratory development.

3. Airtightness of Concrete Structures

3.1. Concrete Material Properties

3.1.1. Mixing Ratio Design

The enclosure structure of high-level biosafety laboratories has high requirements for the airtightness of concrete. The airtightness of concrete structures is closely related to the mixing ratio and molding of concrete materials. As a non-homogeneous material, concrete may inadvertently trap gas bubbles during production, and the evaporation of free water during hydration creates numerous air voids and capillary channels with varying morphologies. These features affect the concrete's compactness and strength, providing pathways for gas migration. Concrete strength and airtightness are closely related to its capillary porosity. Therefore, many factors influencing concrete strength also significantly impact its permeability. According to the Borromeo formula, a lower water/cement ratio results in higher compressive strength. Tang (2017) [6] studied seven different water/cement ratio concretes. They found that a higher water/cement ratio leads to more internal pore structures, resulting in poorer airtightness. Similarly, Liu et al. (2005) [7] found that the gas permeability coefficient of concrete decreases as compressive strength increases, establishing a strong linear relationship between compressive strength and gas permeability coefficient across different water/cement ratios. Xu et al. (2022) [19] found that basalt fiber reinforcement reduces concrete gas permeability by 65.7% (from $3.5 \times 10^{-16} \text{ m}^2$ to $1.2 \times 10^{-16} \text{ m}^2$ under -500 Pa) via pore structure refinement (42% porosity reduction by SEM). Overall, as the water/cement ratio increases, so does the concrete's gas permeability coefficient. The proportion of aggregate in concrete also affects its airtightness. It is generally accepted that adding aggregate to concrete worsens pore connectivity and increases pore tortuosity, thereby reducing its gas permeability coefficient and enhancing airtightness [20].

In summary, key factors affecting the design of the mixing ratio are the material proportioning and the molding process. The strength and airtightness of concrete are closely related to its capillary porosity. A lower water/cement ratio results in higher compressive strength; a higher water/cement ratio leads to more internal pore structure and poorer airtightness. As compressive strength rises, the gas permeability coefficient declines. Additionally, the incorporation of concrete aggregates typically also leads to a decrease in the gas permeability coefficient.

3.1.2. Material Additives

Concrete admixtures are diverse and are widely used to modify concrete properties. Adding these admixtures can lower the gas permeability coefficient, enhance the densification of the hardened concrete, and improve its resistance to salt erosion and freeze/thaw damage. This, in turn, enhances the mechanical properties and overall durability of the

concrete, extending its service life. Numerous studies have examined the effects of various admixtures, with some commonly used concrete admixtures summarized in Table 2.

Table 2. Commonly used concrete admixtures.

Name	Principle of Action	Material Efficacy	Other
Water reducing agent	A typical surfactant that adsorbs at the gas/liquid interface, effectively reducing surface tension and increasing the strength of the liquid films between foam bubbles, thereby improving foam stability.	Incorporation of water-reducing agents is beneficial to improve the airtightness of concrete specimens, among which the sulfonate water-reducing agent is the most effective one [21]. As the dosage of defoamer increases, the workability of the concrete mixture decreases. Additionally, the difference in setting time decreases, while the compressive strength ratio first increases and then decreases. The permeability coefficient of hardened concrete shows a decrease followed by an increase. Organosilicon defoamer has a notably stronger effect on enhancing concrete airtightness [21].	The uneven size and irregular shape of foam may affect the mechanical properties of concrete.
Defoamer	Using defoamers can refine the pore size and thus reduce the gas permeability of concrete.		Mercury pressure test results indicate that the defoamer increased the pore volume of the paste in the pore size range of 6–30 nm, while it decreased the pore volume in the range of 30–60 µm. However, in the pore size range of 60–300 µm, the pore volume increased at defoamer dosages of 0.0025% and 0.0075%, and the pore volume of the cement slurry first reduced and then slightly increased at defoamer dosages of 0.005% and 0.001% [22].
Air-entraining agent	Adding an air-entraining agent can effectively improve the airtightness of concrete.	As the dosage of the air-entraining agent increases, the permeability coefficient of the concrete specimen first decreases and then increases.	Cai (2024) [23] studied the joint effects of defoamers and air-entraining agents, proposing that the process of “first elimination, then air-entraining” can effectively regulate the air content of concrete, thus controlling the apparent quality of concrete components.
Air sealing agent	The amount of air sealing agent has a greater impact on the strength and air permeability coefficient of concrete.	Increasing the dosage of air sealing agent reduces the compressive strength and air permeability coefficient of concrete, while the lowest dosage of air sealing agent results in slightly poorer concrete flowability.	Bai et al. (2018) [24] prepared a new type of concrete airtightness agent using the pre-emulsified seed emulsion polymerization method, which is beneficial for improving the strength and airtightness of concrete.

3.2. Construction Technology

The key technologies in concrete construction include transportation, mixing, pouring, vibration, curing, etc., each of which affects the airtightness of the enclosure structure. Among them, the mixing process, pouring method, and vibration are critical steps. The complete process of concrete construction is shown in Figure 3.

