An Apparatus made from Meccano Parts for the Preparation of Roll-tube Cultures

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SUMMARY: Equipment can be readily made in the laboratory for the mechanical preparation of roll-tubes. Roll-tubes so prepared can largely replace Petri dishes both for counting and for single colony isolations.

The recent shortage of Petri dishes has compelled the adoption of an alternative method for counting viable bacteria. The roll-tube method, which consists essentially in the replacement of Petri dishes by test-tubes, was chosen (Wilson, 1922). Apart from considerations of accuracy, rolled tubes are more economical than Petri dishes since ordinary test-tubes may be used; only one-fifth of the medium is required and the number of dilutions is reduced since it is customary to use a standardized dropping pipette to deliver a known volume of inoculum into the tube. Against this must be set the disadvantage that streaking and picking of individual colonies requires a greater manipulative skill than with Petri dishes.

Withell (1938) obtained a standard error of $\pm 5\%$ with roll-tube counts. Using similar statistical methods Jenninson & Wadsworth (1940) obtained a similar figure for the plate method; with a reasonable number of replicates and only three dilutions the standard error was $\pm 6.6\%$.

Uniform roll-tubes are difficult to prepare without mechanical aid. Thompson (1934) described an apparatus for making them, and a successful commercial model was apparently obtainable in America before the recent war (Prouty, Bendixen & Swenson, 1944). Based on published photographs of this model, an apparatus was constructed from Meccano parts. It is simple in design, ordinary test-tubes (1.5×25 cm.) with cotton-wool plugs are used, and it has proved successful in daily use for 3 months.

THE APPARATUS

Test-tubes containing 2 ml. of melted-agar (2%) medium are inoculated and placed in the machine as shown in Pl. 1, fig. 1. The whole machine is inclined from front to back at an angle of 9° so that the melted agar remains in the bottom 4 cm. of the tubes. The tubes are then rotated. The agar creeps up to within 2 cm. of the cotton-wool plug and sets in about 2 min. Friction between the hinged front plate *D* and the cotton-wool plug reduces the speed of rotation from 950 r.p.m. (unloaded apparatus) to 650 r.p.m. when fully loaded with the four tubes.

The general arrangement of the equipment may be seen in Pl. 1, figs. 1 and 2. The rollers which cause the tubes to rotate and the main shaft are carried on a substantial framework. The frame itself is simple and consists of right-angled

girders made rigid with base plates and cross-girders. The apparatus measures 2 ft. $\times$ 5½ in. at the base and is 12 in. high.

The bearings. Each tube rests on four wheels of which the two pairs of driving wheels *F* (Pl. 1, fig. 2) are fixed on their spindles, while the others are free running. Each pair of driving wheels, which have rubber tyres (a strip of bicycle inner-tubing) to ensure adequate friction, rotates two tubes in an anticlockwise direction. The idler wheels on arm *G* are set at such an angle to the axes of rotation of the driving wheels that the tubes tend to thrust against the back plate *E*, instead of the hinged front plate *D*. Friction against the front plate *D* must be minimized to avoid undue loss of speed and to prevent formation of large amounts of cotton-wool dust. Some cotton-wool dust is inevitable and for this reason the rubber-band drive is preferable to a geared drive.

Method of siting the tubes

Each tube is supported on four wheels and enclosed front and back by metal plates. If the apparatus is perfectly adjusted the hinged front plate *D* becomes unnecessary, but this condition is difficult to maintain in practice.

The tubes are held in contact with the rollers by means of the spring-loaded idler wheels *H*₁–*H*₄, carried on the hinged arm *G* which is loaded with a 200 g. weight (see Pl. 1, figs. 1 and 2). These idler wheels are Meccano pulleys no. 23 fitted with rubber tyres. They are under tension by the action of small compression springs *I*₁–*I*₄, and held in position with a collar *K*. The fixing screw *L* of collar *K* is sufficiently long to enable the screw and collar to slip behind another screw *M*. This arrangement prevents lateral movement of the idler wheels but allows free movement in the vertical plane. Contrary to expectation, it was found in practice that best results were obtained if the tension spring *I*₁ was stronger than the other tension springs.

The arm *G*, which is mounted on a plate *N*, has a sliding adjustment so as to allow the idler wheels *H* to be brought centrally over each tube.

The drive. The drive is clearly shown in Pl. 1, fig. 2. Pulley wheel *A* transmits the power from a 1/5 h.p. motor to pulleys *B* and *C*. The drive to the spindle of the rollers is effected by means of rubber bands attached through a right-angle turn.

The counter. The colonies are counted in the apparatus shown in Pl. 1, fig. 3, which is a slowly rotating support for the tubes set against a black background.

Streak tubes can readily be prepared by means of this equipment. The bottom of a roll-tube containing the sterile medium is gripped in the clip, the tube is rotated (at a greater speed than for counting) and after removing the plug the charged loop is inserted and slowly withdrawn in contact with the agar when a spiral streak will result.

Fig. 1

Fig. 3

Fig. 2

A. HIRSCH—AN APPARATUS MADE FROM MECCANO PARTS FOR THE PREPARATION OF ROLL-TUBE CULTURES. PLATE I

REFERENCES

- JENNINSON, M. W. & WADSWORTH, G. P. (1940). Evaluation of the errors involved in estimating bacterial numbers by the plating method. *J. Bact.* **39**, 389.
- PROUTY, C. C., BENDIXEN, H. A. & SWENSON, S. P. (1944). A comparison of the roll-tube and standard plate method of making bacterial counts in milk. *J. Milk Tech.* **7**, 5.
- THOMPSON, R. (1934). A mechanical device which prepares and inoculates rolled tubes. *J. Bact.* **28**, 279.
- WILSON, G. S. (1922). The proportion of viable bacteria in young cultures with special reference to the technique employed in counting. *J. Bact.* **7**, 405.
- WITHELL, E. R. (1938). The evaluation of bactericidal action. *Quart. J. Pharm.* **11**, 736.

EXPLANATION OF PLATE

- Fig. 1. The apparatus with four roll-tubes in position.
- Fig. 2. The apparatus with the arm bearing the idler wheels raised.
- Fig. 3. Slowly rotating holder for counting roll-tube cultures.

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