

## TECHNICAL METHODS

### A Micro Method for Detecting Indol Formation

NICHOLAS KOVACS

From the Public Health Laboratories, Perth,  
Western Australia

(RECEIVED FOR PUBLICATION JUNE 14, 1958)

The acid in the commonly used indol reagent described by Kovacs (1928, 1932) and *Diagnostic Procedures and Reagents* (1950) changes the reaction of the medium in which the indol production is tested and prevents the further observation of biochemical processes, killing the bacteria themselves.

It was found that, by heating 0.1 ml. or less of culture (broth or peptone water), indol production by enteric bacteria could be easily detected. Four capillary drops of the fluid medium to be tested are put into a test tube of about 12 by 75 mm. and 4 drops of the indol reagent are added from a dropping bottle. The mixture is shaken lightly and if it does not turn red immediately it should be heated gently for a moment, without actual boiling, in the flame of a Bunsen burner. Heating doubles the sensitivity of the reaction seen at room temperature.

For routine work the culture fluid which is to be tested is collected with a spiral-ended wire. This is a simplified version of the one used by Takátsy (1955) for virus work and of the volumetric platinum cylinder described by Berridge (1954). Both are manufactured. The cylindrical spiral can be easily made in the laboratory from a 22 gauge platinum wire by turning eight coils around a 1 in. by 5/32 in. Whitworth metal thread steel screw (about 4 mm. diameter). When preparing the spiral  $\frac{3}{8}$  in. (1 cm.) of wire is left straight at the commencement. When the spiral has been wound, this short end is bent back with forceps into the centre so as to form an axis (Fig. 1). This axis enables the spiral loop to hold the contents more easily. The coils are now pressed together with the forceps so that they are tightly closed. The spiral loop contains about 0.08 to 0.09 ml. of water.

The use of spiral wires dispenses with Pasteur pipettes and saves considerable time and material.



Fig. 1

### Method

Four drops of the indol reagent are filled from a dropping bottle into a Kahn tube. The flame-sterilized spiral wire is loaded with the culture fluid to be tested for indol, and, while holding the tube almost horizontally, the contents of the wire are washed off by rotating the spiral in the reagent. When the indol reaction is not immediately apparent, the tube is heated carefully over the Bunsen burner for one or two seconds only and the reaction observed. In serial examinations, when many cultures are tested in succession for indol formation, the indol reagent is added to a series of Kahn tubes and two or three spiral wires are used alternately as they take much longer to cool than ordinary loops.

The micro-method of indol determination with platinum spiral wire and heat proved a useful and simple test, especially in mass examination of cultures from faeces, egg-pulp, Moore swabs, etc.

### REFERENCES

- Berridge, N. J. (1954). *J. appl. Bact.*, **17**, 15.  
*Diagnostic Procedures and Reagents*, 3rd ed. (1950). Amer. Publ. Hlth Ass., New York.  
Kovacs, N. (1928). *Z. Immun.-Forsch., Orig.*, **55**, 311.  
— (1932). *Zbl. Bakt. I. Abt. Orig.*, **123**, 391.  
Takátsy, Gy. (1955). *Acta microbiol. Acad. Sci. hung.*, **3**, 191.

### A.C.P. Broadsheets

All the broadsheets published to date will cost 1s. 3d. post free each, with a reduction on orders for 25 or more.

### Correction

We regret that there is an error in the paper, "The Antibody Content of Single Cells" (November, 1958, vol. 11, No. 6, p. 544, column 1, line 14). Instead of "an average of 7.8 grains in stripping film," it should read "an average of 1.8 grains in stripping film."