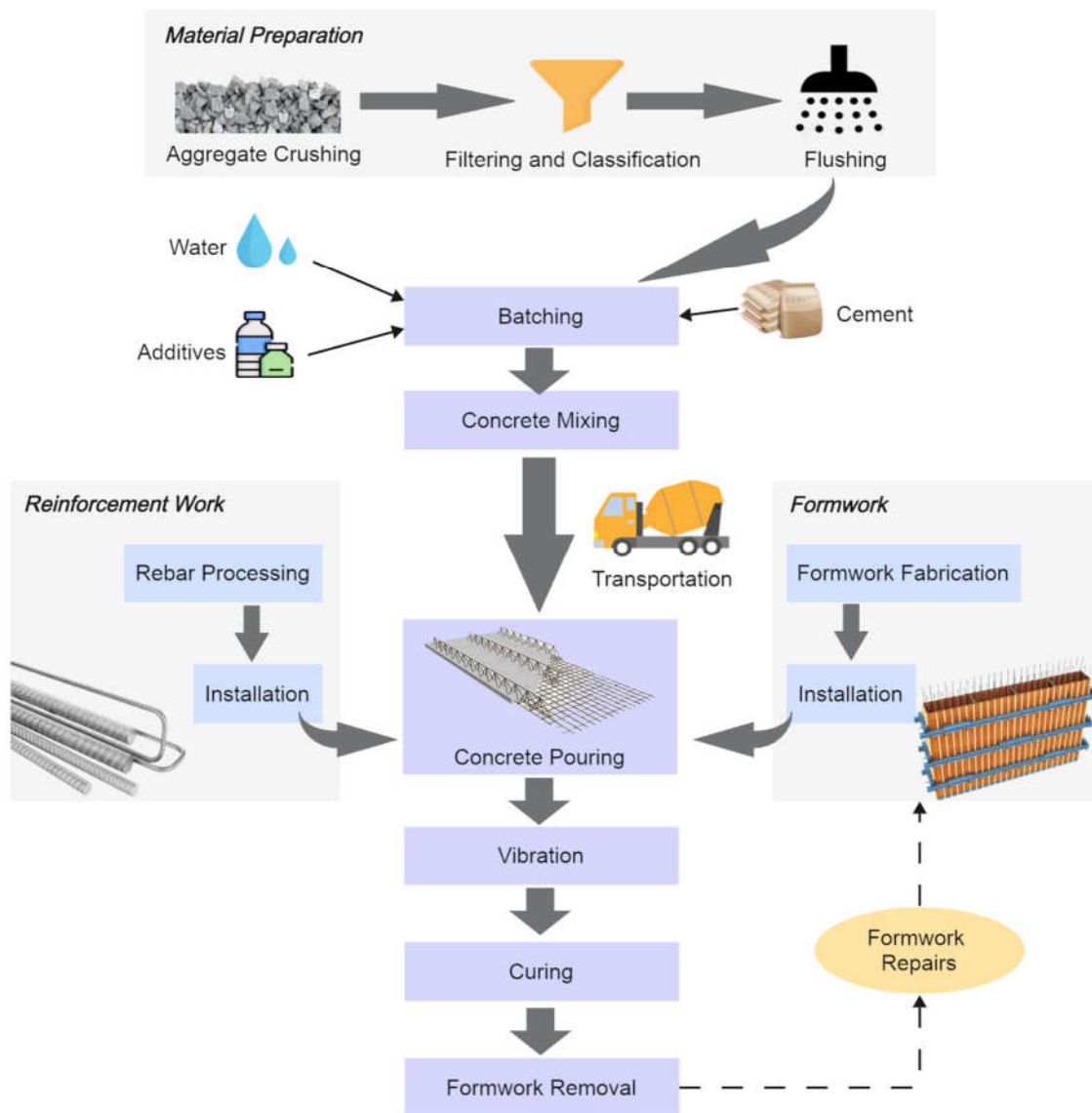


Figure 3. Concrete construction process flowchart.

Concrete site construction must strictly follow the specified sequence of material addition and mixing time during the mixing process. Aggregate gradation should be stored in sealed containers, and moisture content must be measured before each work shift. Adjustments to water consumption and aggregate dosage should be made promptly. To ensure the airtightness of concrete, Lian (2016) [25] mentioned that a recommended way is to dry mix the admixture and fine aggregate first, then add cement along with a portion of the mixing water, and finally combine with the sequence of addition of coarse aggregate, water reducing agent, and the remaining mixing water.

Concrete pouring methods are mainly divided into full-scale pouring, segmental pouring, and inclined pouring [26]. Full-scale pouring is suitable for large-area concrete construction, requiring fast and uniform pouring to avoid prolonged surface exposure time, thereby reducing cracks and interface issues. Segmental pouring is suitable for complex structural construction projects, achieved by dividing the structure into multiple small segments for independent pouring, enhancing structural uniformity and density. Sloped pouring, including the slipform method and free-fall method, is suitable for concrete construction on slopes or inclined surfaces.

After mixing, transportation, and pouring, concrete may contain air bubbles that are formed due to entrapped air. The purpose of vibration is to discharge these bubbles, reduce the number of pores, enhance the density of concrete, and ensure the overall quality of the structure. After sufficient vibration, the trapped air is discharged, and the concrete aggregate particles are more tightly bonded. As a result, the concrete becomes dense, strong, durable, and smooth, with no honeycomb or air bubbles on the surface, making it resistant to infiltration [27].

When using concrete to cast walls, it is important to pour in layers. If the upper layer is too thick, the weight of the concrete will prevent air from being expelled from the lower layer. This trapped air forms pores in the concrete and results in inadequate vibration. The thickness of each layer depends on the selected vibrator. For example, when the length of the selected vibrating rod is 350 mm, the maximum vibrated layer thickness should be 300 mm [27]. Layers should be cast horizontally, being careful not to pile up or tilt the concrete. It is crucial to achieve a dense bond between layers to ensure that each layer of concrete is fully vibrated and compacted. When pouring the next layer, it can still be vibrated to create a dense connection between layers, thereby forming a solid, cohesive structure.

4. Airtightness of Steel Structures

4.1. Selection of Enclosure Structure and Components

Stainless steel enclosure structures are commonly used in high-level biosafety laboratories. This structure not only provides excellent airtightness, meeting the airtightness requirements of such laboratories, but also offers corrosion resistance, high-pressure resistance, and ease of disinfection and cleaning. Stainless steel plate seam-welded enclosure structures are currently the preferred situation for alterations in high-level biosafety laboratories. This solution can be implemented in existing laboratory buildings to minimize the need for extensive structural modifications, resulting in lower costs and shorter construction periods. On the other hand, color-coated steel plate enclosure structures are commonly used in biological safety laboratories where non-airborne biological factors are present. These structures can be safely operated with isolation devices. According to GB 50346-2011 “Technical Specifications for the Construction of Biosafety Laboratories”, the enclosure structures of high-level biosafety laboratories must be able to withstand air pressure loads caused by the abnormalities of the air supply fan or exhaust fan. Stainless steel enclosure structures fulfill this requirement effectively. Due to the limited pressure of color-coated steel plate enclosure structures, their airtightness is typically only tested using visual methods, such as the smoke test, to detect visible leaks in gaps. The indoor relative negative pressure should not exceed -100 Pa. When conducting airtightness testing on color-coated steel plate enclosing structures using the constant pressure method or pressure decay method, stainless steel plate seam-welded enclosure structures or epoxy-treated concrete enclosure structures are generally selected. The former is mainly suitable for new construction, renovation, expansion projects, or projects where the building structure has already been completed, while the latter is appropriate for new construction projects.

To ensure the airtightness of the stainless steel enclosure structure, structural components are made of 304 stainless steel. These components are welded and molded, then processed by aging treatment, CNC gantry milling, sandblasting, and painting. The stainless steel plates undergo laser-cutting undercutting, CNC bending, and molding [8]. In addition, the design of the enclosure structure and selection of components must consider various factors, such as seismic resistance, fire safety, waterproofing, corrosion resistance, etc. These considerations are crucial to ensuring the stability and safety of the laboratory under various extreme conditions. In the construction of high-level biosafety laboratories,

these characteristics of the enclosure structure are essential not only for maintaining biological safety inside the laboratory but also for the long-term stability of laboratory operation and maintenance.

