Detection of Gas Leaks in an Anaerobic Glove Box

GILDA L. JONES AND STANLEY M. DEVER
Center for Disease Control, Atlanta, Georgia 30333

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An inert gas, Freon, can be added to the atmosphere of an anaerobic glove box without deleterious effect to cultures of anaerobic microorganisms. The sensitive probe of a Halogen Leak Detector passing over the outside surface of the box will pinpoint any escaping Freon and therefore locate the leak.

An anaerobic glove box constructed of clear, flexible vinyl plastic was designed by Rolf Freter of the University of Michigan (1). This gas-tight box can maintain the anaerobic atmosphere necessary for the isolation and identification of anaerobic bacteria (2).

Once the glove box has been evacuated of normal atmospheric gases and filled with the recommended gas mixture, it should not be necessary to add more gas to the chamber atmosphere unless, for some reason, a tear or puncture develops in the vinyl plastic. These leaks are generally quite small and therefore difficult to locate. No really good system for locating such a leak has been described. "Snoop," a commercial liquid compound, is helpful in detecting gas leaks in areas where the plastic has been joined to other materials, such as glove and entry ports, to make it more functional. But use of a liquid is awkward and time consuming for covering large surfaces.

A simple and inexpensive system that works well makes use of the colorless refrigerant gas, Freon. This method has been used for leak detection in germfree isolators for a number of years and should prove equally useful in anaerobic glove box procedures. Freon is available in small cylinders which can be easily taken into the chamber. When a very small amount of Freon is released into the atmosphere of a glove box, it mixes with the gases in the box and escapes through any pinpoint hole that would allow the other gases to escape.

Sensitive detectors for a specific type of Freon are available commercially or can be borrowed from the refrigeration maintenance department in most large institutions. An example of such a detector is the General Electric Halogen Leak Detector, type H-10, specific for Freon-12. After Freon-12 has been released inside the glove box, the detector probe is simply moved over the outside area of the box to scan for escaping Freon, particularly at possible points of stress. An indicator light in the probe will register even minute amounts of Freon.

We have noted no effects on the growth of anaerobic cultures in the glove box at the time of its use. However, Bauchop (1967) showed that rumen microbial methane formation could be selectively inhibited in the presence of low concentrations of certain halogenated hydrocarbons such as methylene chloride (CH₂Cl₂), chloroform (CHCl₃), and carbon tetrachloride (CCl₄). He further states that it is probable that Freon (CCl₂F₂) acts in a similar manner. However, with this in mind, the Freon method still offers a simple and inexpensive way of detecting leaks in an anaerobic glove box.

LITERATURE CITED