sulfadiazine were similar to those with sulfathiazole (9 cultures).

The S strain was grown on thioglycollate medium for 20 days, at which time there was luxuriant growth in every tube. Autoclaved thioglycollate medium containing sulfanilamide, sulfathiazole or sulfadiazine was then added to the cultures in amounts to make the total drug concentration 50 or 100 mg per cent. (5 results with each drug in each concentration). Fifteen days later the tubes were subcultured. While the controls grew profusely, only 3 cultures from the sulfonamide groups showed any growth at all, and this was minimal. There was no growth in the subcultures of the sulfathiazole or the sulfadiazine tubes.

The M strain, when grown aerobically, gave a plentiful growth in one month. A similar amount of growth, although somewhat slower in development, occurred in cultures containing 10 mg per cent. of sulfanilamide (3 cultures). No growth occurred in cultures containing 50 or 100 mg per cent. sulfanilamide, or 10, 50 or 100 mg per cent. of sulfathiazole or sulfadiazine (3 results with each drug in each concentration).

Thus it is apparent that any of the 3 sulfonamide drugs used more or less completely inhibited the aerobic growth of both strains of actinomyces. Low concentrations were less effective than high, and sulfanilamide was less effective than sulfathiazole or sulfadiazine.

#### ANAEROBIC CONDITIONS

The S strain was grown anaerobically on Krainsky's medium in sulfanilamide, sulfathiazole and sulfadiazine, at 10, 50 and 100 mg per cent. concentrations (3) results with each drug in each concentration). Growth was plentiful in the control tubes, and in one of the sulfanilamide (10 mg per cent.) tubes in one month. Of the remaining 26 tubes, 8 showed very slight growth, while 18 showed no growth. The positive growths occurred irregularly in the presence of the various drugs in different concentrations.

The S strain was grown anaerobically on thioglycollate medium with sulfanilamide, sulfathiazole and sulfadiazine in concentrations of 10, 50 and 100 mg per cent. (3 results with each drug in each concentration). In comparison with the vigorous growth in the control tubes in one month, sulfanilamide permitted equally good growth in 10 and 50 mg per cent. concentrations, and poor or no growth in 100 mg per cent. concentrations. Sulfathiazole allowed fair growth in 10 mg per cent. concentrations, poor or none in 50 and 100 mg per cent. concentrations. Sulfadiazine allowed little or no growth at any concentration.

The M strain was grown anaerobically with sulfanilamide, sulfathiazole and sulfadiazine in concentrations of 10, 50 and 100 mg per cent. (3 results with each drug in each concentration). Growth in the control tubes was moderate, and that in sulfanilamide (10 mg per cent. concentrations) was almost as good, but it was absent in all other tubes after one month.

The anaerobic results, therefore, agreed with the aerobic.

### Conclusions

(1) Aerobic and anaerobic cultures of two strains of Actinomyces hominis were inhibited to some extent by sulfanilamide in a concentration of 10 mg per cent.

(2) Concentrations of 50 and 100 mg per cent. checked growth more or less completely.

(3) Sulfathiazole and sulfadiazine were definitely more effective than sulfanilamide in similar concentrations. WINDSOR C. CUTTING

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# SCIENTIFIC APPARATUS AND LABORATORY METHODS

## AN APPARATUS FOR ROLLER TUBE TISSUE CULTURE

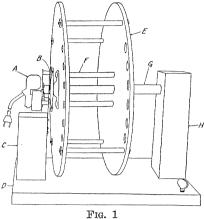
THE roller tube method of tissue culture, originated by Gey<sup>1</sup> and developed by Lewis,<sup>2</sup> is being more and more widely adopted. The apparatus usually employed, consisting of a 1/20 h.p. electric motor, reduction gears and a rotating tube carrier, has several disadvantages. It is expensive and causes more or less noise and vibration. The motor can not be housed inside the incubator because it develops sufficient heat of itself to exceed 38° C. This necessitates a cumbersome apparatus with a shaft running from the motor through the wall of the incubator to the tube carrier.

