MERRICK WHITE PAPER

BOUNDARY INTEGRITY TESTING OF CL3 (BSL3) BIOLOGICAL CONTAINMENT LABORATORIES

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EXECUTIVE SUMMARY

Biological laboratories that work with infectious agents are often required to have some level of boundary integrity testing (also commonly referred to as leak/air tightness testing). This paper is a review of current guidelines and standards in the Biocontainment Industry and proposes a means of testing new construction of CL3 laboratories to ensure that an acceptable level of quality is achieved. Boundary integrity testing of the laboratory space for Biocontainment facilities achieves three primary objectives:1) containment of an aerosolized agent; 2) containment of fumigants (decontamination gases); and 3) verification of the quality of the CL3 (BSL-3, SAPO-3, ACDP-3) boundary construction. Migration of fumigants is of most concern since laboratory ventilation systems are designed to maintain directional airflow into a contaminated zone and room fumigation is usually performed when the ventilation system is isolated at the room or zone level.

For CL3-Ag (BSL-3Ag, SAPO-4) and CL4 (BSL-4, ACDP-4) spaces, industry accepted standards have been pressure decay tests, as identified in the Canadian Biosafety Standard and the USDA ARS Facility Design Standards. This report focuses on boundary integrity testing methods for CL3 (BSL-3, SAPO-3, ACDP-3), laboratories, which are less defined.

Testing strategies and acceptance criteria for CL3 room leakage are often subjective rather than definitive, only referencing that a room should be sealed with no definition of “sealed.”. This lack of definition can create many arguments during construction as to what is considered “sealed.” Other guidelines require smoke pencil testing, but even that is difficult to quantify since interpretation is highly subjective of the person doing the testing. To avoid conflicts, best practice is to specify a combination of testing, including smoke pencil or soap bubble testing followed with a leakage test. The leakage test should be specific, easily quantifiable, allow the contractor to be held liable for achieving, and easily repeatable for future testing. This report analyzes case studies to provide a suggested definitive test criterion that can be applied to newly constructed CL3 facilities that have been constructed using common CL3 construction techniques such as drywall or concrete masonry unit.

Prescriptive requirements from guidelines include a fixed rate range between 2 and 20 l/s (4 and 42CFM) at a differential pressure of 200 Pa (0.8 inches of wc) as published by Australia/New Zealand (AS/NZS); a limit on leakage per room surface area of 0.152 l/s per square meter (0.030 cfm per square foot) at a room differential of 300 Pa (1.2 inches of wc) specified for USDA CL3 greenhouses; and a limit on leakage per room surface area of 0.036 l/s per square meter (0.007 cfm per square foot) at a room differential of 250Pa (1 inch of wc) listed in the German VDI guidelines. Merrick best practice typically specifies a leakage rate of 2% of room volume per minute at a room differential of 500 Pa (2 inches of wc). All these identified criteria were compared with various testing results obtained from facilities around the world. To make comparisons simpler, all rates were adjusted to an equivalent test with a room differential pressure of 250 Pa (1 inch of wc).

If all the rooms have the same construction materials and techniques, it is difficult to imagine why a room that is twice the volume must be twice as tight in construction to achieve an acceptable result as specified by the AS/NZS guideline. This fact creates an argument for improved testing methodologies that consider the room geometry.

The ARS greenhouse leakage rate acceptance criteria of 0.139 l/s per square meter (0.027 cfm per square foot) of room surface area at a room differential pressure of 250 Pa (1 inch of wc) seems to provide a room integrity test standard that is challenging, but at the same time achievable. Fifty-five percent of the rooms tested pass the criteria with typical CL3 construction methodologies, including single and double wall gypsum, as well as CMU.

The Merrick industry best practice leakage rate of 1.41% of room volume at a room differential pressure of 250 Pa (1 inch of wc) appears to offer similar challenging but achievable test results with 59% of rooms tested passing the criteria.

The recommended procedure for testing a room is a two-step process; 1) Initial leaks are identified with smoke pencil or soap bubble testing and repairs are made to seal all visible leaks, and 2) A quantifiable leakage rate to verify the room meets minimum leakage requirements.

Based on multiple facility testing assessments, the USDA ARS greenhouse leakage rate acceptance criterion of 0.139 l/s per square meter (0.027 cfm per square foot) of room surface area at a room differential pressure of 250 Pa (1 inch of wc) is recommended as the base criteria. Merrick best practice leakage rate of 1.41% of room volume at a room differential pressure of 250 Pa (1 inch of wc) is offered as a secondary or alternative test criterion. Finally, for rooms that form the primary containment boundary but are still classified as CL-3, we recommend a more stringent criterion following the German VDI guidelines of 0.036 l/s per square meter (0.007 cfm per square foot) or room surface area at a room differential pressure of 250Pa (1 inch of wc). Achieving pressure differentials in excess of 250Pa can sometimes challenge construction materials, so the final recommendations summarize the room integrity test requirements over a range of room pressure differentials.

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LIST OF ACRONYMS AND ABBREVIATIONS

ACDP-3: Advisory Committee on Dangerous Pathogens Level 3

ACDP-4: Advisory Committee on Dangerous Pathogens Level 4

ASHRAE:

AS/NZS: Australian / New Zealand Standards

ARS: Agricultural Research Service

BSL-3: Biosafety Level 3

BSL-3Ag: Biosafety Level 3 Agricultural

cfm: Cubic Feet per Minute

CBS: Canadian Biosafety Standard

CDC: Centers for Disease Control and Prevention

CL-3: Containment Level 3

CL-4: Containment Level 4

cu.m: Cubic meters

HSE: Health and Safety Executive

l/s: Liters per second

NIH: National Institutes of Health

Pa: Pascals

PC3: Physical Containment Level 3

PC4: Physical Containment Level 4

SAPO-3: Specified Animal Pathogens Order Level 3

SAPO-4: Specified Animal Pathogens Order Level 4

SOP: Standard Operating Procedure

sq.m: Square meters

UK: United Kingdom

USDA: United States Department of Agriculture

USDHHS: United States Department of Health and Human Services

VDI: Verein Deutscher Ingenieure (Association of German Engineers)

wc: Water Column

1 INTRODUCTION

1.1 BACKGROUND:

Biological research, diagnostic and/ bio-response facilities working on infectious agents in which laboratories and rooms have the potential for exposure to biological organisms are often required to have some level of boundary integrity testing (also commonly referred to as leak/air tightness testing). This paper is not intended to be adopted by guidelines or standards, but is a review of current guidelines and standards in the industry, and proposes a means of testing new construction of CL3 laboratories to ensure that an acceptable level of quality is achieved to meet the primary objectives of a CL3 containment envelope.

Boundary integrity testing of the laboratory space for biological containment facilities is performed in order to achieve three primary objectives

1. To minimize the potential for release of an aerosolized infectious agent to the general public and staff, as well as minimize cross-contamination of multiple agent use facilities or larger facilities that may have different program requirements within the same biological containment facility.

2. To minimize the migration of fumigants (decontamination gases) used to decontaminate the laboratory into neighboring spaces within the building thereby reducing fumigant concentrations within the space and creating a health hazard, as well as minimize migration of fumigants to multiple agent use facilities, or larger facilities that may have different concurrent program requirements.

3. To verify construction quality of the space and whether it is sufficient for withstanding the maximum design pressure limit without compromising the structural integrity of the CL3 containment boundary.

For CL3-Ag (BSL-3Ag, SAPO-4) and CL4 (BSL-4, ACDP-4) spaces, industry accepted standards have been pressure decay tests, as identified in the Canadian Biosafety Standard, 2nd Edition and the USDA ARS 242.1-2012 Facility Design Standards. As these standards and guidelines are well defined, this report will focus on boundary integrity testing methods for CL3 (BSL-3, SAPO-3, ACDP-3) laboratories. For the purposes of this report, the acronym for containment level 3 laboratories (CL3) will be used. Note that this acronym is directly comparable to other international acronyms (BSL-3/ABSL-3, ACDP-3, SAPO-3, PC3, etc.).

Typically, laboratory ventilation systems maintain airflow into a contaminated zone and exhaust through HEPA filters to minimize the risk of release of aerosolized agents. Most manipulations of infectious agents within a CL3 laboratory occur within primary containment devices such as Biological Safety Cabinets. The safeguards and practices minimize the risk of an accidental release of biological agents, thus migration of fumigants (decontamination gases) is of greater concern as room fumigation is usually performed when the ventilation system is isolated at the room or zone level.

To minimize leakage rate, CL3 laboratories are sealed using various methods during construction, and known leakage points, such as doors and grilles, are sealed manually before room fumigation. Testing strategies and acceptance criteria for room leakage are often subjective rather than definitive. This report analyzes different testing methodologies and case studies to provide a suggested definitive test criterion that can be applied to newly constructed CL3 facilities using common CL3 construction techniques such as gypsum board and concrete masonry unit (CMU).

1.2 GUIDELINES AND STANDARDS

The only definitive testing criteria for CL3 spaces from the guidelines and standards reviewed are from the Australia/New Zealand standards, and the German VDI standards. Requirements from other standards and guidelines are included for reference. Where applicable we have taken criteria defined within the guidelines and converted it to an equivalent range of acceptance at a room differential pressure of 250 Pa (1 inch of wc) to allow for a cross guideline comparison.

