Letters to the Editor

Detection of bacteraemia by an automated blood culture system

Many clinical microbiology laboratories in the UK and the USA are currently detecting bacteraemia using an automated C\(^4\) labelled substrates blood culture system (Bactec, Johnston Labs, Inc).

Positive blood cultures are initially detected by release of radioactive carbon dioxide into the gaseous space which exceeds a printout value of 40 cpm. We report two cases of endocarditis which, respectively, provide a cautionary reminder and a reassurance to clinicians and microbiologists who rely upon this blood culture system.

CASE 1
A 39-year-old woman was admitted with a ten-day history of malaise, joint pains, sweating attacks and rigors. On examination she was pyrexial and the tip of the spleen was palpable and an Osler's node was seen on a finger. She had a mitral systolic murmur. Six sets of blood cultures were taken over 24 h and antibiotics were commenced using penicillin and gentamicin, a clinical diagnosis of infective endocarditis having been made. The temperature returned to normal within 24 hours of starting treatment and apart from a transient ischaemic attack on day 6 of treatment, no new symptoms or signs were noted.

Blood cultures were examined using the Bactec 460. On the fifth day of incubation one aerobic bottle gave a positive reading. Nothing was seen in the Gram film but *Haemophilus parainfluenzae* was grown in subculture the following day. All other blood culture bottles remained negative. All bottles were then subcultured onto conventional media and *Haemophilus parainfluenzae* was grown from a further two bottles from different sets on day 7. These two bottles persistently gave negative readings up to 21 days of incubation at which time one of the two bottles gave a low positive reading of 59 cpm. *Haemophilus parainfluenzae* was subcultured from both these bottles at 21 days of incubation.

CASE 2
A 28-year-old man with known mixed aortic valve disease presented with a three-month history of a 'flu-like illness with a sore throat, malaise and generalised aches and pains in his limbs. On examination he had a temperature of 37.7°C, hepatosplenomegaly and murmurs of aortic regurgitation and stenosis. A clinical diagnosis of infective endocarditis was made.

Seven sets of blood cultures were taken over three days. These were all processed using the Bactec 460 system: All sets were positive in the aerobic bottles on the first day of reading showing Gram positive cocci on Gram film.

Culture of the blood on standard media failed to produce growth at 24 h but a very scanty growth was visible at 48 h. The organism was alpha-haemolytic on sheep blood agar and optochin resistant and was subsequently identified as *Streptococcus mitior*. The patient was started on ivanogenous benzylpenicillin and netilmicin.

Minimal inhibitory concentration of penicillin was not assayable because the organism would not produce turbid growth in either glucose broth or thioglycollate broth. It was then tested and found to be 0.1% cysyoptin dependent due to a pyridoxine deficiency.

The recent departure from traditional methods to automated technology in medical microbiology has inevitably led to an assumption of the veracity of the machine. This was illustrated by the first case described in which all except one blood culture bottle were believed to be culture negative. This might have led to a failure to appreciate the significance of *H parainfluenzae* in this case but a strong clinical suspicion overcame the inclination to believe the machine printout.

The growing number of reports\(^1\)\(^-\)\(^4\) of nutritionally fastidious organisms causing disease creates concern among microbiologists about the laboratory's capacity to detect routinely their presence in clinical specimens.

The second case provides reassurance about the performance of an automated blood culture system in this context. The inclusion of either pyridoxine or L-cysteine blood culture in the medium supported growth of the organism while traditional sheep blood agar failed to support growth in subculture.

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Quantifying the microbial flora of the cervix

Many studies have been undertaken to define the indigenous bacterial flora of the female genital tract. The majority of these have been qualitative, based on the culture of material obtained from swabs of vagina or cervix. Quantitative studies are few because of difficulties in sampling and also because cervical mucus is both sparse and extremely viscous. It is not, therefore, readily amenable to accurate bacterial enumeration. The few quantitative investigations which have assessed specimens obtained using sterile calibrated loops,\(^1\)\(^-\)\(^4\) pipettes,\(^5\)\(^-\)\(^9\) swabs which are weighed before and after sampling.\(^1\)\(^-\)\(^4\)\(^9\) Difficulties have been encountered with all of these methods. We describe here a further technique for obtaining cervical secretions and quantifying the microbial flora at this site.

Cervical mucus is aspirated using a sterile disposable sampling syringe aspirator (IMV—Aspiagile, Ref UA311), which can be passed easily through the speculum without touching the walls of the vagina. The syringe is calibrated and freshly prepared 10% solution of dithiothreitol (Sputolysin, Calbiochem/Behring Corp, La Jolla, California) is aspirated into it diluting the secretions in a ratio which may vary from 1 to 3:1, depending on the viscosity of the mucus. The syringe is then placed in a sterile test tube and the mixture alternately expelled and aspirated several times until it is liquefied and homogeneous. Six serial tenfold dilutions are made in pre-reduced glucose broth and 10μl samples of the undiluted specimen and each dilution are deposited in the form of drops or plated out, in duplicate, on appropriate media. After incubation colony types are described, counted, and isolated for subsequent identification. From the colony types

References