TECHNICAL METHODS

A SLIDE METHOD FOR DEMONSTRATING SOLUBLE HAEMOLYSIN

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The following method of testing the haemolytic streptococcus for soluble haemolysin has been found to give good results, and no indefinite results (as are sometimes found by the tube method) have been encountered. The main advantage of the method is that positive results are easily read and are not screened by undissolved red cells, as occasionally happens in the tube method, thus making the readings difficult.

Method

The streptococcus to be tested is subcultured in 20% serum broth and incubated overnight. A conveniently sized loopful of the culture is placed on a 1/4 in. square (No. 1) coverslip and mixed with a loopful of normal horse cells.

The mixture is mounted by touching the drop with a slide as in preparing ordinary wet film. Finally the coverslip is sealed all round with vaseline and incubated at 37°C. A positive result is indicated by a complete transparency of amber colour in about three hours as a rule, whereas in a negative result the preparation remains opaque (see Fig.). Late afternoon preparations may be incubated overnight. The results are judged simply by holding up the slide to the light.

The best results are obtained with a proportion of two or three volumes of cells to one volume of culture, but exact amounts are not essential. As a larger volume of cells than of the broth culture is normally picked up by the loop, one loopful of each works well in practice. (The normal horse blood is undiluted, that is, as it is used for adding to nutrient agar for making blood-agar plates.)

Sealing Tool for Wet Preparations.—A quick and effective tool for sealing off wet preparations was devised in the following manner.

A piece of soft metal about 1/4 in. wide and 3 1/2 in. long by 1/32 in. thick was obtained and bent with pliers into a square with sides approximately 1/6 in. (For the metal strip a faeces spoon was used.) A metal swab stick was suitably bent and a handle soldered on vertically: or holes could be drilled and the ends of the swab stick bent and inserted. To use the device it is only necessary to hold the handle and heat the square of metal in a bunsen flame for a few moments. Then the bottom edge of the heated metal is pushed into a petri dish of vaseline, removed, and held in such a way as to touch or almost touch the perimeter of the coverslip. The melted vaseline then runs on to the glass and forms an effective and neat seal.

Photograph of slide showing negative (left) and positive results, and (below) the tool described in the text.
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