

HELIUM AS A REPLACEMENT FOR DICHLORODIFLUOROMETHANE IN CLASS II BIOLOGICAL SAFETY CABINET INTEGRITY TESTING

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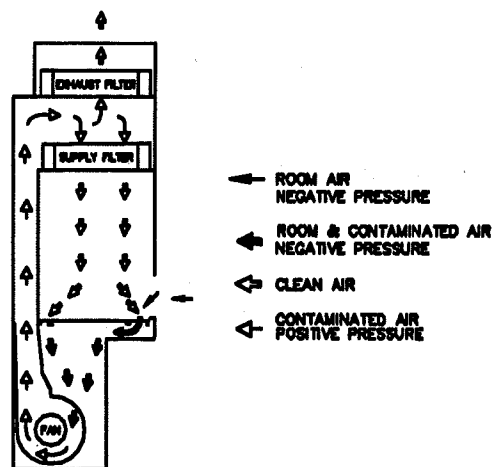
ABSTRACT

Dichlorodifluoromethane was commonly used for many years as a tracer gas for leak testing biological safety cabinets. International agreement to ban release of chlorofluorocarbons into the atmosphere led the United States Environmental Protection Agency to regulate the discharge limit to zero when adequate substitutes are available. Hence the need arose to develop a leak test using an alternative tracer gas that is environmentally friendly. Helium was compared to refrigerant 12 and found to be an adequate substitute in this testing of Class II Type A biological safety cabinets. A waiver was obtained from NSF International to permit the use of the proposed helium leak test as an alternative to the Halogen Leak Test.

INTRODUCTION

NSF International (NSF), formerly the National Sanitation Foundation, produces and publishes a consensus standard (Standard #49) that enables manufacturers and certifiers to test biological safety cabinets (BSCs) in a uniform manner to ensure that they are operating properly (NSF, 1992). NSF 49 requires cabinet manufacturers to perform the "Halogen Leak Test" on "all biologically contaminated air plenums that are under positive pressure to the room" (Figure 1) by pressurizing "the air filled cabinet at atmospheric pressure to 500 Pa (2" wg) with halide gas (dichlorodifluoromethane)" which dilutes the gas to a concentration of 0.5% inside the cabinet. Then scan for leaks with an "industrial-type halogen leak detector". "Absolute leakage from any point in the cabinet shall not exceed a leak rate of 5×10^{-7} atm cc/s to compensate for the dilution of halide gas." (NSF, 1992). What has been used in practice for this purpose is refrigerant 12 (R-12), a well known chlorofluorocarbon (CFC).

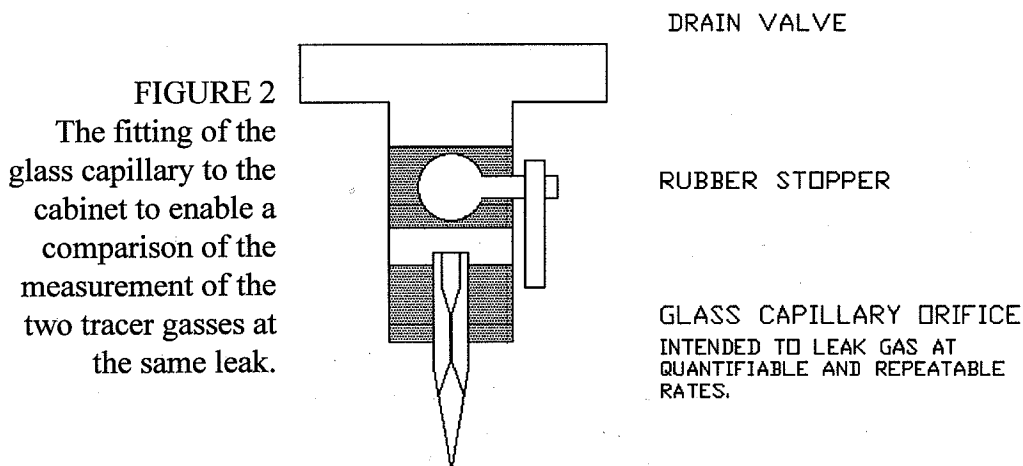
FIGURE 1
An example of the Class II
Type A cabinet airflow pattern
that results in biologically contaminated plenums being under positive pressure to the room.