1.2.1 AUSTRALIAN AS/NZS 2243.3 – 2010 - SAFETY IN LABORATORIES - PART 3: MICROBIOLOGICAL SAFETY AND CONTAINMENT

Appendix H of AS/NZS 2243.3 “Recommendations on Acceptable Room Airtightness,” states a prescriptive recommendation for maximum air leakage: “The recommended maximum leakage rate, β, for PC3 and PC4 laboratories is 10^{-5}, at a test pressure of 200 Pa”, or in other words:

$Q = 10^{-5}$ m3/Pa·s * 200 Pa = 0.002m3/s,

which equates to a recommended maximum leakage rate of 2 l/s at a pressure differential of 200 Pa (4.2 cfm @ 0.8 inch wc).

AS/NZS 2243.3 also makes some exceptions as follows:

1. “Many PC3 and PC4 research laboratories do not need to meet the same level of air tightness as they are not dealing with animals and all work is performed in biological safety cabinets that act as the primary containment device within the laboratory structure.”

2. “It is not recommended that a laboratory be designed for gaseous decontamination if the leakage rate exceeds 10^{-4} at 200 Pa differential pressure, without specialist advice.”

Given these two exceptions, the 10^{-5} criteria is the AS/NZS standard, however a higher leakage rate may be acceptable for rooms that are not the primary containment barriers, as any manipulation of agents is performed in primary containment devices such as isolators or biological safety cabinets. Rooms may be permitted to have a leakage rate as high as 10^{-4} (20 l/s) at 200 Pa differential pressure (42 cfm at 0.8 inches of wc) if precautions are taken during decontamination to ensure that risks of exposure to fumigants are minimized in adjacent spaces.

Therefore, per the Australia/New Zealand standard, the recommended range of leakage from a CL3 laboratory is 2 l/s (4.2 cfm) to 20 l/s (42 cfm), with a room differential pressure of 200 Pa (0.8-inch wc). For comparison purposes in this report, the equivalent range of acceptance at a room differential pressure of 250 Pa is calculated at 2.2 l/s (4.7 cfm) to 22.4 l/s (47.5 cfm).

An argument is made that the Australia/New Zealand standard requires a single leakage rate for all room sizes to force designers to use smaller rooms which makes the facility safer. Another argument is that if they were to allow more leakage in a larger space, the result may be unacceptable leakage at one point causing a safety hazard during fumigations.

Note that this guideline is currently under review for a future update but unlikely to change in this regard.

1.2.2 VDI GUIDELINES TIGHTNESS OF CONTAINMENTS VDI-2083 PART 19 – 2018 - GERMANY

Criteria for testing of leakage is recommended for multiple building types including cleanrooms, for different classifications of containment spaces ranging from level 0 through level 7, For CL3, the guideline recommends that the leakage rate for class 4 be used at a rate of 0.03620L/s per square meter (0.007CFM per square foot) of room surface area at a pressure differential of 250Pa (1 inch w.c.). Also, for CL4 spaces, the guideline recommends that the leakage rate for class 5 be used at a rate of 0.01205L/s per square meter (0.002CFM per square foot) at a pressure differential of 250Pa (1 inch w.c.). Figure 1.1 shows the equivalency table from the guideline for acceptable air permeability rates of different classes at multiple differential test pressures.

Class [a]			Test/reference pressure in Pa							
			10	50	100	250	500	1000	2000	5000
			Air permeability $q_{V,\,Leck,\,spez,\,\Delta p}$ in $\ell/(m^2 \cdot s)$							
0			0,36181	1,02995	1,61616	2,93188	4,60061	7,21913	11,32803	20,55017
1	A	ATC5	0,12060	0,34332	0,53872	0,97729	1,53354	2,40638	3,77601	6,85006
2	B	ATC4	0,04020	0,11444	0,17957	0,32576	0,51118	0,80213	1,25867	2,28335
3	C	ATC3	0,01340	0,03815	0,05986	0,10859	0,17039	0,26738	0,41956	0,76112
4	D	ATC2	0,00447	0,01272	0,01995	0,03620	0,05680	0,08913	0,13985	0,25371
5		ATC1	0,00149	0,00423	0,00664	0,01205	0,01891	0,02968	0,04657	0,08448
6			0,00050	0,00141	0,00221	0,00402	0,00630	0,00989	0,01552	0,02816
7			0,00017	0,00047	0,00074	0,00134	0,00210	0,00330	0,00517	0,00939

Figure 1.1: VDI Guidelines Equivalency Table

1.2.3 CANADIAN BIOSAFETY STANDARD (CBS) – 2ND EDITION – 2015

Paragraph 5.2.12 in the CBS identifies minimum testing requirements of containment boundaries for select CL2 and CL2-Ag zones and all CL3-CL4 zones as follows: “Integrity of the seals of containment barrier penetrations, animal cubicle penetrations, and post mortem room (PM room) penetrations to be tested with a smoke pencil or other aid that does not influence the direction of airflow.”

Paragraph 5.3.5 in the CBS identifies the testing requirements for CL-3Ag and CL4 zones as follows: “Integrity of containment barrier to be tested by pressure decay testing. Acceptance criteria include two consecutive tests with a maximum of 250 Pa (1 inch wc) loss of pressure from an initial 500 Pa (2 inch wc) over a 20-minute period.”

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.4 USDHHS/CDC/NIH – BIOSAFETY IN MICROBIOLOGICAL AND BIOMEDICAL LABORATORIES – 5TH EDITION – 2009, USA (BMBL)

Published by the United States Department of Health and Human Services, the Centers for Disease Control and Prevention, and the National Institutes of Health, the BMBL is considered by many to be an international standard for biological containment facilities. However, for CL3 boundary integrity testing, it simply states, “Seams, floors, walls, and ceiling surfaces should be sealed. Spaces around doors and ventilation openings should be capable of being sealed to facilitate space decontamination.”

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.5 ARS 242.1 – 2012 – ARS FACILITIES DESIGN STANDARDS, USA

Criteria for testing of leakage is only provided for BSL3-Ag zones and is identical to the test identified in the Canadian Biosafety Standard. Acceptance criteria is outlined in paragraph 9B-4.E as, "Two consecutive pressure decay tests demonstrating a minimum of 1 inch wc (250 Pa) negative differential pressure remaining after 20 minutes, from an initial negative pressure differential of 2 inches wc (500 Pa)."

In addition, CL3-Ag (BSL-3Ag) containment for greenhouses is separately identified in paragraph 9B-6 as "the test pressure difference will be 6.24 pounds per square foot positive static pressure (300 Pa); the allowable leakage rate is 0.03 cfm per square foot" (0.152 l/s per square meter). For comparison purposes within this report, the equivalent allowable leakage rate at a room differential pressure of 250 Pa is 0.139 l/s per square meter (0.027 cfm per square foot) of room surface area.

There is no guidance on an acceptable leakage rate for CL3 spaces.

Note that this guideline is currently under review for a future update.

1.2.6 WORLD HEALTH ORGANIZATION (WHO) – LABORATORY BIOSAFETY MANUAL – 3RD EDITION – 2004

The WHO's manual is used in countries around the world, most often for developing countries that have no standard of their own to follow. For boundary integrity testing of CL3 facilities, the manual simply states: "All penetrations in laboratory sealed or sealable for decontamination."

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.7 ANSI/ASSP Z9.14 – 2014, USA

The American National Standard Testing has published a recent standard, "Testing and Performance-Verification Methodologies for Ventilation Systems for Biosafety Level 3 (BSL-3) and Animal Biosafety Level 3 (ABSL-3) Facilities."

Section 8.4.8.2 states, "When required by the facility risk assessment, room tightness (room air-leakage test) shall be performed before initial operation, periodically thereafter (as determined by the facility risk assessment and SOP) …" While no definitive testing criteria is stated, it does identify that the boundary is to be confirmed using smoke or soap bubbles with room pressures ranging from normal operating pressures to higher values (e.g., twice the normal value) as appropriate. It also indicates that additional test methods may be appropriate for initial commissioning of new construction and/or renovations, such as:

1. Operating the room at higher differential pressure values for the smoke/bubble tests (e.g., up to 2 inch wg (500 Pa)): Testing at this pressure may be useful in conjunction with HVAC controls testing.

2. "Room porosity" tests using airflow/pressure testing equipment similar to equipment used for duct-leak testing.

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.8 HEALTH AND SAFETY EXECUTIVE, UK

The HSE publishes a document, “Sealability of Microbiological Containment Level 3 and 4 Facilities,” which requires that, for containment level CL3 and CL4 facilities, the workplace is to be sealable to permit disinfection. The sealability test methods identified include:

1. Smoke testing is the main method, with no test criteria identified. They also go on to state, “Recent research has indicated that the use of smoke plume-generating devices is an inherently sensitive method. However, the limits of its sensitivity are heavily dependent upon the skill of the person performing the test.”

2. Room pressure decay testing, which requires the room to be held at a specified negative pressure relative to atmosphere for a prescribed period and measurement of any loss of the pressure differential at regular intervals during that time. The leak rate must then be compared to a predetermined acceptance value. A reference to Canada’s pressure decay testing methodology is also made.

In addition, there is a pointed note that applies to all responsible owners that deal with room disinfection that states, “Therefore, regardless of whether the fumigant of choice is formaldehyde, hydrogen peroxide or some other chemical, the legal requirement for room sealability remains and it is the responsibility of the duty holder to ensure the sealable status of the facility.”

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.9 ADVISORY COMMITTEE ON DANGEROUS PATHOGENS (ACDP) – 2018, UK

The HSE publishes a document, “Management and Operation of Microbiological Containment Laboratories” which identify the following requirements.

