## Use of Robotics To Dispense Culture Media<sup>†</sup>

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Liquid and agar-containing media can be aseptically dispensed by a laboratory robot. Five errors occurred during the filling of 700 tubes with medium; four of these were due to imperfections of the screw-threaded caps of the culture tubes. The mean volume dispensed was 14.8 ml (standard deviation, 0.5; n = 50) when the system was set to deliver 15 ml.

Some procedures used in microbiological laboratories are repetitive but require skill and concentration on the part of workers to achieve accuracy and avoid contamination. The hand movements of skilled technicians reveal that the trajectories of their motions are consistent. In theory, therefore, a robot arm and hand can mimic these motions. In 1985, a number of laboratory robots appeared on the market and it occurred to us that these were suitable instruments to test the use of robots to reproduce microbiological procedures. In this note, we report on an automated method of dispensing culture media.

The robot arm (A in Fig. 1 and 2), a capping station (C in Fig. 1) with extended jaws, a power-and-event controller (Fig. 3), the interfaces among these components, the computer, and the culture tube racks were purchased from Zymark Corp., Hopkinton, Mass. The counting vanes in the capping station were increased from two to four to improve the precision of rotation. The medium reservoir (R in Fig. 1) was fitted with a standard screw-thread side arm (R1) through which passed the dispensing tube (glass) and a controlling thermometer (T in Fig. 1) to maintain the temperature of agar-containing media through the stirrer hot plate (Fig. 1). Medium was pumped into the culture tubes by the Harvard pump (P in Fig. 1; Ealing Scientific Ltd., St. Laurent, Quebec, Canada), which was turned on and ofl and timed by the power-and-event controller (Fig. 3). A similar pump was used and controlled in the same way to empty the water bath (Fig. 1).

Screw-cap culture tubes (n = 50; 150 by 20 mm) with caps loosely attached, the dispensing line (latex tubing; inside diameter, 8 mm), nozzle (D in Fig. 1), pump cartridge (Autoclude; inside diameter, 6 mm), and thermometer pocket were autoclaved for 20 min on a fast exhaust-and-dry cycle at 120°C. Sterile medium in the reservoir (R in Fig. 1) was mounted on the hot plate, the thermometer pocket was inserted, silicone oil (10 ml) was added, and the controlling thermometer was placed in position (Fig. 1). The dispensing line was then attached, and the nozzle (D in Fig. 1) was clamped in position. The sterile support (E in Fig. 1) used to hold the cap during the filling operation was set in position, and the sterile culture tubes, their caps tightened, were mounted in the rack. Finally, the equipment was irradiated (GE G30T8 lamps) with UV light for 10 min.

Subsequent operations were directed by the computer (Fig. 3). A brief description of these operations follows. The dispensing line was purged of air by pumping medium through it for 52 s. The medium emerging from the nozzle (D

in Fig. 1) during the purge was collected in an Erlenmever flask located in a precisely defined position (Fig. 2). The following describes the motions of each of the 50 culture tubes. The tube was placed in the capping station, and the cap was removed and placed on the support (E in Fig. 1). The tube was placed under the nozzle (D in Fig. 1), medium was pumped into the tube, the tube was replaced in the capping station, and the cap was recovered and replaced. The volume dispensed into the tubes was controlled by the computer, which set the time for which the pump was on. By appropriate programming, this could be changed during a 50-tube run. The volume could also be controlled by setting the pumping speed, but it was found that the viscosity of agar-containing media imposed an optimum speed for greatest accuracy. The average time over 700 fillings for which the tube was open to the air was 40 s (standard deviation, 0.5). The tube was removed from the capping station, its successful removal (and hence all filling operations) was checked by closing the switch (S in Fig. 1), and then it was replaced in the rack.

The mean time for filling 50 tubes (n = 14), was 80 min, and with liquid media the cycle ended after tube 50 had been replaced in the rack. When media containing agar were dispensed, after tube 50, the robot recovered the capstan (C-A in Fig. 1), mounted it in the capping station, and cranked the rack into a position 45° from horizontal. This was achieved by hinging the rack at its forward edge to the base of the water bath (Fig. 1) and connecting the capstan to the top rear edge of the rack by a nylon cord. The water in the bath was then pumped out, and after 1 h the tension on the nylon cord was released and the rack was returned to the horizontal position by the tension on the spring (SP in Fig. 1). Meanwhile, the hot plate under the reservoir (R in Fig. 1) was turned ofl, the robot hand was parked, and the robot arm was placed in a safe position.

Error routines were activated when the switch (S in Fig. 1) was not closed. In both cases, it was assumed that the robot retained the cap in its fingers. Before filling, the cap was dropped, the failure was recorded, and an attempt was made to pick up the next tube in the rack. After filling, it was assumed that the tube remained in the capping station (C in Fig. 1). Thus, a second attempt to cap the tube was made. and if that failed too, an attempt was made to remove the tube from the capping station and return it to the rack; the error was recorded.

The computer programming used in this work (programs are available to interested colleagues on application) used the restricted Easylab language devised by Zymark Corp. Newer versions of the language are said to be more flexible, and in our experience this is desirable. The procedure

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FIG. 1. General view of the apparatus used for dispensing culture medium. A, Robot arm; C, capping station; the other letters are referred to in the text.

described has been in use in our laboratory for about 18 months. During this time, the servomotor controlling the fingers was replaced, and its potentiometer required regular adjustment for smooth action of the fingers; otherwise, only routine maintenance was needed.



FIG. 2. Map of instrument locations. The robot arm is not shown. The dotted circles represent the inner and outer limits of the fingers.

Sterility checks were made by incubating culture tubes containing Pennassay broth (Difco Laboratories, Detroit, Mich.), nutrient broth, and 2% malt agar at 25 and 35°C, but no contamination was seen. We have no evidence of contamination in any of the several thousand cultures that have been maintained on media prepared in this way.

During the dispensing of 700 tubes of media, five errors occurred; in one case, the tube slipped out of the fingers while it was being filled, and in the remaining four cases, the errors were due to imperfections in the glass threads of the culture tubes. The mean volume of liquid medium dispensed into 50 tubes was 14.8 ml (standard deviation, 0.5) when the system was set to dispense 15 ml.



FIG. 3. Electrical connection plan of units used for medium dispensing. The computer, power-and-event controller, printer, and water bath pump are not shown in Fig. 1 and 2.