Cyst of Entamoeba histolytica stained with acridine orange. Tungsten illumination using dark ground condenser. ×400

fluorescence has been observed several weeks after preparation).

It is possible to use the high power dark ground condenser and a tungsten light source as the stain enhances the visual effect of the chromidial bar within the cyst.

AH MOODY
Hospital for Tropical Diseases
4, St Pancras Way,
London NW1 0PE

References

Bacteriological examination of removed cerebrospinal fluid shunts

This paper by Dr Bayston and his colleagues has been read here with interest and their method (A) should prove adequate as a ground plan for laboratories embarking on the detailed examination of internal prosthetic shunts, Hickman lines and similar devices suspected to be colonised. Method A resembles in many details the routine procedure adopted in this Hospital in 1969, since when some 600 complete ventriculo-caval shunts have been examined.

The understandable clinical euphoria which followed the successful implantation and function of ventriculo-caval shunts in 1957 had somewhat abated by 1960. By this time the author had examined several shunt systems "infected" by coagulase-negative staphylococci almost always accompanied by bacteraemia, and often by ventriculitis, caused apparently by the same organism. The investigational procedures were then relatively crude, none of us had had previous experience of such colonisation, and the author regrets that it was not until 1968 that he devised a more critical and searching method, early results of which were reported in 1970.

The complete shunt is placed in a sterile 6" (15-2 cm) Petri dish and sent immediately to the laboratory; it is first examined macroscopically and by plate microscope before the dish lid is opened. Any external pus or detritus is sampled separately with a swab moistened in sterile broth, and in all cases the whole length of the exterior of the shunt is similarly swabbed. Whenever practicable, the surgeon is encouraged to swab the tissue track immediately after shunt removal with a fine wire swab. The surface of each component of the shunt system is thoroughly treated with an iso-propanol swab and fluid aspirated from that site with a fine gauge needle and syringe. It is often necessary to flush the interior of the shunt with about 0.2 ml of sterile infusion broth. In most cases a sample of ventricular fluid, not taken through the proximal catheter, is collected, together with blood culture bottles inoculated at the time of surgery. All swabs and fluid are cultured aerobically and anaerobically on blood-agar plates, onto MacConkey plates, and most of the remainder is cultured in aerobic and anaerobic broth medium. A potent β-lactamase, either by Whatman or Merck, is added to all cultures when a β-lactam antibiotic has been administered to the patient. The β-lactamase is itself tested for sterility. A Gram film is made of each sample. The MacConkey plate is often valuable in distinguishing two or even three biotypes of Staphylococcus albus which may occasionally be concurrently present. All culture and subsamples are incubated for at least 7 days.

Finally, any thrombus or concretion, usually in the shunt lumen or round the tip of the distal catheter, is cut out, fixed in formol saline and thin sections prepared; these are stained by a conventional cytochemical method and by a histological Gram's stain.

Dr Bayston et al are so very right when they point out the twin risks of "mixed infection" and of overdosage in what they term the "conventional" method used in some inexperienced or unthinking departments, and they are to be thanked for demonstrating this so effectively.

It is a pleasure to acknowledge the high technical skills of Mr CH Frankcombe, who has shared the shunt investigations in this Hospital, and as always the unstinting support of our paediatric surgeons, Mr EH Eckstein and Mr DM Forrest and their surgical teams.

RJ HOLT
Queen Mary's Hospital for Children,
Carshalton, Surrey

Not gliding but twitching motility of Actinobacillus calcoaceticus

The surface spreading phenomena as well as the movements of individual cells of strain T15 (Actinobacillus calcoaceticus) are due to twitching3 and not to gliding motility as suggested by Mukerji and Ghopale in a recent letter.4 The movements of Actinobacillus reported by Barker and Maxted5 quoted by Mukerji and Ghopale, likewise, in the light of all available experimental evidence were also due to twitching as later admitted by Barker in a letter to me.

References