

# Five cases of *Alcaligenes pseudobacteraemia*

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## SUMMARY

*A cluster of five cases of pseudobacteraemia due to the organism Alcaligenes denitrificans occurred in three hospital medical wards over a four week period. The same organism was isolated from four of twelve commercially prepared bottles for erythrocyte sedimentation tests. The most likely explanation for the outbreak is that the ESR bottles were filled prior to inoculation of blood culture bottles. The outbreak was brought to an end by advising on correct procedure.*

## INTRODUCTION

Pseudobacteraemia means false – positive bacteraemia, and it implies contamination of the blood culture medium, usually from an environmental source. As opportunistic pathogens become more common it is increasingly difficult to distinguish pseudobacteraemia from true pathogens in blood culture. We describe a cluster of five cases of pseudobacteraemia due to the organism *Alcaligenes denitrificans* over a four week period, occurring in three adjacent hospital medical wards. A case cluster can suggest an outbreak of infection, but since this organism is known to exist in the environment an investigation to identify a possible source was carried out.

## METHODS

Inoculated blood culture bottles are incubated at 37°C and sampled for bacterial growth at intervals using a semi-automated blood culture system. If growth is detected, a sample of the blood culture is aseptically withdrawn from the bottle for gram staining and subculture on solid media. Routinely, these plates are also incubated at 37°C, but once an organism suspected of being an *Alcaligenes* species was isolated, culture plates were incubated at 30°C, which improved recovery.

Informal discussions with junior medical staff failed to reveal any breach of blood sampling protocols. In view of previous reports of pseudobacteraemia traced to contamination of the bottles used for full blood picture and erythrocyte sedimentation rate tests, we tested four of each of these bottles from each of the three medical wards involved. The haematology bottles tested are commercially manufactured by Sherwood Medical, UK. They are made of glass and each contains 0.5 ml sodium citrate. They are designed to take 4.5 ml blood and are

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gamma-irradiated at the end of production to ensure sterility. The ESR bottles tested are commercially manufactured by Sterilin, UK. They are plastic and each contains 0.4 ml of sodium citrate. They are designed to take 2 ml blood and are prepared under aseptic conditions although not irradiated. From both the haematology and the ESR bottles, 20 µl of contained sodium citrate was plated on to blood and MacConkey agars. A gentamicin disk was placed on each plate to screen for any gentamicin-resistant, gram-negative organisms, since the *A. denitrificans* strain had consistently exhibited this characteristic.

Antibiotic sensitivity testing was performed using the Stokes method with *Pseudomonas aeruginosa* National Collection of Type Cultures 10662 (Central Public Health Laboratory, Colindale Avenue, London) as a control organism. Isolates were incubated on blood agar at 30°C for 48 hours. Identification was performed using the API 20NE system (BioMerieux, France) for non-enteric gram-negative bacilli. This is a commercially prepared, semi-automated method using a plastic strip containing twenty capules; the capules contain the necessary reagents for twenty different biochemical tests. A suspension of the test organism is inoculated into each of the capules and the results read after forty-eight hours.

## RESULTS

All five strains isolated from blood cultures had the same biochemical profile on testing with the API 20NE system, and were identified as *Alcaligenes denitrificans*. All had a similar antibiotic sensitivity profile, being resistant to gentamicin and amikacin, and sensitive to netilmicin, ciprofloxacin, aziocillin and ceftazidime.

Twelve haematology bottles were tested and no bacteria were isolated. Twelve ESR bottles were tested, four from each of the three wards, and of these four grew a gram-negative, cytochrome oxidase-positive bacillus which was resistant to gentamicin on the primary culture plates. Of the four positive ESR bottles, two came from one ward and one each from the other wards. The full biochemical and antibiotic sensitivity profiles of these isolates were similar to that of the blood culture strains (Table).

TABLE  
*Isolates from ESR bottles*

<i>Alcaligenes denitrificans</i>		
<u>Resistant</u>	<u>Sensitive</u>	<u>Variable</u>
gentamicin	ceftazidime	netilmicin
amikacin	ciprofloxacin	aziocillin

## DISCUSSION

An organism similar to that which caused five episodes of pseudobacteraemia was isolated from the ESR bottles available in each of the three wards. Gram-negative organisms have frequently been isolated from haematology and ESR bottles, and these contaminated bottles have been shown to be the cause of pseudobacteraemia.<sup>1, 2, 3, 4</sup> In these cases, the most likely route of contamination

has been that after drawing blood from the patient the syringe needle was removed and the ESR bottle filled before the blood culture medium was inoculated. Even if a new sterile needle was placed on the syringe before the blood culture bottles were inoculated, the syringe nozzle could already have been contaminated during filling of the ESR bottle. This contamination might occur either by direct contact with the inside of the ESR bottle or by aerosol.

Successful sampling of blood for culture requires a careful technique with attention to detail. The operator's hands should be washed and gloves may be worn. Various recommendations have been made for skin disinfection of the chosen venepuncture site. Gillies<sup>5</sup> advocated a soap and water wash followed by alcohol and povidone iodine, whereas Tyrrel *et al*<sup>6</sup> advise two applications of 70% alcohol or an alcohol-based disinfectant. It is essential that alcohol is allowed to evaporate to dryness and iodine, if used, is allowed two to three minutes to act. After disinfection the operator should not touch the venepuncture site. Following sampling the needle should be changed prior to inoculation of the blood culture bottles. It is helpful if an assistant is available to rip off the protective cap from the blood culture bottles and disinfect the diaphragm with an alcohol wipe. If additional blood samples for other tests are also required, the extra bottles should be filled after blood culture inoculation to avoid possible contaminating organisms from them being carried over to the blood culture bottles. While the doctors who inoculated the blood culture bottles did not feel that contamination of this type had occurred, the outbreak was brought to an end by advising on the correct procedure.

Pseudobacteraemia is an important phenomenon in terms of patient well-being, clinical and laboratory workload, and the cost of unnecessary treatment. Fortunately, none of our cases were treated unnecessarily with antibiotics because monitoring in the bacteriology department raised the suspicion of pseudobacteraemia at an early stage.

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