

# An Appraisal of the Uricult Dip-Slide Method in the Diagnosis of Urinary Infections\*

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## SUMMARY

Uricult dip-slide urine cultures were compared with standard laboratory plate cultures. Good agreement of bacterial counts was obtained after incubation at 37°C but not at room temperature. Tests of therapeutic activity of various drugs on the commoner organisms producing urinary infections, were done. The results suggested that such tests had no positive value. Uricult dip-slides should be of value as a suitable transport and diagnostic medium for the diagnosis of urinary tract infections.

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Numerous reports assessing the value of the Uricult method in the diagnosis of urinary infections have been published since this procedure was first used in 1965.<sup>1-6</sup> As there appeared to be considerable divergence of views on certain aspects of the recommended procedure, it was decided to carry out a comparison of the Uricult method with the standard quantitative technique as used in this hospital. The following points were especially considered: (i) the value of urine testing on the Uricult dip-slide; (ii) the difference in growths obtained at 37°C and at room temperature; (iii) the difference in the growths obtained on the different media on the Uricult dip-slides; (iv) human error in the use of the Uricult technique; (v) the use of the dip-slide in urine antibiotic sensitivity tests.

## METHODS

The Uricult dip-slides provided were plastic slides coated on one side with 10 cm<sup>2</sup> of MacConkey agar and on the other side with 10 cm<sup>2</sup> Kled agar. The MacConkey agar used appears to inhibit the growth of Gram-positive cocci and is therefore a selective medium for Gram-negative bacteria.

Fresh mid-stream urine specimens submitted to the Hospital laboratories in sterile wide-neck containers were examined by dipping the slide into the urine, draining off excess and incubating at 37°C in the sterile, plastic container supplied by Boehringer Mannheim (S.A.) (Pty) Ltd. Two hundred and fifty urine specimens were examined comparing the Uricult technique and the standard colony count technique as used in this laboratory.<sup>7</sup> A further

200 urine specimens were examined by the Uricult method, both at room temperature and after incubation at 37°C.

In order to determine the value of sensitivity testing using the Uricult dip-slide, urines were inoculated with 10<sup>6</sup> per ml organisms of *E. coli*, *Klebsiella*, *Proteus mirabilis*, *Proteus vulgaris*, *Proteus rettgeri*, *Proteus morgani*, *Enterobacter*, *Salmonella*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus faecalis*. The strains of *Staphylococcus aureus* (NCTC 6571), *E. coli* (NCTC 10418) and *Pseudomonas aeruginosa* (NCTC 10662) were the standard strains recommended by Garrod and Waterhouse.<sup>8</sup> The other strains were selected typical stock test strains. After inoculation of the Uricult dip-slides by dipping in the infected urine, two 6 mm discs, each containing one of the antibiotics used routinely in this laboratory, were placed on the Kled medium, respectively 1.0 cm from the top and 1.0 cm from the bottom of the dip-slide. The antibiotic discs were also placed on the surface of Wellcotest lysed blood agar plates inoculated with the same urine. Results were read after overnight incubation at 37°C. Altogether 74 cultures from 15 different types of bacteria were tested.

## RESULTS

Using the charts supplied, it was possible to distinguish clearly the different bacterial densities. Differences were, however, frequently observed between the densities of growth in the two media on the dip-slides. A clinically significant bacteriuria was diagnosed when the colony count was 10<sup>5</sup> or greater.

Forty-eight of the 250 urine specimens gave a growth of 100 000 or more organisms on both Uricult and colony count, while 8 specimens gave a growth of 100 000 or more on the dip-slides but  $\pm$ 80 000 on the colony count. It was considered that this difference was within experimental error for the method used and that therefore the 56 specimens of urine showed an agreement in the bacterial count, assessed by either the Uricult dip-slide or the colony count.

The majority of these specimens were tested both by the dip-slide technique and the standard laboratory procedures on the arrival of the specimen at the laboratory, usually about 4 hours after voiding of the urine. Fifteen specimens, however, were examined by the dip-slide technique immediately after voiding, and in the laboratory by the standard technique, 4 hours later. In all cases, except one, there was complete agreement between the

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findings obtained. In this one case the dip-slide count was higher than the colony count.

Sixty specimens showed a Uricult count between  $10^4$  and  $10^5$ . Of these, 38 also had a colony count done and in 8 cases there was disagreement in the counts obtained by the two methods. In most cases disagreement was due to a higher count on the colony count than on the dip-slides. In no case, however, did the count approach  $10^5$ .

Although it has been generally accepted that a colony count of  $10^5$  or over should be regarded as of pathological significance, our analysis of the 60 specimens showing a Uricult figure of  $10^4$  -  $10^5$  indicated that 38 of these results would agree with a clinical history of urinary infection and in most cases showed a cellular pathology indicating urinary infection. Although none of the specimens in this series on which colony counts were done, showed a count of  $10^5$  or more; in 18 of the specimens examined the colony count was 30 000 or over.