The challenge arose when the United States Environmental Protection Agency (EPA) extended their regulation of CFC discharge beyond the refrigeration and air-conditioning industries and set the limit to zero when adequate substitutes are available (EPA, 1993). This called for the development of an alternative cabinet integrity test based on an adequate substitute for R-12 in order to meet the spirit of this EPA action. For a helium (He) test to be accepted as an adequate alternative to the R-12 test, it had to be shown that the helium test would quantitatively identify the same leak rate as the halogen leak test, under the test conditions.

METHODS AND MATERIALS

A fine glass capillary was drawn to the point that it would act as a limiting orifice and produce a constant R-12 leak close to 5×10^{-7} atm cc/s while using R-12 under the conditions called for by NSF 49. The capillary was inserted into a rubber stopper and the stopper was firmly pressed and turned into the threads of the drain valve on the bottom of a Class II Type A BSC (Figure 2).



R-12 Procedure

Refrigerant 12 (R-12) (Technical Chemical Company, Dallas, TX.) was introduced at the bottom of the cabinet through the drain valve to pressurize the cabinet to 2 inches of water column (2" wc) and the cabinet blower was run to mix the R-12 and air. A u-tube manometer was used to monitor the pressure inside the cabinet. The drain valve was closed and the R-12 source was disconnected. Then the capillary was fitted to the drain valve, the drain valve was opened and a General Electric Ferret Type H-25 halogen leak detector (now Yokagowa Corporation of America, Shenandoah, GA) was used to measure the R-12 leak rate at the capillary. This leak detector was calibrated to 5×10^{-7} with a calibrated General Electric (Yokagowa) Type LS -20 B standard leak.

Helium Procedure

Helium gas (BOC Gasses, Murry Hill, NJ.) was introduced into the top of the BSC through the aerosol sampling port that is normally used during HEPA filter testing. The helium was mixed with the air inside the cabinet by running the cabinet blower. The air-helium mixture was allowed to flow out of the drain valve at the bottom of the cabinet into a local exhaust to prevent a build up of helium in the room. Helium was continually replaced at the top of the unit until the concentration of helium inside the cabinet rose to 25% (vol/vol). At this point the drain valve was closed and helium introduction continued until the pressure in the cabinet reached 2" wc. The capillary was fitted to the drain valve and the valve was opened. The helium leak detector was calibrated to 4×10^{-5} atm cc/s with a calibrated standard leak (Helium Leak Testing, Inc., Northridge, CA). After the cabinet had been stabilized at 2" wc with the tracer gas, the blower was run to mix the gas with the air in the cabinet. The helium concentration in the cabinet was measured at a valve mounted in a petcock penetration and the helium leak rate was measured at the capillary using an Alcatel ASM 100 mass spectrometer helium leak detector (Alcatel Vacuum Products, Hingham, MA.) set up for atmospheric sampling.

Pressure was maintained at 2" wc during the sampling. Fifty consecutive leak rate measurements were taken at the capillary outlet with the cabinet pressurized with R-12 and again with the cabinet pressurized with helium.

LEAK THEORY

Generally, total leaks less than 10^{-7} atm cc/s are in the molecular flow regime and total leaks greater than 10^{-5} atm cc/s are in the viscous flow regime. In the molecular flow regime the mean free path of the gas molecules is greater than the leak hole diameter so most molecules collide with the passage walls rather than with each other. Each gas in a mixture flows as if it were the only gas present and the rate (Q) at which different gasses will leak under identical conditions is inversely proportional to the square root of their molecular weights (MW) : $Q_a/Q_b = MW_b^{1/2}/MW_a^{1/2}$. (Varian 1980, O'Hanion 1989, Wutz 1989).

In the viscous flow regime the collisions with the passage walls become insignificant and the rate at which two gasses will leak under identical conditions is inversely proportional to the viscosities (η) of the gasses: $Q_a/Q_b = \eta_b/\eta_a$.

