A Simple Apparatus for Making Holes in Agar Plates

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The cup-plate method for vitamin and antibiotic assays is nowadays widely used because of its simple technique and the satisfactory results obtained. Nevertheless, when the number of samples is very large considerable time is taken to make the numerous holes in the agar plates by means of the cork-borer technique. Some mechanical apparatuses combined with suction have been tested, but without satisfactory results as the holes produced are not sufficiently sharp-edged and clean-cut. Moreover, the agar surrounding the holes is often lifted from the bottom of the plate due to the suction.

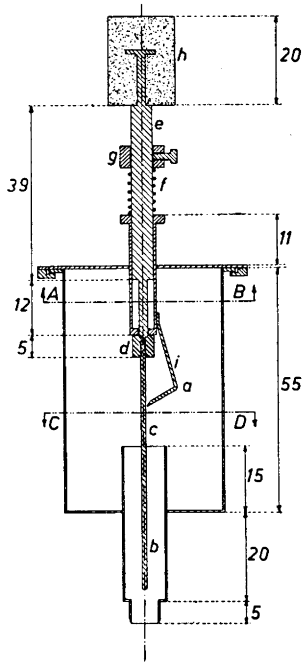

Fig. 1.

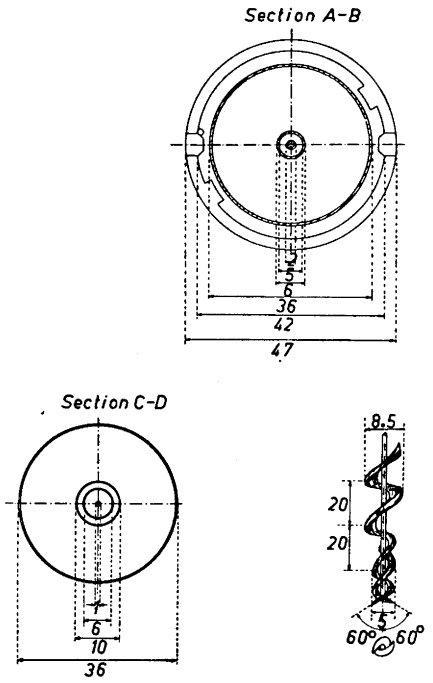

Fig. 2.

The apparatus described below does not require any suction, has a simple design, makes sharpedged and clean holes. It is easy to keep clean and does not need the help of any other device, which provides a high degree of independence in its use. The best material for the construction of this apparatus is stainless steel.

Description. The apparatus consists of two separable parts, part I and part II.

Part I. The shape and the dimensions (in mm) can be seen in Fig. 1. This part consists of the cylinder (a) in which the pieces of agar are collected as they are taken up from the plate, and the cylinder (b) which cuts the agar and brings the pieces of agar into the cylinder (a) by means of a helical conveyer. For the sake of simplicity the helical conveyer does not appear in Fig. 1 but is seen in detail in Fig. 2.

Part II consists of the lid and the following parts attached to it:

(c) Shaft of the helical conveyer.
(d) Device to join the shaft in such a way that the shaft or helical conveyer

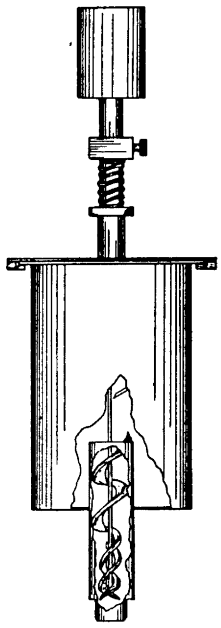

Fig. 3.

can be changed if it breaks or becomes defected during use. Part (d) acts also as a stop for the piston on its way up.

(e) Piston.

(f) Spring which brings the piston up automatically, once the hole has been made. This movement allows a more perfect function of the apparatus because it cleans the terminal and narrowest part of the cylinder (b).

(g) Movable part (attached to the piston by a screw) which acts as a stop for the piston on its way down. It permits adjustment of the effective length of the piston thereby bringing the ends of the helical conveyer into the most favourable working position.

(h) Knob.

(i) Wiper to remove the pieces of agar attached to the shaft by making them fall into cylinder (a).

Procedure. The apparatus is placed on the agar plate and pushed down vertically to cut the agar. The piston is pushed down and turned simply by rotating the knob between the thumb and the forefinger. In this way the pice of agar is taken up by the helical conveyer. On releasing the downward pressure on the screw-cap the piston readily slides up into its initial position.

Capacity. 100–120 holes/min.

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Conjugate Additions of Grignard Reagents to alpha, beta-Unsaturated Esters IV. Cuprous Chloride Catalysed Additions to Ethyl Esters

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Additions of Grignard reagents to *sec*-butyl esters of α,β-unsaturated acids, such as crotonic, tiglic and cinnamic acids, have been found to usually take place at the double bond (1,4-addition) [1] whereas, in general, additions to the corresponding ethyl esters take place predominantly at the carbonyl group (1,2-addition). However, in certain cases 1,4-addition to *sec*-butyl esters does not occur to any appreciable extent. This is for instance the case in some recently described [2] additions of methylmagnesium bromide, but it was found [2,3] that the presence of cuprous chloride during the addition of the esters to the Grignard reagent would secure fair to good yields of 1,4-addition products. It was further found that these yields were obtained not only with *sec*-butyl esters but also with ethyl esters. Crotonic esters thus gave the corresponding *iso*valeric esters, whereas tiglic and cinnamic esters gave saturated ketones as results of both 1,4- and 1,2-addition.

The present investigation was carried out as a consequence of the above-mentioned findings in order to establish if 1,4-addition products would also with higher Grignard reagents be the major products from ethyl esters in the presence of cuprous chloride, thus avoiding the use of *sec*-butyl esters. As model reactions the additions