Letters to the Editor

Counterimmunoelectrophoresis for the detection of bacterial antigens in cerebrospinal fluid

We read with interest Dr DS Tompkins' article comparing Phadebact coagglutination with counterimmunoelectrophoresis (CIE), for the detection of bacterial antigens in cerebrospinal fluid. We would like to draw attention to what may be an important practical point when performing CIE—namely, the dilution of the sample to overcome the potential false negative in the next sample due to the prozone phenomenon. This point is not specifically mentioned in the ACP Broadsheet on the microbiological examination of cerebrospinal fluid, and we wonder if any of your readers have experienced similar cases to the one outlined here.

Case history

A four month old baby girl was admitted with a 24 h history of fever and irritability. No antibiotics had been administered. Investigations included a lumbar puncture, which showed a protein concentration of 1.4 g/l and a cerebrospinal fluid (CSF) glucose concentration that was less than half of a simultaneous blood glucose measurement. The CSF polymorphonuclear leucocyte count was 180 × 10⁶/l, and a number of Gram stains showed a large number of consistently Gram negative cocci bacilli. Some appeared very diplococci in morphology, and despite the Gram reaction we considered the possibility of their being pneumococci, a well described phenomenon in fluid culture. CIE was performed as described in the Broadsheet 108. In view of the large numbers of organisms seen, dilutions of CSF were also tested. All neat specimens were negative when tested against Neisseria meningitidis, Haemophilus influenzae type b, and pneumococcal antisera, but the pneumococcal test proved positive at dilutions greater than 1/16. Culture performed the next day showed a classic Streptococcus pneumoniae, which unfortunately died before it could be serotyped. The patient responded rapidly to parenteral benzyl penicillin, to which the organism was sensitive.

This case indicates how confusing the Gram stain can be. It also highlights the practical importance of the prozone phenomenon. In meningitis treated before lumbar puncture, a large number of bacteria may be lysed and one is unaware of the potential for a prozone. We have so far not assessed the Phadebact coagglutination system, but suspect a prozone may be less of a problem than with CIE.

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References


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Improved blood free selective medium for the isolation of Campylobacter jejuni from faecal specimens

The high incidence of campylobacter enteritis reflects the importance of having available routine isolation facilities and a defined reproducible medium. We have recently described a blood free selective medium—CCD agar—for the isolation of Campylobacter from faeces. Although this medium gave excellent recovery of campylobacters, it was less inhibitory than some other selective agars. The selectivity of the CCD agar was obtained by incorporating cephalin 10 mg/l and sodium deoxycholate 0.1%. In order to improve the selectivity of this medium the cephalo-

lin has been replaced with cefoperazone 32 mg/l, an antimicrobial agent that has proved useful in two other recently described campylobacter isolation media. The modified CCD agar has been compared with the Preston agar for the isolation of C jejuni from faecal specimens. Faecal suspensions prepared in 0-1% peptone water were inoculated on to two plates of each medium using cotton tipped swabs and spread to produce discrete colonies. All plates were incubated microaerobically, one pair of each medium at 42°C and the other at 37°C. Plates were examined after 24 h and 48 h incubation and suspect campylobacter growth was confirmed by the oxidase test and by characteristic cell morphology and motility under dark field microscopy.

Sixty four campylobacter isolations were made from 730 specimens: 62 after incubation at 42°C and 58 after incubation at 37°C (Table). The two specimens which were positive only at 37°C and the six specimens positive only at 42°C produced very scanty growths, indicating that these differences were probably due to sampling. Isolation at both 42°C and 37°C was greater using the modified CCD agar than the Preston agar. Maximum isolation on both media was achieved when plates had been incubated for 48 h.

The modified CCD agar grew contaminants from only 13.4% of specimens at 42°C and from 25% at 37°C compared with 12.3% and 21.8% respectively on the Preston agar. Moreover, of cultures incubated at 42°C the growth of faecal contaminants outside of the primary inoculum area occurred on only 3.4% of modified CCD agar plates and 2% of Preston agar plates. The most frequent contaminants to grow on the modified CCD agar were yeasts, but occasionally streptococci and coliforms were present.

The results with the modified CCD agar used at 37°C are encouraging, and when incubation at 42°C is not available the modified medium should produce acceptable recovery of C jejuni from faecal specimens. We believe that in developing

Growth of campylobacter isolates and contaminants from 730 faecal specimens

<table>
<thead>
<tr>
<th>Incubation (48 h)</th>
<th>Medium</th>
<th>No of campylobacter isolates</th>
<th>Plates growing faecal contaminants (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>42°C (62)*</td>
<td>Modified CCD agar</td>
<td>60</td>
<td>13-4</td>
</tr>
<tr>
<td></td>
<td>Preston agar</td>
<td>59</td>
<td>12-3</td>
</tr>
<tr>
<td>37°C (58)*</td>
<td>Modified CCD agar</td>
<td>56</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Preston agar</td>
<td>52</td>
<td>21-8</td>
</tr>
</tbody>
</table>

*Total number of isolations at stated temperature.
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B Cookson and A Baldwin

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