When the gas is a mixture the viscosity becomes the weighted average viscosity based on the percent volume of each component:

$$[(\%GAS_a)(\eta_a) + (\%GAS_b)(\eta_b)] / 100 = \eta_{mixture}$$

If the gas mixture is leaking according to molecular flow, each component will leak at a rate inversely proportional to the square root of its molecular weight. However, in viscous flow the total leak is a function of the viscosity of the mixture with each component leaking at a rate directly proportional to the volumetric percent makeup of the mixture.

The viscosity values for the various gasses are shown in Table 1.

TABLE 1
Viscosities at approximately 70°F, and Sources of the Values, of the Applicable Gasses

Gas	Viscosity lb/ft. hr	Viscosity centipoises (cp)	Source
air	0.0442	0.0183	Bolz, 1973, page 650
helium	0.0465	0.0192	Extrapolated from Bolz, 1973, page 534
R-12	0.0318	0.0131	ASHRAE, 1993, page 175
SF ₆		0.0150	Extrapolated from L'Air Liquide, 1976, page 861

When a tracer gas is mixed with air the leak detection instrument is measuring the leak rate of the tracer gas and not the total gas leak through the hole (O'Hanion 1989, Wurtz 1989). If a 1% tracer gas leak is measured at 3×10^{-7} atm cc/s the total leak is 3×10^{-5} atm cc/s ($3 \times 10^{-7} / 0.01$). The total leak, in this case is 100 times the detected level of the tracer, and is therefore in the viscous flow regime.

The base criterion, set by NSF in the 1983 revision of Standard #49 for the BSC leak rate is 1×10^{-4} atm cc/s total leak rate that would be measured if there were 100% R-12 in the cabinet. To make the procedure practical for field applications an equivalent leak detection level was established based on pressurizing a sealed air-filled cabinet to 2"wc using 100% R-12. This results in an R-12 concentration of 0.5% based on ideal gas laws: $P_1V_1 = P_2V_2$. Since pressure and volume are inversely proportional, the percent of the BSC volume of R-12 that must be injected into the sealed cabinet is the same as the percent increase in pressure. With an ambient pressure of 407" wc the percent increase is $(409/407) - 1 = 0.5\%$. The R-12 leak rate under these conditions was assumed to be 5×10^{-7} atm cc/s:

$$\begin{aligned} Q_{R-12} &= (Q_{\text{total leak rate}})(\text{Concentration}_{R-12}) & (1) \\ &= (1 \times 10^{-4})(0.005) & = 5 \times 10^{-7} \text{ atm cc/s} \end{aligned}$$

This value is now published in NSF but the calculations for this criterion ignored the difference in viscosity between 100% and 0.5% R-12 as well as the associated effects on the total leak rate. A calculation that considers viscosity yields:

$$\begin{aligned} \eta_{R-12 \text{ mix}} &= [(\text{Concentration}_{R-12})(\eta_{R-12})] + [(\text{Concentration}_{\text{air}})(\eta_{\text{air}})] & (2) \\ &= [(0.005)(0.0131)] + [(0.995)(0.0183)] & = 0.0183 \text{ cp} \end{aligned}$$

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Then:

$$\begin{aligned}
 Q_{R-12 \text{ mix}} &= (\eta_{R-12}/\eta_{\text{mix}})(Q_{R-12}) & (3) \\
 &= (0.0131/0.0183)(1 \times 10^{-4}) = 7.2 \times 10^{-5} \text{ atm cc/s}
 \end{aligned}$$

And:

$$\begin{aligned}
 Q_{R-12} &= (Q_{R-12 \text{ mix}})(\text{Concentration}_{R-12}) & (4) \\
 &= (7.2 \times 10^{-5})(0.005) = 3.6 \times 10^{-7} \text{ atm cc/s}
 \end{aligned}$$

When considering the effects of the viscosity of R-12/air mixture, this result shows the leak rate obtained in equation (1) to be approximately 38% larger than expected.