1. [28] “At CL3, the laboratory must be capable of being sealed to allow it to be effectively disinfected. An ongoing programme of formal assessment, e.g. an annual test is recommended to make sure sealability is maintained. However, more frequent visual inspections should be undertaken, e.g. for cracks, or dust trails, which may provide early indication of breaches in sealability of the facility. In this event, remedial work should be carried out and verified as effective.”

2. [31] “Sealability is usually carried out with the laboratory operating at normal working pressure and leaks are detected by observing any deviation of the smoke plume (see Figure 13) and should be performed by a competent person.”

There is no guidance on an acceptable leakage rate for CL3 spaces.

1.2.10 MERRICK INDUSTRY BEST PRACTICE

Where CL3 leakage criteria is not defined by local standards or guidelines, through coordination with biosafety professionals Merrick has often specified an acceptable leakage rate of 2% of room air volume per minute at a room differential pressure of 500 Pa (2 inches wc). For comparison purposes within this report, the equivalent allowable leakage rate at a room differential pressure of 250 Pa (1 inch of wc) is 1.41% of room volume per minute.

1.2.11 PRIMARY CONTAINMENT DEVICE – BSC

Though the CL3 space is typically considered a secondary containment boundary, it is worth discussing the boundary test criteria for a primary containment device—such as a BSC—to show the correlation between the two boundary types.

Under NSF 49-2016, section 6.2 the following criteria:

1. “The cabinet shall hold 2 in. wg (500 Pa) within ± 10% for 10 min.

2. For manufacturer testing only, the soap bubble method may be used when pressure plates fail: all welds, gaskets, penetrations, or seals on exterior surfaces of air plenums shall be free of soap bubbles when at 2 in. wg (500 Pa) ± 10% pressure above atmospheric.

1.2.12 BUILDING ENVELOPE TESTING

As a point of reference and comparison, there are building envelope testing standards that define the air tightness of the building enclosure or envelope. While these standards set air leakage criteria to help reduce overall energy consumption of buildings, they may not be appropriate for a containment lab environment. Three typical standards with test criteria are as follows:

1. The U.S. Army Corps of Engineers Air Leakage Test Protocol for Building Envelopes; Version 3 identifies a maximum allowable air leakage requirement at 75 Pa (0.3" wc) to be 1.27 l/s per square meter (0.25 cfm per square foot).

2. In the UK, the Air Tightness Testing & Measurement Association (ATTMA) Technical Standard L2 lists normal and best practice air permeability rates for various space types. At 50 Pa (0.2" wc), the most stringent air permeability best practice is listed for Museums as 1.0 cubic meter per hour per square meter or equivalently: 0.278 l/s per square meter (0.055 cfm per square foot).

3. ASHRAE 90.1 – Energy Standards for Buildings Except Low Rise Residential Buildings identifies many leakage rates of building envelopes for different types of building construction and fenestration types, with the one relevant for our purpose being 2.03 l/s per square meter (0.4 cfm per square foot) at a pressure of 75 Pa (0.3" wc)

1.2.13 COMPARISONS

The graphic below illustrates the wide variance in test criteria. Note that the only defined test criteria for CL3 laboratories is the AS/NZS guidelines and German VDI. ASHRAE 90.1 and the NSF/ANSI 49 standards are not relevant to CL3 construction but are shown to illustrate extreme limits.

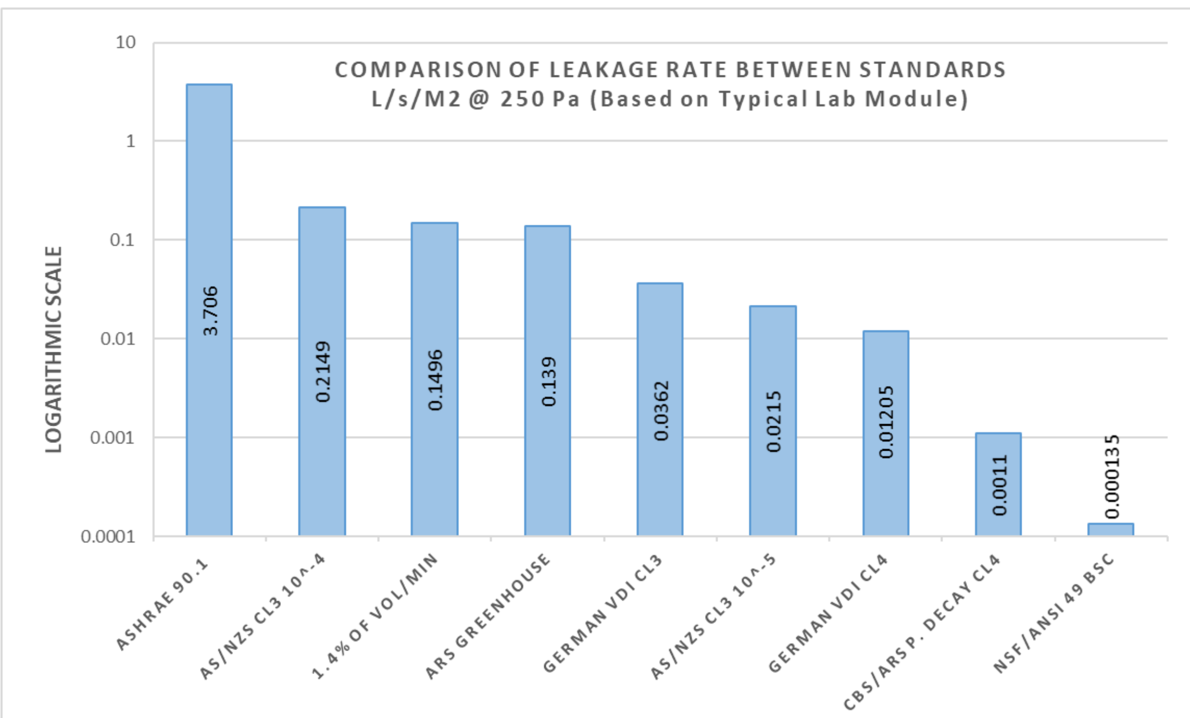

Figure 1.2: Logarithmic comparison of acceptable leakage rate limits between standards normalized to a dP of 250Pa; Based on a typical lab module of 6.7m(L) x 3.3m(W) x 3.0m(H)

2 WHAT IS AN ACCEPTABLE LEAKAGE CRITERION?

2.1 CONTAINING AEROSOLS

CL3 laboratories typically rely on directional airflow to contain potential infectious agents, by supplying less air into the space than what is being exhausted (extracted) from the space. The USDA ARS 242.2 guidelines recommend *"the infiltration of air into the containment boundary be at least 50 cfm (23.6 l/s) per doorway at all times."* The USDA ARS 242.2 guideline also requires *"sufficient exhaust to create a 0.05" water column differential between containment area and the access area"*, which typically drives the airflow across a doorway to be greater than 47l/s (100CFM). The NIH Design Requirements Manual states *"to maintain 47 l/s (100 cfm) air flow from the corridor into each lab module."* To ensure directional airflow is maintained, at a minimum the leakage of the space should be less than allowed for across the doorways. Arguably, increasing the differential between supply and exhaust (extract) air will maintain directional airflow in the leakiest of spaces, so a hard target based on this criterion is difficult to establish. Generally, CL3 spaces are not primary containment, and work with aerosols or animals are within self-contained apparatus, so risk of aerosols within the general CL3 environment is minimal. A risk assessment should be completed to evaluate if there is a considerable risk in which a higher level of containment is required.

2.2 CONTAINING FUMIGANT (DECONTAMINATION GASES)

The applications of expensive welded metal enclosures or monolithic concrete is not required to achieve the desired results for CL3 applications. Many construction techniques can successfully contain fumigants during a room decontamination process, often with the use of controlled pressure differentials to adjacent spaces. Standard stud and drywall construction—with appropriately sealed joints at floors, ceilings, service openings, and door frames—has been successful. Fumigation SOPs need to be considered to ensure that fumigation concentration is maintained, and risk of exposure to adjacent spaces is minimized. The room, or rooms, being decontaminated are usually isolated from the rest of the facility and the building ventilation system, however consideration should be given to extracting a very small amount of air from the space during decontamination to maintain the room at a negative pressure compared to adjacent spaces. Alternatively, adjacent spaces may be slightly positively pressurized.

2.3 CONSTRUCTION QUALITY

Many guidelines only reference that a room should be sealed with no definition of "sealed." This lack of definition can create many arguments during construction as to what is considered sealed. Other guidelines require smoke pencil testing, but even that is difficult to quantify since interpretation is highly subjective of the person doing the testing.

Some guidelines have more prescriptive requirements, such as those for Australia/New Zealand, which specify a fixed rate; Canada and the USDA which specify a volumetric leakage rate for CL3Ag; the USDA greenhouse requirements, which specify a limit on leakage per surface area; and the German VDI guidelines which specify a limit on leakage per surface area.

To avoid conflicts during construction and with testing officials, best practice is to specify a combination of testing, including smoke pencil or soap bubble testing followed with a leakage test. The leakage test should be specific, easily quantifiable, allow the contractor to be held liable for achieving, and easily repeatable in future testing.

Since aerosol and fumigant (decontamination gases) containment can be accomplished with good SOPs, an acceptable leakage criterion for CL3 laboratories does not need to be extreme but should be achievable and repeatable with current construction practices for CL3 laboratories, which often consist of gypsum board or CMU.