4.2. Welding Performance and Welding Materials

The welding performance of steel structures depends on the physicochemical properties and composition of the base material, the selection of welding materials, welding processes, structural design, use conditions, etc. Common physical properties that affect welding performance include the following:

- (1) Differences in melting points make heat treatment difficult. The melting of the base material cannot be synchronized; if the melting point difference is too great, it may prevent successful welding of the materials.
- (2) Differences in electromagnetic properties lead to unstable arc combustion during arc welding.
- (3) Different linear expansion coefficients can easily cause welding stress concentration or cracks, which is also one of the key reasons why welded joints may fail to meet airtightness requirements.
- (4) Differences in thermal conductivity and specific heat capacity prevent the formation of the melt pool from being synchronized. Different rates of heat transfer result in increased thermal stress.

The impact of different welding materials on the welding quality is mainly reflected in the chemical composition matching, strength, corrosion resistance, process performance of welding materials, anti-porosity, and anti-cracking ability. The chemical composition of welding materials should be matched with the welded material to ensure the strength and durability of the weld. The thermal expansion coefficient, melting point range, thermal conductivity, and mechanical properties of the welding material and the material to be welded should be similar to avoid uneven thermal and residual stresses during the welding process, which could lead to cracking or deformation problems of the weld [28]. At the same time, the strength and corrosion resistance of the welding material should meet the requirements of the operating environment. For welded structures working in corrosive environments, welding materials with good corrosion resistance should be selected. Selecting the correct welding material is critical to ensuring welding quality. The suitable welding material should be selected according to the characteristics of the base material, welding method, operating environment, and expected welded joint performance. Welding materials mainly include welding rods, wires, fluxes, shielding gases, etc. Welding rods are coated with flux for arc welding with a molten electrode, while the wire is used as a filler metal or used simultaneously to conduct the wire. The selection of welding materials needs to consider factors such as the base material, welding position, welding current, and welding environment to ensure welding quality and safety. Welding rods are usually made of metal powders, alloys, or other materials, such as carbon steel, low alloy steel, stainless steel, cast iron, copper and copper alloys, aluminum and aluminum alloys, nickel-based alloy rods, and so on. Additionally, the continuous emergence of new welding materials helps improve welding efficiency and quality while reducing problems during the welding process.

4.3. Welding Process

There are many types of welding methods, and the selection of welding methods should take into account both usability and manufacturability. Welded joints obtained by different welding methods may have different organizations and properties. The following are several commonly used welding processes.

(1) Laser Welding.

Laser welding is a welding technology that utilizes a laser beam as a heat source. Compared to traditional welding methods, it provides faster welding speed and higher quality. The laser beam produced by the laser welding machine has extremely high energy density, which means that it can concentrate a large amount of energy in a very small area, thus achieving rapid melting and welding of materials. The size and shape of the laser beam can be precisely controlled by the optical system, allowing the welding process to be carried out with great precision, making it suitable for both small and complex welding tasks. Laser welding is widely used in industries such as automotive manufacturing, aerospace, medical devices, electronics, microelectronics, precision instrument manufacturing, and so on.

(2) Arc Welding.

Arc welding is a method of using the high temperature generated by an electric arc to melt metal, thereby connecting metal parts. Its basic working principle involves creating an electric arc between the welding rod (usually a metal welding rod or wire) and the workpiece. The heat from the arc melts the electrode and the workpiece contact part, forming a molten pool. As the molten pool cools, it forms a weld. There are many types of arc welding, including shielded metal arc welding (SMAW), metal inert gas welding (MIG/MAG), tungsten inert gas welding (TIG), submerged arc welding (SAW), etc.

With the continuous development of technology, various new welding processes have been enhanced from the original. Gorshkova (2021) [29] introduced new techniques that have been developed, such as K-TIG welding, hybrid laser welding, dual-arc welding, and magnetron-controlled electrosag welding. These innovations have increased productivity and improved joint strength, especially in refractory steels and titanium alloys.

Welding process parameters include many aspects, such as power source, current, voltage, polarity selection, welding speed, shielding gas, electrode diameter, etc., all of which directly influence welding outcomes. Additionally, the interplay between these parameters must be considered. For example, increasing welding current increases input heat, enlarging the weld pool and expanding the heat-affected zone; increasing voltage lengthens the arc, leading to weld spatter; and welding speed affects both welding efficiency and weld pool size [30]. All these parameter effects must be considered during the welding process.

5. Evaluation of Concrete, Steel Enclosures, and Use of Other Materials

5.1. Comprehensive Evaluation of Concrete and Steel Enclosures

Case comparisons between the steel/concrete hybrid structure of the Wuhan Institute of Virology and the all-steel enclosures of PHE demonstrate that the steel/concrete hybrid structure reduces initial costs compared to PHE's all-stainless-steel design. However, due to the mismatch in thermal expansion coefficients between concrete and steel, the steel/concrete hybrid structure requires more inspections and maintenance in the long term. PHE's epoxy-coated stainless steel offers higher resistance to chemical disinfection, but its complex geometry results in higher welding costs. Table 3 compares the costs of enclosure structures of the Wuhan and PHE laboratories. Since detailed construction cost data for the laboratories has not been disclosed, this table is estimated based on current international market prices for construction materials, as well as relevant documents and specifications. These differences illustrate how different countries' priorities regarding cost and durability influence their choices of building enclosures and construction techniques.

Table 3. Cost Comparison of PHE and Wuhan Enclosure Structures.

	PHE All-Stainless-Steel Enclosure	Wuhan Steel/Concrete Hybrid Enclosure
Total Cost	Initial Construction: USD 950–1600/m ²	USD 370–600/m ²
Material Cost	304 Stainless steel: USD 650–900/m ² (316 L: +20–30%) Epoxy coating: USD 50–80/m ²	Structural steel (Q235/Q355): USD 150–250/m ² Concrete (C30–C50): USD 80–120/m ² Fireproofing: USD 30–50/m ²
Process Cost	Laser welding: USD 150–200/m ² Precision joints: USD 100–150/m ²	Conventional welding: USD 60–100/m ² Formwork: USD 50–80/m ²
Cost Multiple	2–3 ×	1 × (baseline)
Inspection Frequency	Post-Construction Maintenance: Every 10 years	Every 5 years
Maintenance Cost	Coating refurbishment: USD 50–80/m ² /cycle Weld inspection	Crack repair: USD 20–40/m ² /cycle Joint sealing

Notes: Data sourced from GB 50346-2011, EN 13829:2000, global market prices, relevant industry reports, etc. All cost data reflects current market conditions. Due to regional price differences, European projects may be increased by 15–20%, while Asian projects may be decreased by 10%.

To support decision-making regarding enclosure selection, a comprehensive evaluation of the performance and economy of concrete and steel structures is necessary. Concrete structures offer high initial airtightness and long-term durability, with the potential for further enhancement through admixtures and proper construction techniques. However, they may require more maintenance to ensure joints and connections remain airtight over the long term.

