areas of old infarction. This figure is in close agreement with that of Johansson (1966), who, on clinical criteria, suspected that only 24 out of 142 cases (17%) of chronic heart block were ischaemic in origin. He offered no pathological explanation for 64 out of 142 cases (45%) which were classified as "primary" heart block. In the present post-mortem series 21 out of 53 cases (40%) were due to bundle branch fibrosis. The pathological basis of primary heart block is therefore bilateral bundle-branch fibrosis; the cause remains unknown, but it is certainly not due to coronary artery disease.

Summary

In a post-mortem study of 53 hearts from patients with chronic atrioventricular block only 8 (15%) were due to coronary artery disease. Bundle-branch fibrosis of unknown aetiology, without major damage to the contractile myocardium, is the commonest cause, being present in 21 cases (40%).

In 14 cases of acute myocardial infarction and A-V block major structural damage to the conduction system was unusual, but, when present, involved the bundle branches.

This work was done during tenure of the William Shepherd Research Fellowship from the Board of Governors of St. George's Hospital. We would like to thank Dr. A. Leatham and Dr. W. B. Robertson for their help in preparing this paper.

REFERENCES

- Crawford, T., Dexter, D., and Teare, R. D. (1961). *Lancet*, 1, 181.
 Davies, M. J. (1967). *J. Path. Bact.* In press.
 Eusterman, J. H., Achor, R. W. P., Kincaid, O. W., and Brown, A. L., jun. (1962). *Circulation*, 26, 1288.
 Gilchrist, A. R. (1958). *Scot. med. J.*, 3, 53.
 Harris, A., and Bluestone, R. (1966). *Brit. Heart J.*, 28, 631.
 Johansson, B. W. (1966). *Acta med. scand.*, 180, Suppl. No. 451.
 Penton, G. B., Miller, H., and Levine, S. A. (1956). *Circulation*, 13, 801.
 Portal, R. W., Davies, J. G., Leatham, A., and Siddons, A. H. M. (1962). *Lancet*, 2, 1369.
 Zoob, M., and Smith, K. S. (1963). *Brit. med. J.*, 2, 1149.

Dip-Slide: An Aid to Quantitative Urine Culture in General Practice

DENNIS GUTTMANN,* M.A., M.B., B.CH., B.SC., M.R.C.P.; G. R. E. NAYLOR,† M.A., M.D., M.R.C.P., M.C.PATH.

Brit. med. J., 1967, 3, 343-345

Since Kass (1956) defined criteria of significant bacteriuria, the use of quantitative assessments of the bacterial population as an aid to the differentiation between infection and contamination of the urine has gained wide acceptance. The pour-plate method is recognized as the standard technique for performing viable counts on urine, but requires trained staff for setting up the cultures. If misleading results are to be avoided, urine specimens must be cultured within an hour or two of collection, or be refrigerated; in general practice it is often difficult to fulfil these requirements. Further, the examination of large numbers of specimens, as at antenatal clinics and in population surveys, makes demands on laboratory services which may become prohibitive if the pour-plate method is employed routinely. A number of simpler cultural methods have been described, such as the filter-paper strip inoculation technique of Leigh and Williams (1964), and the semiquantitative methods of Cattell and Lefford (1963) and of Guttman and Stokes (1963). Though these are technically less complicated, they are still dependent on the delivery to the laboratory of freshly passed specimens.