Converting the 5×10^{-7} atm cc/s of 0.5% R-12 NSF requirement to an equivalent 25% helium leak rate requirement results in a calculated helium detection level of 2.5×10^{-5} atm cc/s:

At 25% helium in the cabinet the viscosity of the He/air mixture is:

$$\begin{aligned}
 \eta_{\text{He mix}} &= [(\eta_{\text{He}})(\text{Concentration}_{\text{He}})] + [(\eta_{\text{air}})(\text{Concentration}_{\text{air}})] & (5) \\
 &= [(0.0192)(0.25)] + [(0.0183)(0.75)] = 0.0185 \text{ cp}
 \end{aligned}$$

Starting with 5×10^{-7} atm cc/s leak rate of R-12 at 2" wc and 0.5% R-12 as NSF 49 requires, calculate an equivalent total leak of 100% R-12.

$$\begin{aligned}
 Q_{\text{total R-12 leak rate}} &= (Q_{R-12} / \text{Concentration}_{R-12})(\eta_{R-12 \text{ mix}}/\eta_{R-12}) & (6) \\
 &= (5 \times 10^{-7}/0.005)(0.0185/0.0131) = 1.4 \times 10^{-4} \text{ atm cc/s}
 \end{aligned}$$

Then calculate the total mixture leak rate of the 25% helium mixture at 2" wc:

$$\begin{aligned}
 Q_{\text{He mix}} &= (\eta_{R-12}/\eta_{\text{He mix}})(Q_{R-12}) & (7) \\
 &= (0.0131/0.0185)(1.4 \times 10^{-4}) = 1 \times 10^{-4} \text{ atm cc/s}
 \end{aligned}$$

Since only 25% of the total mixture leak is helium, the detection level is:

$$\begin{aligned}
 Q_{\text{He}} &= (Q_{\text{He mix}})(\text{Concentration}_{\text{He}}) & (8) \\
 &= (1 \times 10^{-4})(0.25) = 2.48 \text{ or } 2.5 \times 10^{-5} \text{ atm cc/s}
 \end{aligned}$$

This is well within the detection capability of the instruments available.

RESULTS AND DISCUSSION

The 50 consecutive leak rate measurements at the capillary outlet using the R-12 method resulted in 35 readings of 3×10^{-7} atm cc/s and 15 readings of 4×10^{-7} atm cc/s. These averaged to 3.3×10^{-7} atm cc/s which was rounded to 3×10^{-7} atm cc/s. This provided the R-12 base line against which the helium method could be compared.

It is impossible to measure a leak rate of helium in the 10^{-7} atm cc/s range because the 5 parts per million of helium in atmospheric air results in a 10^{-6} atm cc/s background leak rate when using the atmospheric apparatus. A leak rate can not be measured if it is smaller than the concentration of the tracer gas in the ambient air (background). Additionally, the leak rates of R-12 and helium are not the same through the same opening under the same pressure. In order to use helium to duplicate the R-12 test, the concentration of helium in the cabinet had to be raised to a level that would make it possible to measure total leak rates that would be equivalent to those measured in the R-12 test.

Using the above theory, with the measured R-12 capillary leak rate as the benchmark, helium at 25% should generate a measured capillary leak rate of 1.5×10^{-5} atm cc/s:

$$\begin{aligned} Q_{\text{He capillary}} &= (Q_{\text{R-12 capillary}})(\eta_{\text{R-12-air mix}}/\eta_{\text{He-air mix}})(\text{Concentration}_{\text{He}}/\text{Concentration}_{\text{R-12}}) & (9) \\ &= (3 \times 10^{-7})(0.0183/0.0185)(0.25/0.005) \\ &= 1.48 \text{ or } 1.5 \times 10^{-5} \text{ atm cc/s} \end{aligned}$$

The cabinet was brought to 25% helium at 2" wc, a local exhaust was used to keep the helium background down, and 50 consecutive measurements of the capillary Q_{He} were taken with the Alcatel helium leak detector. Each of the 50 readings was 1×10^{-5} atm cc/s which was 0.5×10^{-5} atm cc/s less than the theoretical 1.5×10^{-5} atm cc/s.