Steel structures, particularly stainless steel enclosures, provide superior airtightness and pressure stability, along with corrosion resistance and ease of cleaning. The use of advanced welding techniques and high-quality materials ensures minimal leakage and long-term performance. However, steel structures may be more susceptible to damage from impacts and require careful handling during installation.

In terms of economics, concrete structures may have lower initial costs but higher maintenance costs over time. Steel structures, while potentially more expensive upfront, may have lower maintenance costs and a longer service life. When choosing between concrete and steel structures, decisions should be based on specific project requirements, including budget constraints, construction timelines, and long-term operational needs.

Regional policy frameworks and financing mechanisms further complicate the technical and economic trade-offs between enclosure types. As shown in Table 4, the selection of steel/concrete hybrids versus all-steel designs often reflects deeper institutional priorities—EU regulations prioritize seal reliability over the lifecycle by mandating the use of stainless steel, while China’s subsidy policies actively promote hybrid structures as a cost-effective solution. Table 4 systematically compares how these policy-economic factors ultimately manifest as measurable airtightness outcomes across different governance contexts.

Table 4. Policy/Economic Factors Driving Enclosure Structure Selection.

Region	Policy Drivers	Funding Profile	Dominant Enclosure Structure	Airtightness Impact	Cost Ratio *
EU	EN 13829:2000 lifecycle requirements Strict decontamination protocols	High public health budgets 20-year depreciation cycles	Laser-welded stainless steel	Ultra-low leakage Excellent chemical resistance	1.0× (Baseline)
USA	ASTM E779-19 joint testing mandates Defense-funded biosafety priorities GB 19489-2008	Mixed federal/private funding Emphasis on rapid deploy ability	Composite panel systems	Moderate leakage Reliant on gasket seals	0.8×
China	attenuation limits “Localized innovation” subsidies	State capital projects 5-year budget cycles	Steel/concrete hybrid	Variable performance Thermal cracking risks	0.6×
India	ISO 10648-2 compliance for aid projects Tropical climate adaptations	International grants (e.g., WHO) < USD 50 m/project caps	Gasket-sealed color steel	High leakage Frequent gasket replacement	0.3×
Africa	Donor-mandated EU/US standards Emergency outbreak response needs	>80% foreign-funded < USD 20 m/total facility	Modular prefabricated units	Poor long-term sealing Reliant on silicone sealants	0.2×

* Cost ratio normalized to EU stainless steel baseline. Data synthesized from [4,9,10,13,16,31,32] and Table 1 case studies.

5.2. Lifecycle Cost-Benefit Evaluation of Enclosure Systems

The selection of enclosure structures for high-level laboratories requires balancing three key dimensions: (1) initial construction feasibility, (2) long-term maintenance complexity, and (3) operational reliability requirements. While detailed cost data for individual facilities is often proprietary, comparative studies indicate that stainless steel welded structures typically offer better airtightness and longer maintenance intervals, whereas steel/concrete hybrid structures (e.g., Wuhan P4) reduce initial capital expenditures through localized material sourcing but require more frequent joint inspections due to thermal expansion differences.

The WHO Laboratory Biosafety Manual (4th edition, 2020) [33] emphasizes that high-containment facilities require comprehensive lifecycle cost management, though it does not provide specific cost ratios. The manual highlights two key principles: (1) regular maintenance is critical for maintaining isolation integrity, and (2) material selection must balance initial costs with long-term operational requirements (WHO, 2020 [33], Section 5.3).

China’s GB 50346-2011 standard specifies that steel/concrete hybrid structures require more frequent maintenance than all-steel designs, mandating comprehensive inspections every 5 years compared to 10-year intervals for welded stainless steel systems (GB 50346-2011, Clause 5.1.9). This difference stems from the need to monitor thermal expansion differentials at material interfaces. Operational data from reference facilities such as Wuhan P4 (hybrid) and PHE (steel) confirm these maintenance intervals, but actual costs vary by region due to labor and material factors.

5.3. Other Materials Applied in the BSL-4 Laboratories

In addition to concrete and steel, other advanced materials are increasingly being utilized in BSL-4 laboratories to enhance airtightness, durability, and overall safety. These materials are selected based on their unique properties, which complement the stringent requirements of high-containment facilities.

(1) Composite Materials

Composite materials such as fiber-reinforced polymers (FRPs) are gaining attention due to their high strength/weight ratio, corrosion resistance, and adaptability in sealing joints and penetrations [34]. Their flexibility allows for seamless integration with existing structures, reducing potential leakage points [35].

(2) Epoxy Coatings and Sealants

Epoxy coatings are widely applied to concrete and steel surfaces to fill micro-pores and cracks, further enhancing airtightness. These coatings are chemically resistant and can withstand frequent decontamination procedures, making them ideal for laboratory environments [36,37].

(3) Glass and Acrylic Panels

For observation windows and visual access points, laminated glass or acrylic panels with airtight gaskets can be used [32]. These materials provide transparency while maintaining structural integrity and resistance to pressure differentials [38].

(4) High-Performance Gaskets and Membranes

Specialized gaskets made from silicone, EPDM rubber, or fluoropolymers are used to seal doors, vents, and equipment penetrations. These materials exhibit excellent elasticity and longevity, ensuring airtightness under dynamic pressure conditions [39,40].

(5) Nanotechnology-Enhanced Materials

Emerging nanomaterials, such as graphene oxide or nano-silica additives, are being explored to improve the water resistance of traditional materials like concrete and coatings [41]. These innovations aim to reduce porosity at the microscopic level [42].

(6) Fire-Resistant and Thermal Barriers

Materials like intumescent paints and ceramic fibers are integrated into the enclosure design to meet fire safety standards without compromising airtightness. These barriers expand under heat, sealing gaps to prevent smoke or pathogen leakage in an emergency [43,44].

The selection of materials for BSL-4 laboratories extends beyond conventional choices, requiring the integration of composite materials, advanced coatings, and nanotechnology to address multifaceted challenges. Future research should focus on optimizing these materials for long-term performance under operational stresses, including thermal cycling, chemical exposure, and mechanical wear.

6. Airtightness Test Methods and Evaluation Criteria

6.1. Airtightness Test Methods

Building airtightness testing is the fundamental method for evaluating building enclosure performance. Commonly used airtightness testing methods include the pressure decay method, helium mass spectrometry leak detection method, soap liquid method, heating method, flow method, and water pressure method [45]. The introduction of each method and the recommended ratings for their use are shown in Table 5.

Table 5. Recommended ratings for airtightness test methods.