It is our impression that cognizance should be taken of a Uricult count of between  $10^4$  and  $10^5$ , especially if there is clinical evidence suggestive of a urinary infection.

Four hundred Uricult dip-slides from 200 urine samples were used in a parallel comparison of incubation at  $37^\circ\text{C}$  and at room temperature. Room temperature was measured by means of a maximum and minimum thermometer, and during the month of September was found to vary between  $16,3^\circ\text{C}$  and  $22,7^\circ\text{C}$ . Of the 200 urine samples 44 gave a Uricult reading of  $10^5$  or more at  $37^\circ\text{C}$ . However, only 26 of these specimens showed a growth of  $10^5$  or  $>10^5$  organisms per ml urine after overnight incubation at room temperature. It was observed that the majority of specimens which gave unreadable growths or no growths on the Uricult dip-slide after 18 hours at room temperature, gave a readable growth after further incubation in the shirt or waistcoat pocket for 5 to 6 hours. In the case of unreadable growths, difficulty arose in these readings in determining whether the fine appearances present on the Uricult dip-slide were from urinary deposits or the result of bacterial growth.

The two media used, MacConkey agar and Kled agar proved selective for most of the organisms found in infected urines, i.e. *E. coli*, *Proteus spp.*, *Klebsiella*, *Streptococcus faecalis* and *Staphylococcus aureus*. Kled medium, however, in a high percentage of cases, particularly in contaminated urines, showed a heavier growth than the MacConkey agar. This is not surprising in view of the ability of this medium to facilitate the growth of lactobacilli, diphtheroids and Gram-positive cocci. The presence of these organisms is usually an indication of contamination. When present, the true estimate of bacteriuria should be from the colony count on the MacConkey agar. In a minority of cases, Gram-positive cocci will be detected by growth on the Kled medium.

A number of tests were carried out in which the dip-slide technique was used by non-technical staff after demonstration of the method of inoculation. It was clear that with due attention to drainage of the dip-slide, the human error factor was negligible and could be disregarded.

Following the suggestion of Bailey *et al.*,<sup>9</sup> that the Uricult dip-slide with Kled medium be used for rapid

antibiotic sensitivity testing of urinary organisms, tests were carried out on sterile urine samples inoculated with cultures of typical *E. coli*, *Proteus rettgeri*, *Klebsiella*, *Proteus mirabilis*, *P. vulgaris*, *P. morgani*, *Enterobacter*, *Salmonella*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus faecalis*. Each urine sample contained a concentration of bacteria between  $10^5$  and  $10^6$  per ml.

The therapeutic agents tested were those used as a routine in this laboratory, viz. ampicillin, streptomycin, kanamycin, Furadantin, gentamicin, cephalosporin, nalidixic acid, Nicene, trimethoprim sulphamethoxazole and carbenicillin.

Tests were carried out on the various organisms when grown on (i) the Kled medium on the Uricult dip-slide, and (ii) Wellcotest sensitivity agar as recommended by Garrod and Waterhouse.<sup>8</sup>

A clear zone of 10 mm diameter or more was accepted as evidence of sensitivity of the organisms to the chemotherapeutic substance when tests were carried out on Wellcotest agar. As the zone of clearance on the Kled agar on the Uricult dip-slides was invariably smaller than on the Wellcotest agar, a clear zone of 8 mm in diameter was accepted as evidence of sensitivity. (In practice no smaller zones were observed.)

Analysis of the results of sensitivity tests carried out on 74 bacterial cultures showed marked variations in the results obtained. In many cases the Kled medium growths showed apparent sensitivity to an antibiotic, while no such sensitivity was observed on the Wellcotest medium. In the case of other cultures, the reverse phenomenon was observed. These irregular results were particularly noticeable when ampicillin and trimethoprim sulphamethoxazole were tested. Our preliminary tests on 10 cultures of the various strains suggested that the Uricult-Kled medium might have some value in antibiotic testing. Subsequent tests, however, showed so many irregularities that we could not place any reliance on the results obtained. It should be generally accepted that sensitivity tests have to be performed on special media which do not interfere with the therapeutic agents. Media developed for diagnostic purposes, like MacConkey or Kled, should not be used for sensitivity tests.

## DISCUSSION

Our results are in agreement with those obtained by other workers<sup>1-6</sup> in so far that bacterial counts made by the dip-slide technique showed complete agreement with the colony count technique when the dip-slide was incubated at  $37^\circ\text{C}$  overnight.

Unlike Arneil *et al.*<sup>4</sup> and Bailey *et al.*,<sup>9</sup> we did not find agreement between counts done by the two methods when incubation was at room temperature, i.e.  $16,3^\circ\text{C}$  to  $22,7^\circ\text{C}$  overnight. In this regard we are in agreement with Willie *et al.*<sup>5</sup> Incubation at these temperatures overnight followed by incubation in the shirt or waistcoat pocket at  $\pm 37^\circ\text{C}$  for a further 4 to 5 hours gave readable growths on the dip-slide, which were in agreement with the colony counts incubated at  $37^\circ\text{C}$ .



