A temperature regulator for microtiter plates

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Microtiter plates are frequently being used as reaction vessels for a myriad of microanalytical and micropreparative biochemical and molecular biological procedures. As an example, 96-well microtiter plates are used in the preparation of plasmid DNA by alkaline lysis (1) and in the subsequent large-scale restriction enzyme digestion analysis of this DNA. In addition, enzyme reaction-based protocols such as large-scale and high-throughput DNA sequencing can be facilitated by the use of these microtiter plates (2). Other microanalytical applications include large-scale enzyme assays (3) and enzyme-linked immunosorbent assay applications (4, 5). This paper describes an inexpensive and simple microtiter plate temperature regulator that has been developed to allow a rapid attainment of a desired temperature and to maintain this constant temperature over a period of time.

This incubator is schematically diagrammed in Figure 1. A copper strip was obtained and soldered onto a copper plate. Note that the incubator is placed on top of a *separate* commercially available heater block (e.g., VWR Scientific Co.) which is then placed into a modular heater unit (e.g., VWR Scientific Co.). Similar heating units may also be obtained from other vendors; they are interchangeable since the incubator is designed simply to sit on top of any generic heating block. Since the manufacturers caution against placing water directly into their heating blocks (an electrical hazard), washed sea sand is used as the temperature conductor. The individual wells of the microtiter plate are form-fitted in the sand, which facilitates heat transfer regardless of well bottom shapes. Even though the incubator is slightly larger than the heater block, the sand allows for even distribution and constant temperature maintenance across the entire incubator.

A specific application for the microtiter plate incubator is in large-scale and high-throughput DNA sequencing reactions. This is applicable to both radioactive and fluorescent modes of manual DNA sequencing. These reactions typically require heating at 55°C for five minutes and 37°C for seven minutes (2), necessitating rapid changes in temperatures. A set of two incubators allows for optimal consecutive temperature incubations for these reactions. Current alternatives for microtiter plate heating are custom-milled temperature blocks (2), water baths (3, 6), hot air incubators (5) and sub-optimal benchtop incubations performed at room temperature. The geometry of the microtiter plates and the inefficiency of air (dry heating) to conduct heat excludes certain alternatives listed above, such as common bacteriological incubators and forced air microtiter plate incubators (5). Water bath hazards include liquid splashing onto the samples and the possible sinking of the microtiter plates, which may lead to radioactive contamination of the water bath.

Thus, a microtiter plate temperature regulator has been developed to complement large-scale microanalytical and micropreparative procedures, in particular large-scale and high-throughput DNA sequencing. Since this simple and inexpensive

incubator is designed to complement existing common laboratory modular heater units, the need for additional equipment is obviated.

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Note added in proof. A 9.5×13.5×1.5 cm version is available from Bel-Art Products (Pequannock, New Jersey).

Figure 1. Schematic of the microtiter plate incubator. The dimensions of this temperature regulator, are shown in the forms of (A) front view, (B) top view and (C) three-dimensional view. The numbers are given in centimeters.