Mackey and Sandys (1965, 1966) described a dip-inoculum transport medium in which a metal or plastic spoon carries the culture medium, and which may be inoculated by simply dipping in the specimen, after which it need not reach the laboratory for incubation for 24 or 48 hours. Using standard materials readily available in most laboratories, we have investigated a similar method which we feel has a number of advantages. An ordinary 3- by 1-in. (7.5- by 2.5-cm.) glass microscope slide is coated for a length of about 2 in. (5 cm.) with nutrient agar on one side and with MacConkey's agar on the other. It is held in a small cylindrical aluminium container as supplied in the Public Health Laboratory Service for the collection of faeces specimens. This dip-slide is inoculated by dipping the coated end into the fresh urine specimen. After allowing excess urine to drain for a few seconds, the slide is returned to its container and after replacing the screw cap it is sent to the

laboratory, if necessary by post. After overnight incubation the number of colonies which have grown on the slide is counted with a hand lens. The preparation is cheap enough to be discarded after use, and an extensive trial has shown the method to give clinically valuable results. Fig. 1 shows the dip-slide and its aluminium container. Full details of the preparation of the dip-slides are given elsewhere (Naylor and Guttman, 1967).

Dilutions of broth cultures of Gram-negative rods and Gram-positive cocci isolated from pyogenic infections of the urinary tract were examined by dip-slide inoculation and simultaneous viable bacterial counts: we found a straight-line relationship between the number of colonies on the dip-slide and the number of organisms in the fluid into which it had been dipped; 50 to 100 colonies on the dip-slide correspond to a viable bacterial count of 10,000 per ml. (Naylor and Guttman, 1967). Fig. 2 shows a series of dip-slides prepared from dilutions of a culture of *Escherichia coli*, and it will be seen that separate colonies readily countable with a hand lens were obtained up to a concentration of 0.6×10^5 organisms per ml. The results of a clinical trial are reported in more detail below.

Methods

From outpatients attending a urinary infection clinic 385 consecutive mid-stream urine specimens were obtained and examined in parallel by the dip-slide, a pour-plate viable count, and the semiquantitative method of Guttman and Stokes (1963). The last 100 specimens were inoculated on to dip-slides with cycloheximide added to the nutrient agar at a concentration of 100 µg./ml. of medium, as well as on to ordinary dip-slides. Cycloheximide prevents the growth of air-borne mould spores which occasionally were found to contaminate some of the slides during preparation, thus creating difficulties if the dip-slides were stored for some weeks before use. Such a concentration of cycloheximide did not interfere with the growth of bacteria isolated from urine specimens.

Microscopy of the Gram-stained deposit and tests for protein were performed, and white cell excretion rates were estimated by

* Addenbrooke's Hospital, Cambridge.

† Public Health Laboratory Service, Cambridge.

the method of Houghton and Pears (1957), the patients having been instructed to empty the bladder two to three hours before attending the clinic. Preparation of female patients consisted of washing the vulva and perineum with soap and water and drying with a clean towel. Where possible the vaginal orifice was plugged with cotton-wool. A clean catch mid-stream specimen was obtained for bacteriological examination, which was subsequently combined with the remainder of the voided

FIG. 1.—The dip-slide.

FIG. 2.—Dip-slides inoculated from tenfold serial dilutions of a culture of *E. coli*.

bladder contents for estimation of white cell excretion rates. All specimens were processed within three hours of collection, those which could not be cultured within one hour being refrigerated meanwhile.

All patients attending the urinary infection clinic had evidence of past or present infection of the urinary tract, and over half the specimens were obtained from patients receiving chemotherapy.

Results

Forty-two specimens were found to contain more than 10^6 organisms per ml., and all these gave a confluent growth on both sides of the dip-slide, except one specimen containing a heavy growth of a coagulase-negative staphylococcus, where no growth was obtained on MacConkey's agar. Fifty-two specimens contained fewer than 100 organisms per ml. and gave no growth on the dip-slide. Culture results on the remaining 291 specimens are shown in Fig. 3; these include 30 specimens containing fewer than 100 organisms per ml., but showing some growth on the dip-slide. The dip-slide colony counts refer to the side of the slide coated with nutrient agar, except where this was overgrown with swarming organisms when the side coated with MacConkey's agar was counted. It will be seen that there is good correlation between the pour-plate counts and the numbers of colonies grown on the dip-slides, the moderate degree of scatter causing no practical difficulty in the

interpretation of the results. Essentially similar results were obtained with Gram-negative rods and Gram-positive cocci.