Method Name	Recommended Rating	Method Description	Reason for Recommendation
Pressure Decay Method	★★★★★	Apply a certain pressure to the product or system being tested, record the initial pressure value, then turn off the gas or liquid supply source and measure the pressure change over a period of time to determine airtightness.	This method enables quantitative airtightness assessment and is the most widely adopted in BSL-4 laboratories. It is suitable for testing the airtightness of high-level biosafety laboratories, such as BSL-3 or BSL-4 laboratories.
Helium Mass Spectrometry Leak Detection Method	★★★★★	Apply gas around the product or test sample and use a leak detector to detect gas leakage, thereby assessing its airtightness performance.	The most reliable method, offering advantages of high accuracy, fast response, locatable leaks, and environmental adaptability. However, it requires a specialized helium mass spectrometry leak detector, which is relatively expensive.
Soap Liquid Method	★★★★☆	Apply soap liquid to the surface of the tested component, then fill it with gas at a certain pressure. The gas will blow soap bubbles through the leakage point, thus exposing the leakage location and observing whether there are bubbles generated to determine the airtightness.	Simple and easy, safe and reliable, can determine the leak location. However, the bubble formation can be influenced by leakage size and pressure difference. If the leakage volume is small, bubble formation takes longer, and bubbles may be too small or fragile to observe.
Heating Method	★★★★☆	Utilizing the thermal expansion and contraction properties of gas, the component is heated to expand the internal gas, then observe whether there are bubbles or water column formation to determine the airtightness.	Simple to operate, no complex equipment required, and bubbles or water columns can be directly observed. However, it is unsuitable for temperature-sensitive equipment and lacks sensitivity for detecting small leaks. Often used for chemical laboratory equipment.
Flow Rate Method	★★★★☆	Intermittently inject gas at a certain pressure into the tested component, use a mass flowmeter or volume flowmeter to detect changes in gas flow rate, and determine whether the leakage rate of the component is within the required acceptable range.	This system has a simple design, wide applicability, accurate test results, and can quantitatively assess airtightness performance. However, it can only detect overall airtightness and cannot locate the leakage position.
Water Pressure Method	★★★☆☆	Based on the principle of pressure, the component is submerged in water, subjected to a certain water pressure, and observed for water ingress or the formation of bubbles to determine its airtightness.	This method is simple and easy to perform, enabling direct visualization of leak points. However, once water enters the component, it may cause irreparable damage, and the water must be dried, wasting manpower and resources.

Note: 5 stars—Excellent, 4 stars—Good, 3 stars—Average, 2 stars—Below average, 1 star—Poor.

6.2. Airtightness Testing Standards

6.2.1. International Standards

Developed countries have prioritized research on building airtightness for decades, establishing more comprehensive building codes with stringent airtightness requirements.

Building airtightness testing has been conducted extensively, with detailed technical specifications for overall building airtightness and standardized approaches that demonstrate notable regional variations. This review examines six internationally recognized standards—ISO 9972:2015 [9], EN 13829:2000 [31], ASTM E779-19 [10], JIS A 2201:2018 [46], ATTMA TS1 (2022) [47], and CAN/CGSB-149.15 (2020) [48]—comparing their technical specifications, applicability, and impact on the building enclosure performance. The assessment is presented in Table 6, focusing on five critical dimensions: regional applicability, testing protocols, quantitative metrics, calculation methods, and regulatory implementation.

Table 6. Comparative analysis of international building airtightness testing standards.

Criteria	ISO 9972:2015 [9]	EN 13829:2000 [31]	ASTM E779-19 [10]	JIS A 2201:2018 [46]	ATTMA TS1 (2022) [47]	CAN/CGSB-149.15 (2020) [48]
Regional Scope	International benchmark standard	Mandatory in EU member states	Mainstream in North America	Japan-specific	UK construction standard	Canadian national standard
Test Method	Single-mode fan pressurization	Dual-mode (Method A/B)	Single-point/Multi-point options	ISO 9972 + Japanese adaptation	Residential/Non-residential classification	Single-zone/Multi-zone testing
Metrics	Q_{50} (m ³ /h) ACH_{50} (h ^{−1}) A_{50} (m ²)	Same as ISO 9972	CFM_{50} (ft ³ /min) ACH_{50} (h ^{−1}) ELA (in ²)	Q_{50} (m ³ /h) ACH_{50} (h ^{−1}) C-value (cm ² /m ²)	Q_{50} (m ³ /h) ACH_{50} (h ^{−1}) ELA (m ²)	Q_{50} (m ³ /h) ACH_{50} (h ^{−1}) ELA (cm ²)
Equations	$Q = C \cdot (\Delta P)^n$ $ACH_{50} = Q_{50} / V$ $A_{50} = Q_{50} / (0.61 \cdot \sqrt{50})$	Same as ISO 9972	Imperial unit adaptation ELA = $CFM_{50} / (0.61 \cdot \sqrt{50})$	Same as ISO 9972 C-value = ELA / floor area	Same as ISO 9972	Same as ISO 9972 Climate zone adjustments
Compliance	Referenced by other standards	Incorporated in EU regulations Design verification method B	Used for US certification	Japanese Building Code reference	UK Building Regulations	Canadian National Building Code
Special Notes	None		Single point allowed	Mandatory C-value reporting	Building type differentiation	Adaptation to extreme climates

Abbreviations: ACH_{50} : Air Changes per Hour at 50 Pa; ELA: Equivalent Leakage Area; C-value: Leakage area per unit floor area (Japan-specific). Unit Consistency: ISO/EN/JIS/ATTMA/CGSB use metric units (m³/h, m²); ASTM uses imperial units (CFM, in²).

The analysis demonstrates how regional building practices, climatic factors, and performance requirements have influenced the development of distinct testing frameworks. While ISO 9972 establishes an international benchmark, EN 13829 and ASTM E779 reflect European and North American regulatory approaches, respectively. Notably, JIS A 2201 incorporates Japan-specific parameters such as the C-value, while CAN/CGSB-149.15 addresses Canada's strict climate demands. These variations highlight the necessity of adopting context-specific standards to accurately assess building enclosure performance.

6.2.2. China's Standards

China's research on building airtightness is still in its infancy, with most existing literature concentrated in the past decade and a lack of referenceable airtightness measurement data. In recent years, the concern for building airtightness has been rising. For concrete and stainless steel monolithic welded enclosure structures, airtightness testing methods are generally the pressure decay method test or the constant pressure method [11].