When raising the concentration of helium in the cabinet by flowing the gas through it, it takes less time and gas to reach 15% than it does to reach 25%. Therefore, it was deemed desirable to use 15% rather than 25% helium in the testing, which results in a leak rate that is 15/25 of the calculated 25% helium requirement (equation 8) To calculate an equivalent to the NSF-defined leak rate, multiply the helium leak rate at a 25% (2.5×10^{-5} atm cc/s) by 15/25. This results in a helium test requirement of 1.5×10^{-5} atm cc/s to be equivalent to the 5×10^{-7} atm cc/s of the NSF R-12 test.

$$\begin{aligned} Q_{\text{He at 15\%}} &= (\text{Concentration ratio})(Q_{\text{He at 25\%}}) & (10) \\ &= (15/25)(2.5 \times 10^{-5}) & = 1.5 \times 10^{-5} \text{ atm cc/s} \end{aligned}$$

This leak rate was reduced by the observed experimental error to derive a leak rate requirement (1×10^{-5} atm cc/s) that would provide a helium test equivalent to the R-12 test.

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An extrapolation from this work is that sulfahexafluoride (SF₆) could be used as another acceptable substitute for R-12 for Class II type A cabinet integrity testing. Using the viscosity of SF₆ as 0.015 cp and that of R-12 as 0.0131 cp in the viscous flow calculation yields:

$$\begin{aligned} \eta_{\text{SF}_6 \text{ mix}} &= [(\text{Concentration}_{\text{SF}_6})(\eta_{\text{SF}_6})] + [(\text{Concentration}_{\text{air}})(\eta_{\text{air}})] && (2) \\ &= [(0.005)(0.015)] + [(0.995)(0.0183)] = 0.0183 \text{ cp} \end{aligned}$$

$$\begin{aligned} \eta_{\text{R-12 mix}} &= [(\text{Concentration}_{\text{R-12}})(\eta_{\text{R-12}})] + [(\text{Concentration}_{\text{air}})(\eta_{\text{air}})] && (2) \\ &= [(0.005)(0.0131)] + [(0.995)(0.0183)] = 0.0183 \text{ cp} \end{aligned}$$

Therefore, the theory shows SF₆ and R-12 to behave the same when leaking as a 0.5% mixture with air. This means that one should be able to duplicate the R-12 test by following the NSF 49 procedure for the “halogen leak test” while using SF₆ in place of R-12 and setting the alarm on an appropriate SF₆ leak meter at 5.0 x 10⁻⁷ atm cc/s.

A summary of the equivalent leak information is shown in Table 2.

TABLE 2
Equivalent Leaks

Leak Rates atm cc/s		Gas Mixtures in Sealed BSC at 2" w.c.			
		100% R-12	0.5% R-12	25% He	15% He
Base Criterion (NSF)	Total Mixture	1 x 10 ^{-4 a}	7.2 x 10 ^{-5 (3)}		
	Tracer Gas	1 x 10 ^{-4 b}	3.6 x 10 ^{-7 (4)}		
Current Criterion (NSF)	Total Mixture	1.4 x 10 ^{-4 (6)}		1 x 10 ^{-4 (7)}	
	Tracer Gas	1.4 x 10 ^{-4 (6)}	5 x 10 ^{-7 (1)}	2.5 x 10 ^{-5 (8)}	1.5 x 10 ^{-5 (10)}
	Recommended Based on Empirical Results				1 x 10 ^{-5 e}
Empirical Data	Measured Tracer Gas from Capillary		3 x 10 ^{-7 c}	1 x 10 ^{-5 d}	
	Calculated Equivalent			1.5 x 10 ^{-5 (9)}	

(#) is equation number from text.

- a) Base criterion (NSF 1983)
- b) Same as a) since gas is 100%
- c) Measure from capillary leak
- d) Measure from capillary leak
- e) Recommendation adjusted based on empirical results.

CONCLUSIONS

It was concluded that helium can be used as an adequate substitute for R-12 in cabinet integrity testing of Class II type A biological safety cabinets.

A waiver was obtained from NSF to use helium, as a replacement for R-12 in the testing of these cabinets, following an agreed upon method using 1×10^{-5} atm cc/s with 15% helium and 2" wc as the requirement.

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