2.4 INTEGRITY TEST DESCRIPTIONS

Facility leakage tests and pressure decay tests are similar tests that measure the amount of air that escapes or is drawn into a space under pressure. One test measures the amount of air leaking into or out of a room to maintain a pressure, while the other records pressure changes as the amount of air in the room changes due to pressurization and flow through the leak points. For practical purposes, airflows less than 2 l/s (4 cfm) are difficult to accurately measure and control, while for most CL3 rooms air leakage greater than 2 l/s (4 cfm) result in a pressure decay rate that is relatively short at less than a few seconds. The sensitivity of instruments used for testing has to be taken into consideration to ensure the expected leakage rates can be accurately measured.

From the Australia/New Zealand standards, we see a range of acceptable leakage is suggested at between 2 to 20 l/s (4 and 42 cfm) when the room is subjected to a differential pressure of 200 Pa (0.8 inches of wc). However, an acceptable room volume or surface area through which the leakage would occur is not provided. Thus, for the Australia/New Zealand standard, the size of the room is irrelevant in the calculation, and the same leakage rage is assumed for any size of room. We suggest that a small procedure room should not have the same leakage rate as a large open laboratory, when the large laboratory has significantly more surface area, penetrations, and other potential leak points all of which contribute to overall leakage. Similar to the pressure decay testing methodology, we suggest that a leakage standard for CL3 spaces should be proportional to the room size.

When looking at a pressure decay testing specified for CL3-Ag and CL4 facilities, the same rate of change in pressure is required for any size room. From the ideal gas law, the change in the amount of air in the space is proportional to the room volume, thus the larger the room, the larger the amount of displaced air is required to meet the same change in pressure. In other words, for the pressure decay test criteria, the rate of leakage from a space is directly proportional to the room volume.

$$V=n \cdot R \cdot T/P$$

Room geometry can also play a factor in leakage rates. Consider two rooms with the same width, with one room having a square floor area and the second room having rectangular area where the length of one side is twice the length of the width of the other side. Although both rooms hove double the volume of air, the surface area of the rectangular room is marginally less than double of the surface area of the square room.

$A_{sq} = 2*[(w \cdot h)+(l \cdot h)+(l \cdot w)]$, where l=w $\qquad$ $A_{rect} = 2*[(w \cdot h)+(l \cdot h)+(l \cdot w)]$, where l=2w

$A_{sq} = 2*[(w \cdot h)+(w \cdot h)+(w \cdot w)]$ $\qquad$ $A_{rect} = 2*[(w \cdot h)+(2w \cdot h)+(2w \cdot w)]$

$A_{sq} = 4wh+2w^2$ $\qquad$ $A_{rect} = 6wh+4w^2$

This observation suggests that acceptable rates of air leakage from containment spaces should be proportional to the room surface area, rather than the room volume, however it is noted that the difference in room volumes vs the difference in surface areas between the two geometries is marginal and would have minimal consequences on calculating an acceptable leakage rate.

2.5 RATE VS. VOLUME VS. AREA

Some simple examples to demonstrate the differences between the three established measurement criteria are identified as follows:

1. A typical lab module of 6.7m(L) x 3.3m(W) x 3.0m(H).
2. A double wide lab module of 6.7m(L) x 6.7m(W) x 3.0m(H) is twice as wide as a typical lab module.
3. A double long lab module of 13.4m(L) x 3.3m(W) x 3.0m(H), is twice as long as a typical lab module.

Each of these examples is used to demonstrate the comparable leakage rates. The surface areas and volumes for each of the rooms is summarized as follows:

Room #	Room Name	Volume cu.m	Surf. Area sq.m
1	Typical	66.33	104.22
2	Double Wide	134.67	170.18
3	Double Long	132.66	188.64

Figure 2.1: Area and Volume Calculations

2.5.1 FIXED FLOW RATE: AUSTRALIAN AS/NZS 2243.3

The AS/NZS standard requires leakage of any size space to be between 2.2 and 22.4 l/s (4.7 and 47 cfm) when subjected to a room differential pressure of 250 Pa (1 inch of wc). Figure 2.2 below indicates the minimum fixed leakage of 2.2 l/s (4.7cfm) for lab modules of different geometry and compares it to the equivalent leakage per unit area, leakage of %volume per minute, and an equivalent pressure decay rate. From the figure, we see that comparable leakage per unit area and leakage of percent volume per minute are essentially double for a typical lab module that is half the size of the other two lab modules. Comparable pressure decay rates for the typical lab module is approximately half of that compared to the other two lab modules. If all the lab modules have the same construction materials and techniques, then it is difficult to imagine why a room that is twice the volume must be twice as tight in construction to achieve an acceptable result. This demonstrates that a fixed leakage rate may not be the best solution as a measure of acceptable leakage as compared to guidelines which factor in room geometry.

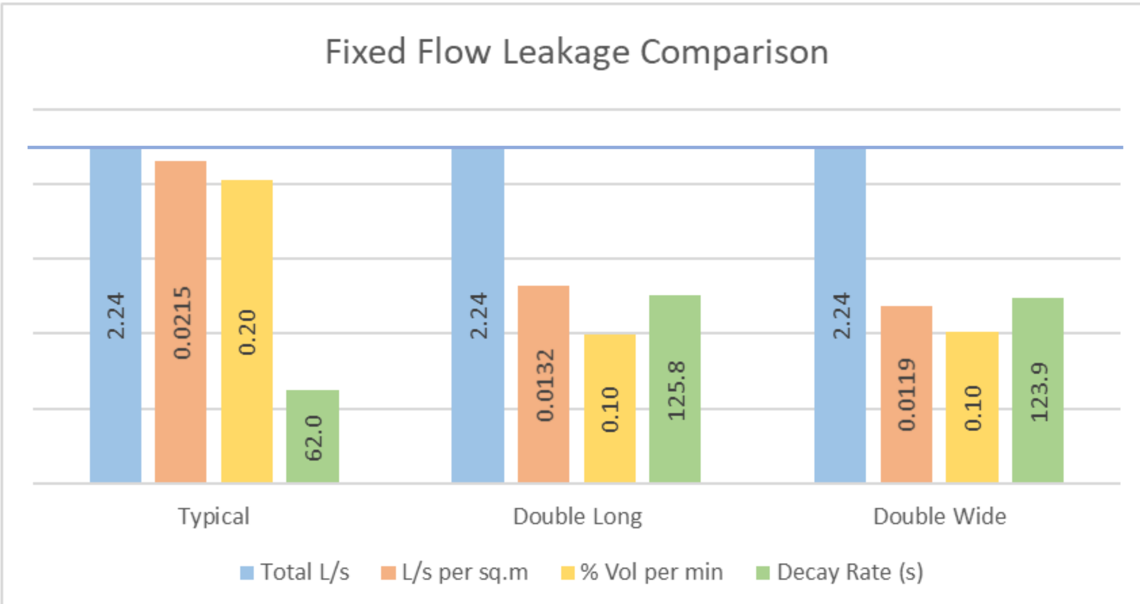

Figure 2.2: Fixed Flow Rate: AS/NZS Minimum Leakage Comparison

2.5.2 FLOW PER UNIT AREA: VDI-2083 PART 19 AND ARS 242.1

The German VDI guideline requirement states a leakage per room surface area criteria of 0.0362 l/s per square meter (0.007 cfm per square foot) at a room differential pressure of 250 Pa (1 inch of wc).The ARS greenhouse requirement states a leakage per room surface area criteria of 0.139 l/s per square meter (0.027cfm per square foot) at a room differential pressure of 250 Pa (1 inch of wc). Figure 2.3 below indicates the ARS fixed leakage per room surface area criteria of 0.139 l/s per square meter (0.027cfm per square foot) at a room differential pressure of 250 Pa (1 inch of wc) for lab modules of different geometry and compares it to the equivalent total leakage flow rate, leakage of %volume per minute, and an equivalent pressure decay rate. From the chart, we see that total leakage increases as surface area increases, as would be expected when using the same construction materials and techniques. The chart also shows that the comparable percent volume per minute calculation and pressure decay rate only vary slightly as the room geometry changes.

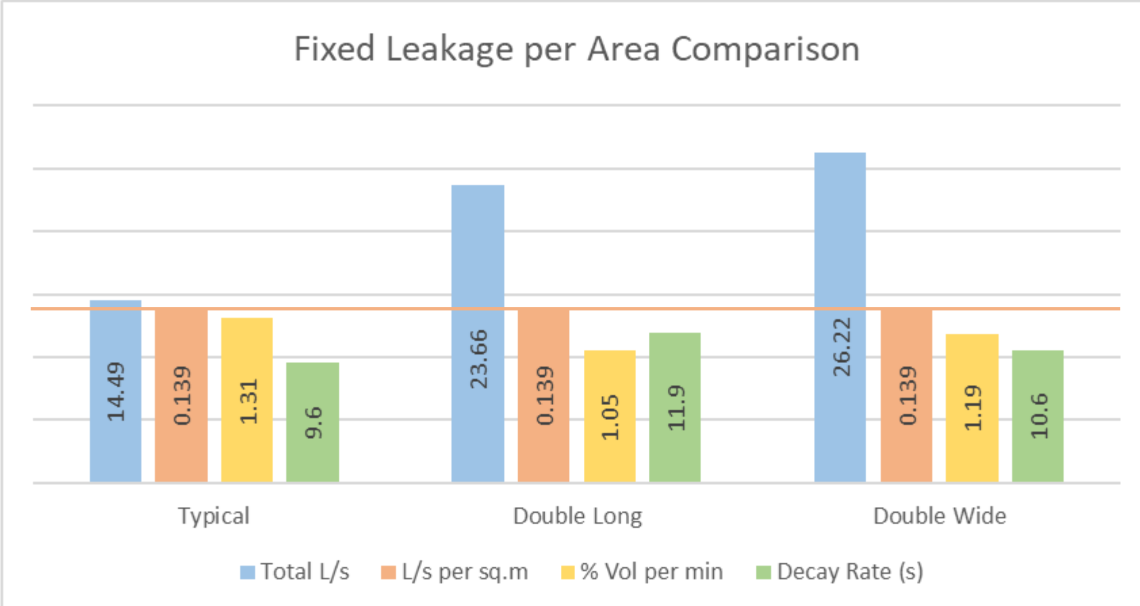

Figure 2.3: Fixed Leakage per Area: ARS Greenhouse Leakage Comparison

2.5.3 FLOW AS % VOLUME PER MINUTE

For CL3 biocontainment construction Merrick typically specifies 1.41% of room volume per minute as an acceptable leakage rate with a room differential pressure of 250 Pa (1 inch of wc). From the tables, we see that total leakage increases as volume increases, as would be expected when using the same construction materials and techniques. As previously suggested, we also see that the comparable leakage per surface area calculation vary slightly as the room geometry changes.