(1) Pressure Decay Method

The pressure decay method involves testing the temperature and pressure inside a sealed enclosure at regular time intervals from the initial to the final stage. The actual leakage rate of the enclosure is calculated based on the results of temperature and pressure changes over time. Usually, the initial and final readings are used for calculation, and the remaining intermediate readings are used to control the test conditions. ISO 10648-2:

1994 “Sealed chambers Part 2: Classification of sealing and its test methods” [32] gives the formula for calculating the hourly leakage rate T_f using the pressure decay method:

$$T_f = \frac{60}{\tau} \left(\frac{p_n T_1}{p_1 T_n} - 1 \right) \quad (1)$$

where T_f is the cavity hourly leakage rate, h^{-1} ; τ is the duration of the test, min; p_n is the final reading of the absolute pressure in the room, Pa; p_1 is the initial reading of the absolute pressure in the room, Pa; T_1 is the initial reading of the temperature, K; T_n is the final reading of the temperature, K.

(2) Constant Pressure Method

The constant pressure method maintains the negative pressure in the sealed enclosure at a constant level, measures the flow rate of the discharged gas, and calculates the leakage rate based on the ratio between the leakage volume and the volume of the enclosure. As the accuracy of the cavity volume measurement directly affects the numerical calculation of the leakage rate, it is usually used to evaluate the leakage rate of sealing cavities with higher leakage rates or regular internal dimensions of the cavity. GB 19489-2008 [13] gives the formula for calculating the hourly leakage rate T_f by the constant pressure method:

$$T_f = Q / (V_1 - V_2) \quad (2)$$

where Q is the air volume maintaining a constant pressure difference, m^3/h ; V_1 is the room’s spatial volume, m^3 ; V_2 is the room’s object volume, m^3 .

Both the pressure decay method and the constant pressure method reflect the extent of gas leakage from the enclosure. The two methods are interchangeable in terms of their testing principles, although there are differences in their formulas [32]. Due to the presence of gaps in the building enclosure, when a pressure difference exists between the interior and exterior of the enclosure, air flows through the gaps from the high-pressure side to the low-pressure side. The volume of air flowing into or out of the room through the gap represents the leakage rate of the enclosure structure.

China’s current national standards GB 19489-2008 [13] and GB 50346-2011 [16] specify the airtightness requirements for P4 laboratories, as shown in Table 7.

Table 7. Laboratory airtightness requirements.

Clause No.		Airtightness Requirements
GB 19489	6.4.8	The airtightness of the enclosure of the laboratory protection zone shall meet the following requirements: under the condition of closing all access to the test room and maintaining the temperature in the room at the upper limit of the design range, when the air pressure in the room rises to 500 Pa, the natural decay of the air pressure within 20 min shall be less than 250 Pa.
GB 50346	10.1.6 No.3	Level 4 biosafety laboratories shall use the primary laboratory pressure decay test method under both positive and negative pressure conditions.
	3.3.2	The relative negative pressure of the main laboratory room is -500 Pa, and after 20 min of natural decay, the relative negative pressure should not exceed -250 Pa.

7. Other Measures to Improve Airtightness

7.1. Setting the Airtight Valve and Airtight Door

7.1.1. Airtight Valve

The airtightness requirements of the enclosure structure are extremely high, and the ventilation and air conditioning piping system is the biggest risk to the enclosure structure,

with many penetrating ducts, large diameters, and sealing difficulties. Installing biosafety airtight valves at key nodes of the ventilation and air conditioning duct system and ventilation and filtration equipment ducts in the building enclosure can effectively guarantee the airtightness of the enclosure structure, isolate the laboratory with biosafety protection equipment from the external environment, and realize the possibility of providing test conditions for different types of ventilation and filtration equipment to provide the possibility of test conditions [49].

7.1.2. Airtight Doors

To meet the airtightness requirements of biosafety laboratories, mechanical compression doors and inflatable airtight doors are usually used to provide safe isolation between the protected and non-protected areas of the laboratory. The mechanical compression door is mainly composed of a door frame, door panel, seal ring, mechanical compression mechanism, and access control device. Its working principle is as follows: when the door is closed, the door panel and door frame are pressed together by the seal ring through the compression mechanism, ensuring no leakage between them. After the installation of an airtight door, the ultrasonic penetration method is used for leakage detection. Mechanical compression airtight doors are durable and, unlike inflatable airtight doors, do not require inflation of the seal rings. Additionally, they do not need to be replaced regularly due to seal ring aging, thereby reducing maintenance costs [8]. Liu et al. (2025) [50] found that nano-silica epoxy coatings achieve a 90% reduction in steel joint leakage rates ($0.05 \rightarrow 0.005 \text{ m}^3/(\text{h} \cdot \text{m})$) by filling micro-cracks $<50 \text{ }\mu\text{m}$, which is critical for the sealing of doors and windows in BSL-4 laboratories. In addition, the study on airflow distribution in mobile BSL-4 laboratories also highlighted the importance of airtight doors in maintaining the directional airflow and preventing cross-infection [51]. The opening and closing of airtight doors can significantly affect the airflow patterns and the generation of vortices, which in turn influence the deposition and removal of bioaerosols. Therefore, the use of mechanical compression doors not only ensures airtightness but also helps in maintaining a stable and controlled airflow environment within the laboratory.

7.2. Through-Wall Equipment and Sealing Measures

High-level biosafety laboratories through-wall (floor, ceiling) equipment mainly includes airtight doors, chemical shower devices, autoclaves, transfer windows, transfer channels, high-efficiency filtration air supply and exhaust vents, airtight floor drains, etc. The airtightness of these devices themselves and the installation of the airtightness of the frame are important components of the laboratory airtightness. When installing the outer frames, stainless steel outer frames should be welded and embedded in the enclosure to ensure sealing. High-level biosafety laboratory through-wall (floor, ceiling) pipes mainly include ventilation pipes, water supply and drainage pipes, and strong and weak electric cables. The airtightness of these pipes themselves and the sealing treatment of wall penetration openings also significantly impact the laboratory's airtightness. Overseas, specialized embedded components are often used to eliminate temporary construction openings, such as professional pipe wall penetration sealers or liquid groove seals with centralized cable trays to ensure the integrity and airtightness of laboratory walls [52]. Common sealing measures use materials such as tape, silicone strips, silicone coatings, liquid coatings, foam sealing strips, and polyurethane foam [53]. These materials can achieve good airtightness when installed perfectly. However, the performance of sealing materials is highly sensitive to construction quality, especially tape, silicone-based materials, and foam sealing strips, whose performance mainly depends on construction quality.

7.3. Adopting the Dynamic Pressure Control Design Method

The dynamic pressure control design method can effectively improve the stability of the differential pressure of the BSL-4 laboratory. The core technology of the dynamic pressure control design method involves extending the cycle of the variable air volume valve for indoor pressure measurement. By adopting a dynamic pressure control design method that combines a constant supply and variable exhaust system with parallel variable air volume valves, the stability of laboratory pressure gradients is ensured, interference resistance is enhanced, and the directionality of airflow organization is achieved [49].

