Matters arising

hominis could be pathogenic. Because of space limitations, this is only implicit in the biotyping scheme presented in Warsaw (Marples 1981) where a taxonomic collection, mostly for non-clinical sources, was studied. The isolates were ornithine decarboxylase positive and two were from the blood. At that time, we had not recognised any anomalies in clumping factor tests, though we have become aware of these in this and other species of coagulase negative staphylococci.

Recognition that tests for clumping may not be specific for *S aureus* when rapid bench kits are used is important, as it may affect treatment and management. In our case, the initial misidentification as an anomalous *S aureus* was not of clinical importance because appropriate treatment was given. There continues to be a need to identify organisms causing endocarditis.

Comparison of latex and haemolysin tests for determination of anti-streptolysin (ASO) antibodies

Curtis *et al* compared latex and standard haemolysis inhibition tests for determining ASO antibodies.¹ May we offer one obvious explanation for the discrepancies in results referred to in their table 3? They make no distinction between antistreptolysin antibody (ASO) and the entity that we have designated as antistreptolysin factor (ASF).² The latter is produced by the activity of cholesterol esterase on low density lipoproteins and is contained in peptide fragments where cholesterol is spatially orientated in such a manner that it is capable of binding to streptolysin O.³ For years we have routinely distinguished between ASO and ASF by measuring activity before and after precipitation of lipoproteins with dextran sulphate. Only the latter fraction contains true antibody activity and titres are almost always lower than they are before precipitation. Raised ASF titres may be found in some patients with chronic staphylococcal infection, in rheumatoid arthritis, and in glomerulonephritis. Even in patients with Lancefield group A infections, the titre measured by haemolysis inhibition is usually due to ASF with true ASO only about two thirds that of ASF. Occasional contaminated sera give high ASF concentrations due to production of cholesterol esterase by *Staphylococcus aureus* or *Pseudomonas aeruginosa*. Both produce high ASF concentrations when grown in serum broth mixtures.

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Marking planes of surgical excision on specimens with mixture of India ink and acetone

In a recent paper by Paterson and Davies,¹ the use of artists' pigments suspended in acetone to mark planes of surgical excision on breast specimens was advocated. The pigment preparations were reported to be better than India ink in that spreading did not occur and drying was much quicker. These authors did not mention the use of India ink in acetone or other volatile liquids in their methods nor in their discussions. In our laboratory the pathologists routinely use India ink (Pelikan AG, West Germany) to mark planes of surgical excision. The possibility of improving the performance of India ink by mixing it with a suitable volatile reagent immediately came across our minds. We were unaware of anyone who had explored this possibility.

We mixed equal parts of acetone with India ink (Pelikan AG, West Germany) and compared the application of this mixture with undiluted India ink on two breast specimens. When the India ink and acetone mixture was applied with a brush, no spreading of the marker beyond the area painted was noted. Drying was quick and completed in less than five minutes. The mixture was easy to prepare and was free from potential health hazards associated with the artists' pigments.¹ If the mixture was allowed to dry, however, a hardened gel would result that could be very difficult to break up. The undiluted India ink was slow to dry and tended to spread beyond the area painted. The intensity of labelling using the two preparations was indistinguishable when viewed under the microscope.

We tried to use absolute ethanol to replace acetone in the mixture. Spreading of the marker was noted, and we concluded that ethanol was not a good substitute for acetone for this purpose.

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Reference

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Dr Davies *et al* comment:

We were intrigued to see the above letter from Drs Chan *et al* about the use of India ink mixed with acetone. A brief trial in this laboratory confirms the apparent benefit of their novel method, although the very fluidity of the mixture—admittedly an asset in rapidly coating a biopsy specimen—would make differential marking difficult. Indeed, it must be pointed out that by itself India ink, even admixed with acetone, precludes microscopic identification of more than one plane of resection. Coloured pigments are necessary for this purpose. Use of the many older, if more muted artists' pigments¹ would avoid the potential toxic hazards of cadmium, cobalt, and other bright modern colours. Furthermore, the use of India ink diluted in acetone does not prevent the unfortunate tendency for the marker to penetrate between lobules of fat. The main advantage of acetone as an ancillary mountant for India ink is that it shortens the drying time. Like Chan *et al*, we, too, found in pilot studies that ethanol was a poor substitute for acetone.

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