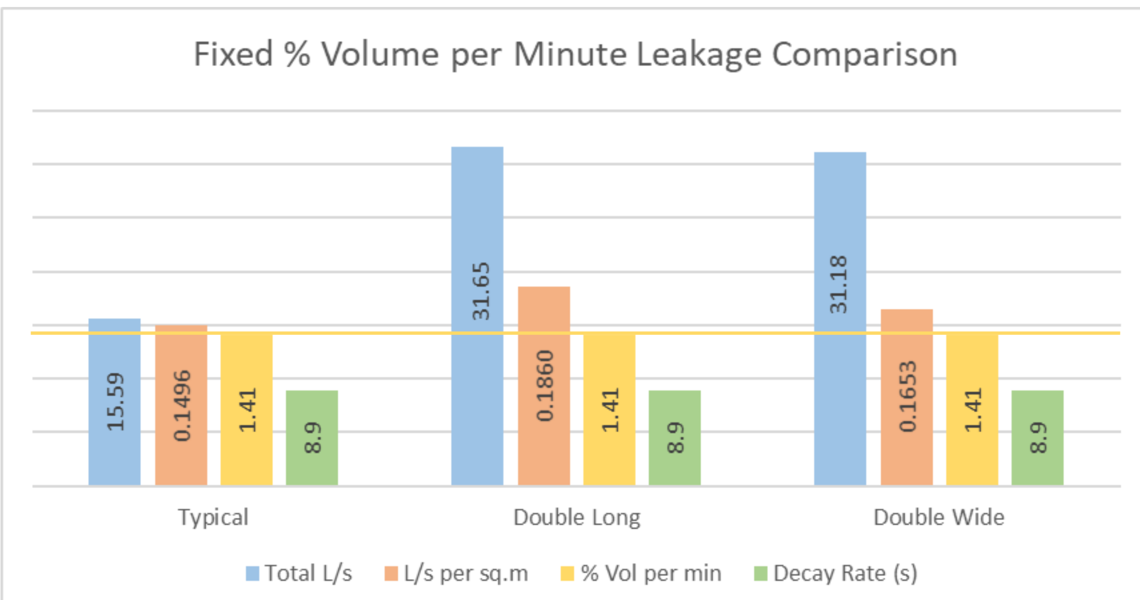

Figure 2.4: Industry Percent of Room Volume Leakage Comparison

2.5.4 PRESSURE DECAY: CANADIAN BIOSAFETY STANDARD (CBS) – ARS 242.1

Although extreme for CL3, the CL3-Ag and CL4 pressure decay test criteria identified in the CBS and ARS guidelines is something worth considering for primary containment spaces. The guidelines require a pressure decay rate exceeding 20 minutes from a pressure of 500 Pa to 250 Pa (2 to 1 inch of wc). Figure 2.4 shows how flow as %volume per minute, and an equivalent pressure decay rate are directly related.

3 QUANTIFIABLE TESTING RESULTS

3.1 INTRODUCTION

The following examples are of real CL3 laboratories around the world where we have compiled quantifiable leak testing results. The results are compared to different guideline testing requirements of the AS/NZS total leakage of 22.4L/s, the ARS Greenhouse standard of 0.139 l/s per square meter of room surface area, and the biocontainment industry standard of 1.41% of room volume per minute, all at a differential pressure of 250Pa.. Since testing at all facilities was at slightly different pressures, all data is modified using the orifice equations and ideal gas laws to equate comparable leakage rates at 250 Pa, and green highlights indicate the spaces that pass the criteria in the relevant column. For simplicity of reading, all units of measure are presented in metric.

3.2 AUSTRALIAN STUDY

In 2009, Gordon B. McGurk published "A Study of Air-tightness in Australian High-level Bio-containment Facilities" in the ABSA journal, Applied Biosafety. This study included leakage testing comparisons to the Australian standards for 18 different PC3 (CL3) facilities in Australia. Two facilities could not be successfully tested due to structural integrity. Tabulation of the results, including the construction material for each, from the 16 facilities are shown in Figure 3-1.

Only the wall surface area for each facility was recorded in the publication, so for comparison purposes in this report, the total surface area including floor and ceiling was calculated. To calculate the total surface area, the room volume was divided by a typical room height of 3 meters to get the room area. This room area was used as the floor and ceiling area and added to the wall area to achieve a calculated total room surface area.

We analyzed the 16 Australian facilities and compared accepted leakage rates based on the AS/NZS 2243.3 standard and calculated flow per square meter of room surface area, percent of room volume of leakage per minute at equivalent room differential pressures of 250 Pa, and pressure decay rate from 500 Pa to 250 Pa. Of the 16 facilities tested, only 10 met the minimum Australian criteria of 22.4 l/s when measured at a differential pressure of 250 Pa, and only two facilities achieved the leakage rate of 2.2 l/s.

Australia Facility #	Construction Material	Volume cu.m	Est Total S. Area sq.m	Leakage @ 250Pa (L/s) (Pass =22.4)	Leakage per Area (L/s per sq.m) (Pass = 0.139)	Leakage per Vol %vol per min (Pass = 1.41)	Decay Rate 500Pa to 250Pa (s)
1	Sandwich Panel	79.69	112	1.80	0.0160	0.14	103.6
2	Double Gypsum	42.2	77	1.86	0.0241	0.26	53.2
3	Double Gypsum	48.11	109	2.54	0.0234	0.32	44.4
4	Sandwich Panel	57.82	109	2.96	0.0273	0.31	45.7
5	Double Gypsum	48.11	109	4.51	0.0415	0.56	25.0
6	Gypsum	50.83	80	8.36	0.1041	0.99	14.2
7	Double Gypsum	143.07	158	16.12	0.1019	0.68	20.8
8	Sandwich Panel	61.97	110	16.47	0.1499	1.59	8.8
9	Sandwich Panel	67.35	105	17.71	0.1679	1.58	8.9
10	Gypsum	48.91	85	21.18	0.2494	2.60	5.4
11	Gypsum	82.8	115	26.48	0.2299	1.92	7.3
12	Gypsum	112.83	173	27.75	0.1604	1.48	9.5
13	Gypsum	42.04	76	28.17	0.3700	4.02	3.5
14	Gypsum	49.15	86	29.96	0.3503	3.66	3.8
15	Gypsum	66.86	99	35.03	0.3546	3.14	4.5
16	Gypsum	156.68	248	63.28	0.2548	2.42	5.8

Figure 3.1: Australian Study Comparison

In terms of l/s per square meter of room surface area, we see that the first nine facilities and facility 12 are all under 0.2 l/s per square meter at a pressure differential of 250Pa. This is expected because facility 12 has a large surface area about 60% greater than the other facilities that passed the AS/NZS standard. If we look at the ARS guideline with an acceptable leakage criterion of 0.139 l/s per square meter of room surface area at a differential pressure of 250Pa, we see that only the first seven facilities pass the requirement as indicated by the green highlight. We also note that the leakage rate from facility numbers 8,9, and 12 are almost identical between 0.15 and 0.17 l/s per square meter, while facility 10—which passed the AS/NZS criteria—grossly exceeds the ARS rate with a leakage of 0.25 l/s per square meter at a pressure differential of 250Pa. This data supports the suggestion that leakage per unit area is an improved acceptance criterion over a fixed rate for all spaces.

When looking at percentage of room volume per minute, again as indicated by the green highlight only the first seven facilities fall under the CL3 biocontainment industry standard recommended criteria of 1.41% of room volume per minute at a pressure differential of 250Pa. We also notice that facility 12's leakage rate of 1.48%vol per minute at a pressure differential of 250Pa is less than that of facilities 8,9 and 10, all of which passed the AS/NZS criteria. This data shows similar acceptance results when compared to the ARS greenhouse leakage rate of 0.139 l/s per square meter of room surface area at 250 Pa differential pressure, which again supports the suggestion that leakage per volume is an improved acceptance criterion over a fixed rate for all spaces.

We also note that the best constructed facility has a pressure decay rate from 500 Pa to 250 Pa of just over one and a half minutes, which is not even close to the 20 minutes identified in the CBS and ARS guidelines for CL3-Ag and CL4 facilities. This data supports that CL3 facilities are not currently held to the same construction standards as CL3-Ag and CL4.