When the dip-slide showed a growth of between  $10^4$  and  $10^5$  at  $37^\circ\text{C}$  we found agreement with the colony count in less than two-thirds of the specimens. In most of these cases there was a history of urinary infection and/or cellular pathology.

We are of the opinion that the dip-slide technique is a reasonably accurate and rapid method of determining the presence of bacteriuria when incubated at  $37^\circ\text{C}$ . We accept the criterion of Cohen and Kass<sup>2,10</sup> that counts of  $10^5$  or more should be regarded as of pathological significance. We are of the opinion that where laboratory services are not available, the clinician using the dip-slide technique may regard such counts as highly suspicious of infection, especially if associated with clinical evidence and/or cellular pathology.

As over 50% of the infected urines examined in this laboratory showed a growth of *E. coli* and a further 40% a growth of other Gram-negative bacteria, in particular *Proteus spp.*, *Klebsiella*, and *Enterobacter*, it is our opinion that the growth on the MacConkey medium is of more significance than is that on the Kled medium. Occasionally a heavy growth was obtained on the Kled medium and not on the MacConkey medium. In these cases contamination with lactobacilli, diphtheroids and cocci was the usual cause. The MacConkey medium inhibited growth of Gram-positive cocci, including *Streptococcus faecalis* and *Staphylococcus aureus*.

Although the technique of inoculation of the dip-slide is exceedingly simple, we consider that proper drainage must be emphasized and that the slide must be placed in the container immediately after inoculation and drainage.

The possible use of the dip-slide Kled medium as an indicator of therapeutic activity, suggested by Bailey *et al.*,<sup>9</sup> is of particular interest to us. Our laboratory examines specimens from two hospitals, and a technique which would enable not only rapid growth to be obtained from an infected urine specimen, but also a speedy indication of the appropriate therapeutic agent would be of advantage.

We therefore carried out a series of experiments with the routine therapeutic agents used in this laboratory, i.e. ampicillin, streptomycin, kanamycin, Nitrofurantoin, gentamicin, cephaloridine, nalidixic acid, Nicene, trimetho-

prim sulphamethoxazole, and carbenicillin. The bacterial strains used were grown on Wellcotest agar as suggested by Garrod and Waterhouse<sup>8</sup> and the concentrations of therapeutic agents used were those recommended by the same authors. Nicene was tested at a concentration of  $50\ \mu\text{g/ml}$ , and trimethoprim sulphamethoxazole at a concentration of  $25\ \mu\text{g/ml}$ .

The results of tests carried out on 74 different cultures from 15 types of bacteria, showed such a degree of variation that no reliance could be placed on sensitivity tests carried out on the Uricult dip-slide Kled medium. In view of the many and complex factors which may affect the suitability of a culture medium for the testing of antibiotic sensitivity, and which have led to the development of special media, such as Wellcotest agar, this is not surprising.

Our results indicate that the use of the Uricult dip-slide Kled agar for this purpose is not to be recommended.

In the case of the country practitioner where laboratory services are not readily available, the Uricult dip-slide could perform a useful function under the conditions of incubation outlined above. We do not consider that the dip-slide technique should take the place of a full laboratory examination of the urine when such facilities are available. The Uricult dip-slide is in our opinion an excellent and time-saving transport medium for urine cultures, combining as it does both transportation of the specimen and growth of the bacterial content, if the temperature during the transport period allows the organisms to multiply.

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#### REFERENCES

1. Gultman, D. and Naylor, G. R. E. (1967): *Brit. Med. J.*, **3**, 343.
2. Cohen, S. N. and Kass, E. M. (1967): *New Engl. J. Med.*, **277**, 176.
3. McFadyen, I. R. and Eykyn, S. J. (1971): *Lancet*, **1**, 132.
4. Arneil, G. C., McAllister, T. A. and Kay, P. (1970): *Ibid.*, **1**, 119.
5. Willie, L., Scharer, K. and Bickel, M. (1970): *Ibid.*, **2**, 105.
6. Arneil, G. C., McAllister, T. A. and Kay, P. (1970): *Ibid.*, **1**, 364.
7. Franke, S. and Reitman, S., eds. (1963): *Gradwohl's Chemical Laboratory Methods and Diagnostics*, vol. I, p. 513. St Louis: C. V. Mosby.
8. Garrod, L. P. and Waterhouse, P. M. (1971): *J. Clin. Pathol.*, **24**, 779.
9. Bailey, M. J., Neary, J. T. F. and Notelovitz, M. (1972): *S. Afr. Med. J.*, **46**, 1323.
10. Leading Article (1964): *Lancet*, **1**, 77.