7.4. Microbial Testing of Laboratory Tightness

The integrity of containment systems in high-containment laboratories, particularly those operating at BSL-4, is critical for preventing the escape of hazardous microorganisms, necessitating rigorous microbial testing of laboratory tightness. Aerosol challenge testing serves as a direct method to detect microbial leakage, involving the aerosolization of non-pathogenic tracer microorganisms such as *Bacillus subtilis* spores or *Serratia marcescens* within the lab and subsequent air sampling outside critical barriers. The acceptance criteria for this test require no viable tracer microorganisms to be detected outside containment. Complementary to this approach, particle counting tests utilize inert particles like polystyrene latex spheres to simulate microbial aerosols, with laser particle counters scanning door seals and filter housings to verify that external particle counts remain significantly lower than internal levels, typically below 0.01% leakage [54,55].

Filter integrity testing is another critical component of containment verification, particularly for HEPA/ULPA filters that serve as final barriers against microbial escape. This testing involves introducing aerosolized dioctyl phthalate (DOP) or polyalphaolefin (PAO) upstream and scanning downstream with photometers or particle counters, with permissible leakage thresholds set at $\leq 0.01\%$ of upstream concentration. Smoke testing offers visual confirmation of proper airflow patterns, ensuring smoke flows inward in negative-pressure laboratories without escaping containment zones [56]. Additionally, biological indicator testing serves to verify decontamination efficacy by placing spore strips in critical areas, incubating them post-fumigation with agents like hydrogen peroxide vapor, and confirming the absence of microbial growth [57].

These diverse testing methods, when combined and aligned with rigorous regulatory standards such as those established by the WHO, CDC, or ISO 14644, create a robust system for maintaining and verifying containment integrity in BSL-4 laboratories [58]. Regular application of these tests ensures the ongoing effectiveness of containment systems, providing multiple layers of protection against accidental microbial leakage and safeguarding both laboratory personnel and the external environment from potential exposure to hazardous pathogens. The comprehensive nature of this testing regime reflects the extraordinary precautions necessary for working with the world's most dangerous microorganisms in the highest-level containment facilities.

8. Conclusions

This paper discusses the current state of global P4 laboratory construction and the specifications for airtightness requirements. It examines the material properties, construction methods, and evaluation techniques related to the airtightness of concrete and steel structures. The analysis focuses on how various materials, structural designs, and construction methods affect the airtightness of enclosure structures. Based on these findings, construction plans to enhance airtightness are proposed, and the following conclusions are drawn.

- (1) From the operation practice of P4 laboratories, the number of high-level biosafety laboratories in China needs to be further increased, with plans to form a reasonable national network of high-level biosafety laboratories by 2025. There are still some safety loopholes in the management of P4 facilities, which need to be unified. In order to improve the accuracy and effectiveness of enclosure structure airtightness testing, relevant national standards still need to be formulated and revised. Currently, China particularly needs to accumulate information related to the long-term operational reliability of P4 facilities, including laboratory design, building materials, construction processes, construction supervision, and qualification certification. Relevant regulations and standards still need to be further improved. In the future, China can draw on the mature experiences and relevant standards of other countries in the construction of high-level biosafety laboratories to improve its national standards, laboratory management, and maintenance, providing references for the biosafety construction and application of high-level biosafety laboratories in China. Given the high requirements for the construction and management of high-level biosafety laboratories and the significant investment costs involved, it is advisable to first strengthen support for the development of key technologies and related disciplines, gradually forming and leveraging their capabilities and advantages.
- (2) The airtightness of concrete is closely related to its capillary porosity, with influencing factors including material proportions and forming processes. Admixtures significantly affect concrete airtightness. Water-reducing agents improve the airtightness of concrete specimens; defoamers refine pores and reduce concrete permeability; air-entraining agents enhance airtightness, but increased dosage leads to a temporary decrease followed by an increase in permeability coefficient; when compressive strength and permeability coefficient decrease, airtightness agent dosage increases.
- (3) Stainless steel enclosure structures are airtight, corrosion-resistant, high-pressure resistant, and easy to disinfect and clean. They are widely used in high-level biosafety laboratories. A stainless steel plate seam-welded enclosure structure is the preferred option for remodeling projects, with low cost and a short construction period. Color steel plate enclosure structures for non-airborne biological factors of the laboratory to ensure airtightness are usually made of 304 stainless steel, after welding, molding, aging treatment, CNC machining, sandblasting and painting, and other processes. The welding performance of steel structures is affected by the physical and chemical properties of the base material, chemical composition, welding material selection, process, structural design, use conditions, and other aspects. Welding material selection affects the welding quality; the chemical composition matching, strength, corrosion resistance, process performance, anti-porosity, and crack resistance need to be considered. Different welding methods to obtain the organization and properties of the welded joints include new technologies such as K-TIG welding, hybrid laser welding, dual-arc welding, and magnetron electro slag welding to improve productivity and joint strength.
- (4) Airtightness test methods include the pressure decay method, helium mass spectrometry leak detection method, soap method, heating method, flow method, water pressure method, etc., among which the pressure decay method and helium mass spectrometry leak detection method are more recommended. In developed countries, the research on the airtightness of the enclosure structure is sufficient, and the relevant building codes are more complete, with higher requirements for airtightness. China's research on building airtightness is still in the primary stage, and the current national standards still need to be further improved.

- (5) From a holistic perspective, to systematically improve China's biosafety laboratory infrastructure, a three-phase roadmap is proposed. In the short term, interdisciplinary training programs (e.g., biosafety engineering) should be prioritized to build technical capacity. For the mid-term, piloting regional laboratories incorporating hybrid materials could optimize cost/performance ratios. Long-term efforts should focus on establishing a centralized database for airtightness performance metrics, enabling data-driven standardization. This tiered approach balances immediate needs, incremental innovation, and sustainable governance for future biosafety challenges.

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Nomenclature

The following acronyms and abbreviations are used in this manuscript:

BSL-4	Biosafety Level 4
P4	Physical Containment Level 4 (equivalent to BSL-4)
CNAS	China National Accreditation Service for Conformity Assessment
GB	Guobiao (Chinese National Standards)
ISO	International Organization for Standardization
ASTM	American Society for Testing and Materials
EN	European Standard
JIS	Japanese Industrial Standards
ATTMA	Air Tightness Testing & Measurement Association
CGSB	Canadian General Standards Board
ACH ₅₀	Air Changes per Hour at 50 Pa
ELA	Equivalent Leakage Area
CFM	Cubic Feet per Minute

Appendix A

Table A1. Global Construction of BSL-4 Laboratories.