3.3 US FACILITY 1

The construction of the walls of this facility was concrete masonry unit (CMU), and the testing criteria was originally specified as a pressure decay test, starting with the room at a negative differential pressure of 188 Pa, and measuring the time it took until the negative pressure differential reached 94 Pa. The passing criteria was an elapsed time greater than 20 minutes. During initial testing, the elapsed time for most of the rooms was less than 15 seconds.

Merrick recommended a different approach to measure the room volumetric offset to maintain a certain pressure. Leakage points were identified and sealed, and tests were re-run using a blower door to pressurize each room and measure the airflow required to maintain pressure.

We compared the final measured leakage rates and calculated flow per square meter of room surface area, and percent of room volume of leakage per minute at equivalent differential pressures of 250 Pa, and a pressure decay rate from 500 Pa to 250 Pa. From these results, we see that all the large labs fall under the Merrick acceptance leakage rate of 1.41% of room volume per minute at a pressure differential of 250Pa.

US Facility 1		Volume	Surf. Area	Leakage Measured @ 250Pa (L/s)	Leakage per Area L/s per sq.m	Leakage per Vol %vol per min	Decay Rate
Room #	Room Name	cu.m	sq.m	(Pass =22.4)	(Pass = 0.139)	(Pass = 1.41)	500Pa to 250Pa (s)
MB00.58	Large Lab	59.47	96.62	5.7	0.0587	0.57	21.9
MB00.59	Large Lab	59.47	96.62	13.3	0.1381	1.35	9.3
MB00.60	Large Lab	59.47	96.62	9.3	0.0967	0.94	13.3
MB00.61	Large Lab	59.47	96.62	9.3	0.0967	0.94	13.3
MB00.62	Large Lab	59.47	96.62	13.7	0.1415	1.38	9.1
MB00.63	Large Lab	59.47	96.62	9.7	0.1001	0.98	12.9
MB00.64	Large Lab	59.47	96.62	11.0	0.1139	1.11	11.3
MB00.66	Procedure Room	24.07	50.17	8.9	0.1775	2.22	5.7
MB00.67	Procedure Room	48.14	81.75	13.7	0.1673	1.70	7.4
MB00.73	Iso Suite	99.11	137.50	13.7	0.0995	0.83	15.2

Figure 3.2: US Facility 1 Comparison

If we look at the ARS guideline with an acceptable leakage criterion of 0.139 l/s per square meter of room surface area at a pressure differential of 250Pa, we see that all but one of the large labs pass the requirement. This data supports the suggestion that ARS greenhouse criteria is slightly more stringent than the Merrick best practice acceptance criteria.

If we compare the leakage rates to the AS/NZS acceptance criteria of 22.4 l/s at a room differential pressure of 250 Pa, we see that all the rooms easily pass the criteria. Again, this data supports the suggestion that leakage per unit area or % of volume per minute is an improved acceptance criterion over a fixed rate for all spaces.

3.4 US FACILITY 2

The construction of the walls of this facility was single layer drywall on metal stud. No specific testing criteria were identified for this project, however Merrick recommended performing multiple tests in succession in which leaks were identified and sealed and the room re-tested. Using the same approach, tests were run using a blower door to pressurize each room and measure the airflow required to maintain pressure.

We compared the final measured leakage rates and calculated flow per square meter of room surface area, and percent of room volume of leakage per minute at equivalent differential pressures of 250 Pa, and a pressure decay rate from 500 Pa to 250 Pa. From these results, we see that most of the larger labs fall above the ARS greenhouse requirement of 0.139 l/s per square meter or room surface area at a pressure differential of 250Pa and fall above the Merrick acceptance leakage rate of 1.41% of room volume per minute at a pressure differential of 250Pa. However, we note that the animal rooms, which are primary containment, all passed the listed criteria in the table.

US Facility 2		Volume	Surf. Area	Leakage Measured @ 250Pa (L/s) (Pass =22.4)	Leakage per Area L/s per sq.m (Pass = 0.139)	Leakage per Vol %vol per min (Pass = 1.41)	Decay Rate 500Pa to 250Pa (s)
Room #	Room Name	cu.m	sq.m				
8079	Large Lab	67.96	107.77	33.5	0.3108	2.96	4.2
8078	Large Lab	67.96	107.77	19.3	0.1795	1.71	7.4
8077	Large Lab	67.96	107.77	27.4	0.2539	2.42	5.2
8076	Large Lab	67.96	107.77	22.6	0.2101	2.00	6.3
8075	Large Lab	67.96	107.77	22.6	0.2101	2.00	6.3
8074	Large Lab	67.96	107.77	13.7	0.1269	1.21	10.4
8073	Large Lab	67.96	107.77	14.2	0.1313	1.25	10.1
8023E	Lab	45.31	78.04	9.4	0.1209	1.25	10.1
8024E	Lab	45.31	78.04	7.1	0.0907	0.94	13.4
8025E	Lab	45.31	78.04	7.1	0.0907	0.94	13.4
8026E	Lab	45.31	78.04	7.1	0.0907	0.94	13.4
8023F	Procedure Room	40.78	73.21	7.1	0.0966	1.04	12.1
8023G	Animal Room	73.62	109.63	7.1	0.0645	0.58	21.8
8024F	Procedure Room	40.78	73.21	7.1	0.0966	1.04	12.1
8024G	Animal Room	73.62	109.63	7.1	0.0645	0.58	21.8
8025F	Procedure Room	40.78	73.21	7.1	0.0966	1.04	12.1
8025G	Animal Room	73.62	109.63	7.1	0.0645	0.58	21.8
8026F	Procedure Room	40.78	73.21	7.1	0.0966	1.04	12.1
8026G	Animal Room	73.62	109.63	7.1	0.0645	0.58	21.8
8027E	Aerobiology	124.88	159.98	23.6	0.1474	1.13	11.1
8027I	Necropsy	39.64	70.61	23.6	0.3340	3.57	3.5
8027J	Animal Room	27.18	55.00	23.6	0.4288	5.21	2.4

Figure 3.3: US Facility 2 Comparison

If we compare the leakage rates to the AS/NZS acceptance criteria of 22.4 l/s at a room differential pressure of 250 Pa, we see that the larger rooms have about twice the air leakage as the smaller rooms. Again, this data supports the suggestion that leakage per unit area or volume is an improved acceptance criterion over a fixed rate for all spaces.

3.5 US FACILITY 3

The construction of the walls of this facility was precast concrete with a polyurethane sealant, and the testing criteria was specified as 1% of room volume per minute at a room differential pressure of 125 Pa. The facility was tested as a whole at the containment perimeter using a blower door to pressurize the space and measure the airflow required to maintain pressure.

We compared the final measured leakage rates and calculated flow per square meter of room surface area, and percent of room volume of leakage per minute at equivalent differential pressures of 250 Pa, and a pressure decay rate from 500 Pa to 250 Pa. From these results we see the facility falls well below the ARS greenhouse requirement of 0.139 l/s per square meter or room surface area at a pressure differential of 250Pa, and also falls well below the Merrick acceptance leakage rate of 1.41% of room volume per minute at a pressure differential of 250Pa. The facility also passes the German VDI recommendation of 0.036 l/s per square meter (0.007 cfm per square foot) of room surface area at a room differential of 250Pa (1 inch of wc). In contrast, due to the volume of the space the facility would have no hope of passing the AS/NZS requirement of 22.4 l/s at a room differential pressure of 250 Pa. Again, this data supports the suggestion that leakage per unit area or volume is an improved acceptance criterion over a fixed rate for all spaces.

US Facility 3		Volume	Surf. Area	Leakage Measured @ 250Pa (L/s) (Pass =22.4)	Leakage per Area L/s per sq.m (Pass = 0.139)	Leakage per Vol %vol per min (Pass = 1.41)	Decay Rate 500Pa to 250Pa (s)
Room #	Room Name	cu.m	sq.m				
101	Mechanical Space	8563	2742	73.4	0.0268	0.05	244.2

Figure 3.4: US Facility 3 Comparison

3.6 CANADIAN FACILITIES

The first Canadian facility listed here was tested on a room-by-room basis for each lab room, whereas the second Canadian facility was tested as a whole at the containment perimeter. Both the Canadian facilities' walls were constructed with double layer gypsum drywall and were tested using a pressure decay of each space from 500 Pa to 0 Pa, for the time required for the pressure to drop to 0 Pa. The starting and ending atmospheric and gauge pressures were known, along with total room volume and space temperature. These values were used to calculate a l/s (cfm) leakage rate normalized to room differential pressure of 250 Pa, flow per square meter of surface area, and percentage of room volume per minute.

We compared the final measured leakage rates, calculated flow per square meter of room surface area, percent of room volume of leakage per minute at equivalent differential pressures of 250 Pa, and pressure decay rate from 500 Pa to 250 Pa. From these results, we see that about half of the labs fall below the ARS greenhouse requirement of 0.139 l/s per square meter of room surface area at a pressure differential of 250Pa. We also see that the leakage rates for the same rooms also fall below the Merrick acceptance leakage rate of 1.41% of room volume per minute at a pressure differential of 250Pa.