FIG. 3.—Comparison of dip-slide colony counts and viable bacterial counts by the pour-plate method. ○=Ordinary media. ●=Nutrient agar containing cycloheximide 100 µg./ml. (See text.)

In our series of 385 specimens there were 37 which contained between 10^4 and 10^5 organisms per ml., and produced dip-slide counts from 50 to 500 colonies. Six of this group, all from patients receiving chemotherapy, were found by routine methods to give pure cultures of pathogenic organisms, showed protein and increased white cell excretion rates, and were considered to reflect active urinary infection. These could be readily distinguished from heavily contaminated specimens by examination of the dip-slides, the purity or mixed nature of the growth being obvious at this concentration. We believe that there is little danger of missing an active infection of the urine by the dip-slide method even if the viable bacterial count is less than 10^5 organisms per ml. In 4 out of the remaining 31 specimens containing 10^4 to 10^5 organisms per ml. which by routine methods were considered to be heavily contaminated, the dip-slide was wrongly interpreted as indicating true infection; the incidence of false-positive results is therefore acceptably small when the whole series of 385 specimens is considered.

Dip-slide in General Practice

Dip-slides have been used by a number of general practitioners in the Cambridge area, the inoculated slides being sent to the Public Health Laboratory for incubation and reporting. No practical difficulties were experienced and the culture results have been clinically helpful. They have been particularly useful when urine specimens have had to be sent by post, and also when patients attended evening surgeries held after normal laboratory working hours.

A number of patients attending the urinary infection clinic were asked to inoculate dip-slides at home. They were instructed to dip the slide into a mid-stream specimen passed, after perineal toilet, into a basin rinsed with boiling water and allowed to cool, or to wet both sides of the dip-slide in the urine stream while holding the uncoated end in a clothes peg. Preliminary tests showed that the media adhere to the slide firmly enough not to be washed off when the preparation is inoculated in this way. The dip-slide could also be inoculated by pouring urine over both sides if the specimen was collected in a vessel unsuitable for dipping. No difficulty was experienced with excessive contamination of dip-slides inoculated by patients themselves. Home inoculation was found to be of particular value in patients who developed urinary symptoms between

visits to the clinic and were unable to attend for a specimen to be collected and examined by routine methods. The response of established infections to chemotherapy could also be followed on an outpatient basis, slides being inoculated on several consecutive days after starting treatment.

Discussion

The dip-slide is based on Mackey and Sandys's idea of a dip-inoculum transport medium. It is made from easily available materials and is cheap. It has two further advantages. Firstly, two media are inoculated simultaneously. This helps considerably in distinguishing between pure cultures of pathogenic organisms and mixed growths due to contamination. Secondly, a larger surface area of medium is available for inoculation. This makes colony counting easier, and the transparency of the preparation is also helpful in this respect. Up to 500 colonies can be easily separated on one side of the dip-slide, the growth becoming confluent only with counts approaching 1,000 colonies. Our clinical trial shows that all specimens containing more than 10^5 organisms per ml. gave dip-slide counts of 200 or more colonies, and specimens with more than 10^4 organisms per ml. produced counts above 40 colonies. Colony counts between 40 and 200 are particularly convenient to read.

We suggest that dip-slide colony counts between 100 and 500 should be regarded as doubtful and an indication for further investigation of the patient. In our series there were 45 specimens in this range, of which eight were considered to be truly infected. It must be remembered that our patients were highly selected, and it is probable that in a random population the incidence of doubtful results would be smaller.

The dip-slide has been in clinical use for over one year. In general practice it was found to overcome difficulties in bacteriological diagnosis which may arise when there is an unavoidable delay between collection of the specimen and its delivery to the laboratory. The ease of inoculation makes it useful in screening clinics where large numbers of specimens have to be processed. With a little experience the majority of dip-slides may be assigned to significantly or non-significantly infected groups by inspection without counting colonies. The quantitative accuracy of the method is greater than that obtained by standard-size loop inoculation methods; on the other hand, the identification of bacterial species and subculturing are easier than from a pour plate. In our series, which included 60

specimens shown to be infected by conventional methods, the dip-slide yielded no false-negative results, and false-positive results were also not a problem.

