Device for Ensuring Continuity of Air Supplied to Carbon Dioxide Incubators for Tissue Culture

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A simple device to ensure the continuity of air supplied to carbon dioxide incubators for tissue cultures is described.

Tissue culture incubators that are flushed continuously with a precisely regulated mixture of air and carbon dioxide have proved to be a remarkable improvement over older designs which provided no control over the ambient incubator atmosphere. The CO₂ incubators permit monitoring of the culture atmosphere which is essentially an extension of the incubator atmosphere. Therefore, pH can be maintained within narrow limits, and it is possible to use petri dishes and other culture vessels which are not sealed.

There are several methods of providing the supply of air and carbon dioxide to flush such incubators. Compressed-gas cylinders with the proper air-CO₂ ratio may be purchased. Alternatively, compressed-CO₂ gas cylinders may be used in conjunction with institutional compressed-air lines to establish an appropriate mixture. This latter approach requires attention and the use of flowmeters, but this method is cheaper than purchasing cylinders of the established mixture. Further, after opening the door of a CO₂ incubator, the air-CO₂ ratio is destroyed because of the rapid admixture of room air. The proper mixture may be readily established by "purging" the incubator with a brief flow of CO₂ without added air. This technique is not possible with a premixed, and therefore fixed, gas ratio provided from a single gas cylinder.

Mixing CO₂ with the available house air has one major liability. The continuity of service of the institutional air supplied to the laboratory must be reliable. Any interruption of the air provided to the mixing apparatus will result in the entrance of unadulterated carbon dioxide. The gravity of this event is dependent upon the duration of the exposure. With an interruption of air supply on a weekend or evening, most cell cultures would be destroyed before the situation is detected. After experiencing several such events, we developed a device which has been proven to be quite reliable in preventing such occurrences.

This device (Fig. 1) consists of a compressed-air cylinder linked in parallel with the house air-supply line. Pressure regulators and one-way flow-check valves prevent air from the compressed-air cylinder from entering the house air lines, regard-

Fig. 1. Parallel air supply to CO₂ incubator. (1) Air filter. (2) Line regulator, (3) inlet nut and nipple (NCG 513410-74 and 513520-74), and (4) check valve (NCG 200303-62), all from the National Cylinder Gas Co., King of Prussia, Pa. (5) Standard pipe adapter, 3/4 to 1/2 inch (1.9 to 1.2 cm).

less of the ambient cylinder pressure. In operation, there is no flow of air from the compressed-air cylinder unless the pressure in the house air line drops lower than the flow-pressure regulator-valve setting of the air cylinder. We find that it is best to set the flow pressure of the compressed air cylinder regulator valve to a level which is about 15 psi less than the pressure in the house air line.

The device has been demonstrated to be highly reliable. We have simulated house air supply

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failures on approximately 100 occasions. In each case, the compressed-air cylinder provided air to the incubator when the house air pressure dropped below the preset threshold. When the pressure was restored in the house air line, the flow from the compressed-air cylinder was automatically shut off. The unit is relatively inexpensive (about $50, less air cylinder and its regulator valve), and can readily be installed by a plumber in about 1 hr.

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