Canadian Facilities		Volume	Surf. Area	Leakage Measured @ 250Pa (L/s)	Leakage per Area L/s per sq.m	Leakage per Vol %vol per min	Decay Rate
Room #	Room Name	cu.m	sq.m	(Pass =22.4)	(Pass = 0.139)	(Pass = 1.41)	500Pa to 250Pa (s)
20-097	Lab	141.6	189.1	286.85	1.5171	12.15	1.0
20-098	Procedure	45.1	79.3	13.87	0.1750	1.85	6.8
20-099	Lab	112.5	156.9	160.59	1.0237	8.57	1.5
20-106	Procedure	42.1	74.9	3.67	0.0490	0.52	24.0
20-107	Procedure	42.1	75.2	12.94	0.1722	1.85	6.8
20-108	Procedure	41.3	73.7	11.99	0.1626	1.74	7.2
20-109	Procedure	42.1	74.9	9.46	0.1263	1.35	9.3
20-110	Lab	96.5	153.5	12.12	0.0789	0.75	16.7
MM	Total Vol	289.9	352.7	23.32	0.0661	0.48	26.0

Figure 3.5: Canadian Facilities Comparison

If we compare the leakage rates to the AS/NZS acceptance criteria of 22.4 l/s at a differential pressure of 250Pa, we also see that most of the rooms passed the criteria. However, note the wide fluctuations in flow ranging from 5 l/s to over 22 l/s, which is almost directly related to the size of the room.

3.7 EUROPEAN FACILITY

The construction of the walls of this facility was cast-in-place concrete with an epoxy coating, and the specification for this facility was for a very tight rate specified at 0.0091 l/s per square meter of surface area at a test pressure of 200 Pa. Tests were run using a blower door to pressurize each room and measure the airflow required to maintain pressure.

The airflow leakage rate measured here is significantly tighter than other facilities and comparable to the CBS/ARS requirement of 20 minutes (1200 seconds) for a pressure decay from 500 Pa to 250 Pa. This is expected as the construction materials and techniques are like those applied to CL3-Ag and CL4 facilities.

European Facility #1		Volume	Surf. Area	Leakage Measured @ 250Pa (L/s)	Leakage per Area L/s per sq.m	Leakage per Vol %vol per min	Decay Rate
Room #	Room Name	cu.m	sq.m	(Pass =22.4)	(Pass = 0.139)	(Pass = 1.41)	500Pa to 250Pa (s)
GE07B	Animal	49.725	76.5	0.19	0.0025	0.02	547.5
GE08	Animal	126.425	194.5	0.27	0.0014	0.01	986.0
GE09	Animal	77.831	119.74	0.13	0.0011	0.01	1214.0
GE10	Animal	77.831	119.74	0.30	0.0025	0.02	539.6
GE11	Animal	77.831	119.74	0.31	0.0026	0.02	520.3
GE12	Animal	77.831	119.74	0.20	0.0017	0.02	809.3
GE13	Animal	96.98	149.2	0.32	0.0022	0.02	625.9
GE14	Animal	48.867	75.18	0.15	0.0019	0.02	703.6
GE16	Animal	81.38	125.2	0.15	0.0012	0.01	1171.7

Figure 3.6: European Facility Comparison

4 CONCLUSIONS

A fixed leakage rate for all facility sizes does not make sense since leakage is a function of surface area. So a room with a large surface are and large volume will inherently leak more than a smaller room constructed of the same materials and construction methods. A fixed leakage rate is comparable to saying a 3000kg truck and a 600kg car can both drive the same distance on 1 liter of gasoline. Although it may be possible, we recommend that improved testing methodologies consider the size of the room.

The rooms tested were tallied with pass rates identified for each of the three comparative leakage rates at a room differential of 250 Pa (1 inch w.c.). Data from US facility 3 and the European facility were ignored in this tabulation, since these facilities were constructed with higher standards and construction methodologies that exceed typical CL3 construction. The results are shown in Figure 4.1 below.

	Room Pass Rate at Room Differential Pressure of 250Pa				
Facility	AS/NZS (2.24L/s)	AS/NZS (22.4L/s)	ARS Greenhouse (0.139L/s per sq.m)	Room vol/min 1.41%	German VDI (0.036L/s per sq.m)
Australian	13%	63%	44%	44%	25%
US Facility #1	0%	100%	70%	80%	0%
US Facility #2	0%	68%	64%	68%	0%
Canada Facilities	0%	67%	44%	44%	0%
Average	3%	74%	55%	59%	6%

Figure 4.1: Leakage Pass Rate Comparisons Across Projects

Note that none of the above facilities in the US or Canada passed the German VDI standard, and yet all the facilities reported are acceptable to national biosafety regulations and have been certified to operate at CL3. Additional testing has also proven that fumigation of these spaces has been carried out without adverse effects to surrounding areas within the building. This suggests that the German VDI standard may be more stringent than is required for typical CL3 construction and should not be used to validate facilities constructed of drywall or CMU.

The ARS greenhouse leakage rate acceptance criteria of 0.139 l/s per square meter (0.027 cfm per square foot) or room surface area at a room differential pressure of 250 Pa (1 inch of wc) seems to provide a room integrity test standard that is challenging but achievable. Fifty-five percent of the rooms tested pass the criteria with typical CL3 construction methodologies, including single and double wall gypsum and CMU.

The Merrick best practice rate of 1.41% of room volume at a room differential pressure of 250 Pa appears to offer similar challenging but achievable test results with 59% of rooms tested passing the criteria with the same CL3 construction methodologies.

Although not all rooms tested pass the above suggested criteria, it is important to note that such requirements were not originally specified and thus there was no reason for the contractor to apply better quality control techniques to achieve improved performance. For example, if you look closely at the data for US facility 2, you see that two of the large labs passed all criteria, while five others of the same volume and surface area had almost twice the leakage and failed all criteria. If two of the same sized labs can easily pass the criteria, then surely the others can be sealed to the same standard of care to achieve the same results.

We also note that for rooms that are primary containment but still rated as CL3 (not CL3Ag or CL4), a risk assessment should be undertaken to validate acceptable leakage rates. Such spaces may have an increased requirement to contain potentially aerosolized agents and will likely see more frequent room fumigations occurring several times per year. Under these circumstances, consideration should be given to higher quality construction, and increased testing criteria such as that identified by the German VDI Guidelines.

5 RECOMMENDATIONS:

Industry standard construction materials and methods for CL3 Laboratories around the world typically use gypsum board or CMU with impermeable coatings. It is often difficult to obtain room pressure differentials above 250Pa for such construction and often, it is desirable to perform testing at lower pressures so as not to cause any damage to the structural integrity of the space. At the same time, pressures less than 125 Pa lead to inaccurate leakage rates and are not recommended. If the structure can handle the pressures, it is recommended to test the spaces at 250Pa (1 inch of wc) or greater, and could be positive or negative pressure differentials, or both.

The recommended procedure is a two-step process,

1. Initial leaks are identified with smoke pencil or soap bubble testing and repairs are made to seal all visible leaks.

2. Use a quantifiable leakage rate to verify the room meets minimum leakage requirements.

This procedure avoids several problems common to CL3 construction and testing practices and ensures a better end product for the user:

1. If only relying on visual methods, contractors may introduce gross leaks that are easily identified and repaired, ignoring small leaks.

2. Even if done by a third party, the quality of smoke pencil and soap bubble testing is a function of the operator, and someone with less experience may not find all the leak points.

3. Quantitative testing alone may leave small holes that would be found with visual indicators such as smoke pencil or soap bubbles.

The ARS greenhouse recommended leakage rate at a room differential pressure of 300 Pa (1.2 inches of wc) of 0.152 l/s per square meter (0.03cfm per square foot) of surface area is recommended as an acceptable criterion for testing construction boundaries of CL3 laboratories constructed using todays common CL3 construction practices. We also note that a specific containment facility in the United Kingdom has adopted an acceptable leak test rate for CL3 spaces which is equivalent to the ARS greenhouse standard. The table in figure 5.1 below identifies the ARS greenhouse flow per unit area recommended leakage equivalence for different room differential pressures, with the yellow highlight as the actual published value.

ARS Greenhouse Leakage Equivalency Table				
Pressure Pa	**Leakage L/s per sq.m**		**Pressure inch of w.c.**	**Leakage CFM per sq.ft**
500	0.196		2	0.039
400	0.176		1.6	0.035
375	0.170		1.5	0.033
350	0.164		1.4	0.032
300	0.152		1.2	0.030
250	0.139		1	0.027
200	0.124		0.8	0.024
150	0.107		0.6	0.021
125	0.098		0.5	0.019

Figure 5.1: ARS Greenhouse Leakage Equivalence vs Room Differential Pressure

The Merrick best practice leakage rate at a room differential pressure of 500 Pa (2 inches of wc) of 2% of room volume per minute also produces similar results and could be considered as an alternative test method. Note that the equivalent pressure decay rate is very short and thus difficult to quantify. If using a pressure decay rate, the time should be taken measured to decay from starting pressure to half the starting pressure for accuracy. The table in figure 5.2 below identifies the percent of room volume recommended leakage equivalence for different room differential pressures and equivalent pressure decay rates to half of the starting pressure.

Percent Volume Leakage Equivalency Table						
Pressure Pa	Pressure inch of w.c.	Leakage %Vol per Min		Decay to Press. (Pa)	Decay to Press. of w.c.	Decay Rate (seconds)
500	2.0	2.00		250	1.00	8.9
400	1.6	1.79		200	0.80	7.9
375	1.5	1.73		188	0.75	7.7
350	1.4	1.67		175	0.70	7.4
300	1.2	1.55		150	0.60	6.9
250	1.0	1.41		125	0.50	6.3
200	0.8	1.26		100	0.40	5.6
150	0.6	1.10				
125	0.5	1.00				

Figure 5.2: Percent Volume Leakage and Pressure Decay Equivalence vs Room Differential Pressure

For rooms that are primary containment but still rated as CL3 (not CL3Ag or CL4), or CL3 spaces that may have an increased frequency of room fumigations, should follow the more stringent requirements of the German VDI guidelines. The acceptable leakage rate at a room pressure differential of 250 Pa (1 inch of wc) is noted 0.03620L/s per square meter (0.007CFM per square foot) of room surface area. The table in figure 5.3 below identifies the VDI flow per unit area recommended leakage equivalence for different room differential pressures, with the yellow highlight as the actual published value at 250Pa room differential pressure. The acceptable airflows as a result of following the VDI standard are quite low, so appropriate instrumentation such as a rotameter needs to be used for accurate measurements.