We regard the dip-slide as an adjunct to conventional methods of urine examination. It does not obviate the need for microscopy of the deposit, tests for protein, or cultures for the identification of the infecting organism. We do not suggest its introduction for routine use where specimens can be delivered to the laboratory quickly after collection. It may, however, be conveniently used as the only screening method in surveys such as at antenatal clinics. Patients from whom specimens are obtained with dip-slide colony counts in the infected or doubtful range would then be recalled for further investigation by routine methods.

Summary

A dip-inoculum transport medium for the diagnosis of significant bacteriuria is described, consisting of a microscope slide coated with culture media. Laboratory and clinical testing shows the dip-slide to give consistent quantitative results which compare well with pour-plate viable counting.

The dip-slide enables ready differentiation between heavy contamination of urine specimens and genuine infection. In a series of 385 specimens no false-negative results were obtained, and there was an insignificant number of false-positive cultures.

The dip-slide is recommended whenever there is likely to be delay in delivery of urine specimens to the laboratory. It is particularly useful in general practice if specimens have to be sent by post. Owing to the ease of inoculation and reading it is also suited to the routine screening of large numbers of specimens such as at antenatal clinics.

We wish to thank Miss I. J. Barker, Miss V. A. Lane, and Miss S. E. Minns for technical assistance, and the outpatient nursing staff of Addenbrooke's Hospital for their skill in obtaining satisfactory urine specimens. One of us (D.G.) is grateful to Dr. M. H. Gleeson-White for provision of laboratory facilities.

REFERENCES

- Cattell, W. R., and Lefford, M. J. (1963). *Brit. med. J.*, 1, 97.
Guttman, D., and Stokes, E. J. (1963). *Ibid.*, 1, 1384.
Houghton, B. J., and Pears, M. A. (1957). *Ibid.*, 1, 622.
Kass, E. H. (1956). *Trans. Ass. Amer. Physns*, 69, 56.
Leigh, D. A., and Williams, J. D. (1964). *J. clin. Path.*, 17, 498.
Mackey, J. P., and Sandys, G. H. (1965). *Brit. med. J.*, 2, 1286.
— (1966). *Ibid.*, 1, 1173.
Naylor, G. R. E., and Guttman, D. (1967). *J. Hyg. (Camb.)*, 65. In press.

Sodium Uptake in Peritoneal Dialysis

J. D. SWALES,* M.B., M.R.C.P.

Brit. med. J., 1967, 3, 345-347

Peritoneal dialysis has come to occupy a prominent place in the treatment of renal disease, and has, indeed, largely replaced haemodialysis in some centres (Cameron, 1966). Pulmonary complications frequently occur, though in many cases they consist of asymptomatic segmental collapse (Berlyne *et al.*, 1966). An occasional, and more often fatal, complication is acute heart failure. In a recent series of 91 patients this complication accounted for two deaths (Stewart *et al.*, 1966).

When dialysis fluid cannot be drained from the peritoneal cavity, fluid overload and heart failure are easily accounted for in terms of the slow absorption of dialysis fluid. However,

even where the patient is not in positive fluid balance, heart failure may occur during the course of peritoneal dialysis (see Case Report below). The purpose of this paper is to present data on sodium absorption during 13 consecutive peritoneal dialyses, to suggest that it is the uptake of sodium in hyponatraemic patients which is responsible for the development of heart failure, and to suggest how this potentially fatal complication may be avoided.

Case Report

A 14-year-old schoolboy gave a history of lifelong nocturia. For five years he had had polydipsia and polyuria with recurrent dysuria. In March 1965 he was found to have a blood urea of 450 mg./

* Registrar, Medical Professorial Unit, Westminster Hospital, London S.W.1