Letters to the Editor

A tiny colony-forming *Staphylococcus aureus* from a clinical specimen which grew better on MacConkey's medium

An ear swab from a patient with otitis externa was cultured on blood agar and MacConkey's plates. Both were incubated aerobically at 37°C. Another blood agar plate, inoculated with the specimen, was also incubated anaerobically. After overnight incubation there was scanty growth of tiny (0.2-0.3 mm) translucent colonies on both blood agar plates; but the MacConkey's plate produced medium-sized (about 1 mm), opaque, pinkish-white colonies. Gram staining from all three plates showed Gram-positive cocci in clusters. Standard laboratory tests,¹ including coagulase, DNase, and mannitol fermentation, established that the growths from all three plates were *Staphylococcus aureus*. By disc-diffusion test the organism was found to be sensitive to all commonly used antibiotics, including penicillin and gentamicin. On repeated subcultures the organism continued to produce tiny translucent colonies on nutrient agar, blood agar, and chocolate agar plates even after 48 hours' incubation in the presence or absence of carbon dioxide, aerobically or anaerobically. On all occasions MacConkey's medium produced better growth with medium-sized opaque colonies.

To exclude the possibility of the presence of any inhibitory substances in our ordinary media, a control strain of *Staph aureus* (Oxford strain) was cultured in parallel with the test strain on blood agar and MacConkey plates. The control strain produced large (about 3 mm) opaque colonies on the blood agar and medium-sized (about 1 mm) colonies on MacConkey, while the test strain produced two different types of colonies on two plates, as before. This colonial character of the test strain and the fact that it was *Staph aureus* were also confirmed by a neighbouring laboratory. To see whether any of the constituents of MacConkey's medium (not present in blood agar) were responsible for the improved growth of this strain, cultures were made on specially prepared blood agar plates containing any one of neutral red, lactose, or bile salt, in the same concentrations as are used in MacConkey's medium. The plates were examined after incubation at

37°C for 24 hours and 48 hours, and all produced exactly similar tiny colonies; only MacConkey's medium produced better growth. It was thus proved that the MacConkey's medium was better for supporting this particular strain than other commonly used media, and that none of the constituents of MacConkey's medium could improve the growth when used alone in the ordinary media.

Small or 'dwarf' colony-forming *Staph aureus* have been described in the literature.²⁻⁴ They are usually produced under inhibitory conditions and tend to revert to their usual size and growth character when such conditions are removed.⁵ The strains studied by Lacey and Mitchell^{3,4} were highly pigmented ones and resistant to aminoglycosides. The strain described here was different from those of other authors in many ways: (a) it grew better on an inhibitory medium such as MacConkey's than on the usual non-inhibitory media; (b) tiny colonies produced on blood agar were translucent and not pigmented; (c) it was sensitive to antibiotics; (d) repeated subcultures failed to improve the size of the colonies.

The clinical implication of this finding is that such organisms are likely to be missed or ignored if this finding is not borne in mind. Had it not grown better on MacConkey's, the scanty growth of tiny colonies on the blood agar plate from an ear swab might have been ignored in this case.

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References

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- ³ Lacey RW. Dwarf-colony variants of *Staphylococcus aureus* resistant to aminoglycoside antibiotics and to a fatty acid. *J Med Microbiol* 1969;2:187-96.

⁴ Lacey RW, Mitchell AAB. Gentamicin-resistant *Staphylococcus aureus*. *Lancet* 1969;ii:1425-6.

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Blood thixotropy

In the *Journal of Clinical Pathology*, May 1980 (p 418), J Stuart and M Kenny reported that 'blood, like non drip thixotropic paint, shows a fall in viscosity with time during shearing'. This has not been our experience. At a constant shear stress of 1.73 Nm⁻² we observed a variation of shear rates within 1% over a period of 2 hours. The only occasions when a thixotropic-like behaviour has been demonstrated were when blood was allowed to cool before shearing at 37°C, or allowed to stand before shearing. In the first case, the decrease in viscosity can be attributed to a simple temperature effect; in the second, from a breakup of preformed rouleaux. It may be argued that the thixotropic nature of blood is therefore an artefact of insufficient mixing.

If the breakup of a rouleaux formation is to be defined as thixotropy, then, at low shearing, blood also shows rheopecty as rouleaux formation begins. Using a couette system linked to a Deer Rheometer, we observed that at a shear stress of 0.86 Nm⁻² or less, sedimentation occurred, resulting in a steady increase in viscosity over a period of time. If at the end of this period blood was sheared at a higher rate the viscosity returned to its original level. When an insufficiently mixed sample is first sheared at a low stress the resulting viscosity is extraneously high, and, because the degree of separation is virtually impossible to quantify, reproducibility must also be poor.

We believe that it is perfectly acceptable to use one sample for a whole shear range, but, first, shearing must be carried out at a high rate before low shear rate readings are taken, and not the reverse as reported. In practice, we find that after shearing at 1.73 Nm⁻² we can take reproducible readings at progressively lower shear over a time limit of 10 minutes before it is necessary to 'remix' at the higher shear stress.