German Leakage Class 4				
Pressure Pa	Leakage L/s per sq.m		Pressure inch of w.c.	Leakage CFM per sq.ft
500	0.057		2	0.011
400	0.049		1.6	0.010
375	0.047		1.5	0.009
350	0.045		1.4	0.009
300	0.041		1.2	0.008
250	0.036		1	0.007
200	0.031		0.8	0.006
150	0.026		0.6	0.005
125	0.023		0.5	0.005

Figure 5.3: German VDI Leakage per Unit Area vs Different Room Differential Pressure

APPENDIX A: EXPLANATORY NOTES ON LEAK TESTING

The recommended testing procedure is a two-step process; 1) Initial leaks are identified with smoke pencil or soap bubble testing in a qualitative manner and repairs are made to seal all visible leaks.2) A quantifiable leakage rate is used to verify the room meets minimum leakage requirements.

Quantifiable leakage rate can be determined from tables in the recommendations of the body of the report or through a project specific risk assessment.

A-1 ROOM PREPARATION

Fill all P-Traps with water.

Seal ventilation openings with plastic and tape. If provided, use bubble tight dampers in ductwork.

Close and seal all doors with plastic and tape.

Door Plate Installation (If using)

- Must be installed on containment perimeter such that applied test pressure pulls/pushes door plate against door stop and allows for door frame to be part of leak test.
- Door closers and any bump-stops located on door stop surface must be removed prior to fitting the door plate.

Provide a calibrated digital manometer installed across the containment perimeter in a manner to minimize interference with wind or ventilation turbulence and to accurately represent the interior and exterior differential air pressure.

Provide a portable fan to be used to achieve required test pressures in room.

- Provide all adapters, fittings, bubble tight isolation valves, and flow monitoring stations required to perform testing as defined herein

A-2 STEP 1 (QUALITATIVE BOUNDARY TESTING)

1 Use portable pressurization fan to pressurize room to a negative pressure (500 Pa (2 inch wc) or maximum structural tolerance pressure). Use fan controls to maintain room at a steady negative pressure through the duration of smoke pencil/soap bubble testing. Be careful not to over pressurize the room such that structural damage may occur.

2 Use a smoke pencil and/or a liquid detergent for soap bubble testing at locations that could be point sources of air leakage.
   2.1 Note that a room positive pressure is desirable for smoke pencil testing to easily see the smoke escaping through pin holes, while a room negative pressure is desirable for soap bubble testing to easily see bubbles forming on air being drawn in through pin holes.
   2.1 Identify all relevant leak points for repair
   2.2 Any leaks found through smoke pencil and/or soap bubble testing shall be repaired.

3 Repeat test identified in item #1 above.

4 Acceptance Criteria: No visible leaks found after repairs completed.

A-3 STEP 2 (QUANTITATIVE BOUNDARY TESTING)

1 Use portable pressurization fan to pressurize room to a negative pressure (recommended 250Pa (1 inch wc). Use fan controls to maintain room at a steady negative pressure through the duration of leakage testing. Be careful not to over pressurize the room such that structural damage may occur.

2 Record/trend room pressure and airflow over a 20-minute period at five second intervals.

3 Acceptance Criteria: Leakage rate is less then pre-determined quantifiable leakage rate.

APPENDIX B: FORMULAS AND SUPPORTING CALCULATIONS

B-1 ORIFICE EQUATION

Using the published ASHRAE formula for crack leakage, airflow through a crack can be calculated as:

$$Q = C * A * dP^{0.5},$$

Where Q = flow, C = the flow constant, A = orifice area, and dP = the pressure differential across the crack. To calculate the estimated flow at different pressures, the flow constant and the orifice area can be assumed to be equal, thus the formula can be reduced to:

$$Q2 = Q1 * (dP2/dP1)^{0.5}.$$

To compare the results from the different test facilities, all leakage rates were correlated to an equivalent leakage rate at a differential pressure of 250Pa (1 inch wc) using this formula.

B-2 IDEAL GAS LAW

The ideal gas law is used to analytically calculate certain properties of gases based on known factors and basic assumptions. The generic form of the ideal gas law is as follows:

$$PV=nRT,$$

Where P = absolute pressure in Pascals, V = the volume of the occupied gas in cubic meters, n = the number of moles of the gas, R = the universal gas constant (8.314 Pa·m3/(K·mol)), and T = the absolute temperature in Kelvin.

For an ideal gas, such as air, at room temperature and pressure, 1 mole of gas occupies 24.5 liters of space.

$$n = 101{,}300\text{Pa} * 0.0245\text{m}^3 / (8.314 * 298.5\text{K})$$
$$n = 1 \text{ mole}$$

The ideal gas laws are used to calculate the number of mols of air that escape during a pressure decay test, which can then be converted to a flow rate. Refer to B-6 for more information on these calculations.

B-3 UNIT CONVERSION

1 cfm = 0.472 L/s
1 cfm per ft2 = 5.08 l/s per m2 and vice versa
1 Inch wc = 250 Pa
1 PSI = 6895 Pa

B-4 CONVERSION OF PRESSURE DECAY TO LEAKAGE RATE

For the purposes of this analysis, we will assume a typical laboratory space, 10m long, by 3.3m wide, by 3m tall. We will demonstrate the equivalences of each test procedure using this typical space, by calculating the equivalent leakage rate. For these purposes, we will assume the starting gauge pressure is 120 Pa. Assuming the atmospheric pressure is standard 101.3 kPa, then the actual starting pressure is 101,420 Pa. The ending gauge pressure is 20 Pa, or equivalent actual pressure of 101,320 Pa. The time it took for the pressure to decay the specified amounts is five minutes.

Surface Area of space (SA) = (2 x 10 x 3.3) + (2 x 3.3 x 3) + (2 x 10 x 3)
SA = 66 + 20 + 60
SA = 146m2
Volume of space (V) = 10 x 3.3 x 3
V = 99m3

At starting temperature and pressure, the starting mols equals:

n1 = P1V/RT
n1 = 101,420Pa * 99m3 / 8.314 x 298K
n1 = 4,052.6 mols

After decay the number of mols remaining in the space is:

n2 = P2V/RT
n2 = 101,320Pa * 99m3 / 8.314 x 298K
n2 = 4,048.6 mols

And after decay, the number of mols escaping the space is:

ne = 4,052.6 – 4048.6
ne = 4 mols

Assuming that on average, I mol of gas occupies 24.5 liters of volume at room temperature and pressure, the volume of standard air escaped from the space after five minutes is:

Va = 24.5l * 4 mols
Va = 98l

The average airflow leakage (AL) from the space is calculated as:

AL = 98l / 5min * 1min/60s
AL = 0.327l/s or 1.176m3/hr

Using the AHRAE formula for crack leakage, the proposed leakage rate at a pressure of 250 Pa can be calculated:

Q2 = Q1 * (dP2/dP1)^0.5.
Q2 = 0.327L/s * (250Pa/70Pa)^0.5
Q2 = 0.61L/s (1.3CFM)

This leakage rate can be easily converted into a cfm per square foot or percentage of room leakage value based off the assumed surface area and volumes of the room, as demonstrated in other sections of this report.

B-5 EFFECTS OF TEMPERATURE

Using the ideal gas law, the effects of temperature on pressure were calculated and recorded in Figure B-1. Note that small temperature changes of 0.05°C (0.09°F), can result in pressure fluctuations of 17 Pa (0.07"), which can be detrimental to pressure measurements when trying to record changes less than 25 Pa (0.1"). Therefore when performing any boundary integrity test, it is critical that the space temperature is also monitored and confirmed to be stable throughout the duration of the test.

P1g (in)	P1a (Pa)	T1 (K)	T2 (K)	P2a (Pa)	P2g (in)
0.5	101425	298	297.5	101254	-0.1834
0.5	101425	298	297.55	101271	-0.1151
0.5	101425	298	297.6	101288	-0.0467
0.5	101425	298	297.65	101305	0.0216
0.5	101425	298	297.7	101322	0.0899
0.5	101425	298	297.75	101339	0.1583
0.5	101425	298	297.8	101356	0.2266
0.5	101425	298	297.85	101373	0.2950
0.5	101425	298	297.9	101390	0.3633
0.5	101425	298	297.95	101407	0.4317
0.5	101425	298	298	101425	0.5000
0.5	101425	298	298.05	101442	0.5683
0.5	101425	298	298.1	101459	0.6367
0.5	101425	298	298.15	101476	0.7050
0.5	101425	298	298.2	101493	0.7734
0.5	101425	298	298.25	101510	0.8417
0.5	101425	298	298.3	101527	0.9101
0.5	101425	298	298.35	101544	0.9784
0.5	101425	298	298.4	101561	1.0467
0.5	101425	298	298.45	101578	1.1151
0.5	101425	298	298.5	101595	1.1834

Figure B-1: Temperature Effects

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