A relatively simple and inexpensive device which does not have these disadvantages has been successfully used in this laboratory. The apparatus consists of a motor and tube carrier conveniently mounted to form one complete unit.

The general structure of this apparatus is shown in Fig. 1. The motor, A, was obtained from a General Electric clock, of the dressing table type, having an automatic starting mechanism. The motor was detached from the clock and removed from its metal housing. The exposed gears were removed until there

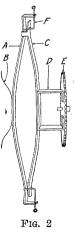
<sup>&</sup>lt;sup>1</sup>G. O. Gey, *Am. Jour. Cancer*, Vol. 17, 1933. <sup>2</sup> Warren H. Lewis, Contributions to Embryology No. 150, July, 1935.

remained only the gear on the main drive shaft and two gears connected to it. One of these was found to turn at a speed of 9 revolutions per hour, a satisfactory rate for rotating the cultures. A piece of 1/32 inch sheet brass, B, cut so as to leave the desired gear well exposed, was fastened to the face-posts of



the clock, and a small hole drilled through this sheet supported the end of the shaft of the intermediate gear. The sheet of metal also served to fasten the motor to its mount, C, a  $45/16 \times 2 \times 2$  inches wooden post fastened with screws to the  $12 \times 9 \times 3/4$  inches plywood base, D.

The roller tube carrier, E, consists of two circular pieces of 1/4 inch plywood, 12 inches in diameter, with eighty-four 5/8 inch holes to accommodate the test-tubes. These wooden discs were mounted on the frame of a bicycle pedal, F, and further secured to each other by 4 metal rods at their peripheries. The



distance between the discs so mounted is  $3 \ 1/2$  inches. The posterior disc was fitted with a cardboard backing with holes 1/2 inch in diameter to prevent the tubes from sliding out of the carrier. The bicycle pedal should be of good construction, about 4 1/2inches long, with ball bearings. The shaft of the pedal was screwed into a steel bar, G, 4 3/8 inches long, fitted snugly into a hole drilled through a wooden post, H, 7  $1/2 \times 2 \times 2$  inches. This post was screwed to the base.

The motor was attached to the tube carrier, as shown in Fig. 2.

A thin strip of metal, A, 3 inches

long and 1/4 inch wide, was soldered to the end of the bicycle pedal, B. A similar strip of metal, C, was soldered to a metal table, D, which was in turn soldered to the face of the clock gear, E. These two metal strips, A and C, were given a tendency to spring apart at the ends, and were clipped together by a sliding wire loop, F, at either end. This connection is sufficiently flexible to center itself as the gear turns and makes unnecessary a precise alignment of the clock gear with the shaft of the pedal. The connection is easily demountable.

The base of the unit is furnished with two thumbscrews at the anterior end for adjusting the tilt of the apparatus necessary to keep the tubes in place and to prevent fluid from wetting the stoppers in the tubes.

This apparatus can be housed in any incubator having adequate interior dimensions and an A.C. electric outlet inside the oven. In our laboratory we use it in a home-made plywood oven which is insulated with rock-wool and heated by thermostatically controlled electric light bulbs. The inside dimensions of this oven are  $19 \times 15 \times 12$  inches, and the walls are 3 inches thick. An inexpensive electric temperature control, manufactured by the Lyon Electric Company, San Diego, California, called the Lyon T 22 Breaker, is used. An A.C. outlet is installed in the back wall of the oven into which the roller tube apparatus is plugged.

This unit is silent in operation and produces no vibration. The synchronous motor assures a constant speed. The single unit construction allows the apparatus to be easily transported; it can be used in or out of the incubator as desired and can be changed from one place to another in a few seconds. The total cost of materials used in constructing the apparatus, including the motor, should not exceed seven dollars.

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