Region	Country	Number	Name of Laboratory and Organization	Year of Construction	Area (m ²)
Europe	UK	7	Centre for Emergency Preparedness and Response, Public Health England (PHE)	1974	105
			Defence Science and Technology Laboratory (Dstl), Ministry of Defense	2005	335
			High Containment Large Animal Facility (HCLAF), Pirbright Institute	2015	257
			The Francis Crick Institute Containment 4 facility (formerly NIMR), The Francis Crick Institute	2016	298
			Centre for Infections (CFL), United Kingdom Health Security Agency (UKHSA)	2013	30
			National Institute for Biological Standards and Control (NIBSC), The Medicines and Healthcare Products Regulatory Agency, Department of Health & Social Care	1975	118
			Public Health England (PHE) Harlow, United Kingdom Health Security Agency (UKHSA)	2024	unknown
	Germany	4	Institute for Virology, Philipps University of Marburg	2007	152
			Bernhard Nocht Institute for Tropical Medicine (BNI)	2014	400
			Centre for Biological Threats and Special Pathogens, Robert Koch Institute, Federal Ministry of Health	2018	330
			Friedrich Loeffler Institute (FLI), Federal Research Institute for Animal Health	2015	405
	France	3	DGA Maîtrise NRBC, General Directorate of Armaments (DGA)	2013	178
			Institut de Recherche Biomédicale des Armées (IRBA), Army Health Service (SSA)	2015	240
			Jean Mérieux-Inserm (Institut national de la santé et de la recherche médicale), French National Institute of Health and Medical Research	1999	400
	Switzerland	3	Spiez Laboratory	2014	118
			Laboratory of Virology, Geneva University Hospitals	unknown	36
			Institute of Medical Virology, University of Zurich	unknown	25
	Italy	2	National Institute for Infectious Diseases Lazzaro Spallanzani (IRCCS)	2000	unknown
			Laboratory of Clinical Microbiology, Virology and Bioemergencies (CLIMVIB), Luigi Sacco University Hospital, University of Milan	unknown	unknown
	Czech Republic	2	Laboratory for Biological Monitoring and Protection, National Institute for Nuclear, Chemical, and Biological Protection	2007	28
			Department for Biological Defence, Military Institute of Health	2007	50
	Hungary	2	National Biosafety Laboratory (OKI), National Public Health Institute (former National Center for Epidemiology)	2002	163
			Szentágothai Research Center, University of Pécs	2017	55
	Russia	2	State Research Center of Virology and Biotechnology (VECTOR), Russian Federal Service for Surveillance on Consumer Rights Protection and Human Wellbeing	1980	1440
			48th Central Scientific Research Institute Sergiev Posad, Ministry of Defense	1979	unknown
	Sweden	1	Folkhälsomyndigheten (FOHM), Public Health Agency of Sweden	2001	136

Table A1. Cont.

Region	Country	Number	Name of Laboratory and Organization	Year of Construction	Area (m ²)
North America	US	11	Integrated Research Facility at Rocky Mountain Lab (IRF, RML), National Institutes of Health (NIH), National Institute of Allergy and Infectious Diseases (NIAID)	2008	1145
			The Betty Slick and Lewis J. Moorman, Jr. Laboratory Complex, Texas Biomedical Research Institute	1970	114
			Center for Biodefense and Emerging Infectious Diseases Shope Laboratory, The University of Texas Medical Branch	2003	186
			Galveston National Laboratory, The University of Texas Medical Branch	2010	1022
			National Bio and Agro-Defense Facility (NBAF), US Department of Homeland Security, US Department of Agriculture	2024	4084
			High Containment Core (HCC) (formerly the Viral Immunology Center), Georgia State University	1988	60
			Special Pathogens Branch, Centers for Disease Control and Prevention (CDC)	1988	533
			National Biodefense Analysis and Countermeasures Center (NBACC), US Department of Homeland Security	2008	980
			Integrated Research Facility at Fort Detrick (IRF - Frederick), National Institutes of Health (NIH), National Institute of Allergy and Infectious Diseases (NIAID)	2013	1305
			US Army Medical Research Institute of Infectious Diseases (USAMRIID), US Army Medical Research and Materiel Command, Department of Defense	1969	1186
			National Emerging Infectious Diseases Laboratories (NEIDL), Boston University	2017	1202
Asia	Canada	2	Vaccine and Infectious Disease Organization, Centre for Pandemic Research, University of Saskatchewan	2024	unknown
			National Microbiology Laboratory (NML), Public Health Agency of Canada	1999	185
	China	4	High-Level Biosafety Primate Experimental Center, Institute of Medical Biology, Chinese Academy of Medical Sciences	2019	3000
			Wuhan Institute of Virology, Chinese Academy of Sciences	2016	200
			Chinese Center for Disease Control and Prevention, National Health Commission	Under construction	Pending
			Chinese National High Containment Facilities for Animal Diseases Control and Prevention, Harbin Veterinary Research Institute	2018	17000
	India	4	Microbial Containment Complex (MCC), National Institute of Virology	2012	848
			National Institute of High-Security Animal Diseases (NIHSAD), Indian Council of Agricultural Research	2000	unknown
			Advanced Biological Defence Research Centre (ABDRC)	Under construction	Pending
			Rajiv Gandhi Centre for Biotechnology (RGCB)	Under construction	Pending
	Japan	2	Murayama Annex, National Institute of Infectious Diseases (NIID)	2015	unknown
			National Research Center for the Control and Prevention of Infectious Diseases, Nagasaki University	2021	unknown
	Taiwan, China	1	Institute of Preventive Medical Research (IPMR), National Defense University	unknown	unknown
	Korea	1	Osong BSL-4 Laboratory, Korea Centers for Disease Control and Prevention Agency	2017	300
	Saudi Arabia	1	National Health Laboratory, Saudi Ministry of Health	Pending	Pending
	Singapore	1	Defence Science Organisation (DSO) National Laboratories	2025	Pending
	Philippines	1	Virology Institute of the Philippines	2024	Pending

Table A1. Cont.

Region	Country	Number	Name of Laboratory and Organization	Year of Construction	Area (m ²)
Africa	South Africa	1	Special Pathogens Unit, National Institute for Communicable Diseases (NICD)	1979	unknown
	Gabon	1	International Center for Medical Research of Franceville (CIRMF)	2008	unknown
	Côte d'Ivoire	1	Institut Pasteur de Côte d'Ivoire	Pending	Pending
Oceania	Australia	4	Australian Centre for Disease Preparedness (formerly Australian Animal Health Laboratory), Commonwealth Scientific and Industrial Research Organization (CSIRO)	1985	567
			Emerging Infections and Biohazard Response Unit (EIBRU), Westmead Hospital	2007	86
			Queensland Health Forensic and Scientific Services (QHFSS)	2007	150
			National High-Security Quarantine Laboratory (NHSQL), Peter Doherty Institute for Infection and Immunity	2014	90
South America	Brazil	1	Pan American Foot and Mouth Disease Center (PANAFTOSA)	2021	unknown

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