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BRIEF COMMUNICATION

A Modified PAS Staining Technique for Polysaccharides in Gels*

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Standard procedures of the periodic acid–Schiff reaction when used to stain polysaccharide components in immuno-electrophoretic analyses in agar gels or in electrophoretic analyses in polyacrylamide gels have some disadvantages. Firstly with agar gels there is a strong tendency for the gels to be stained with increasing intensity on exposure to the atmosphere, thus reducing the definition of the specific reaction. Secondly the use of a reducing rinse after staining to counteract this effect removes stain from the specific reaction at the same time.

In the attempts to stain the polysaccharide precipitin lines in agar gels the disadvantages mentioned could not be eliminated with the usual PAS techniques. Consequently, the PAS staining method, described previously (Buchanan, Dekker and Long, 1950; Baddiley, Buchanan, Handschumacher and Prescott, 1956) for the detection of α-glycols in paper chromatograms, was modified. The modified technique was found to be suitable for detecting polysaccharide precipitin lines in agar gels or polysaccharide bands in polyacrylamide gels.

AGAR GELS

The gel was pre-dried on the glass plate, covered with a 1 per cent solution of sodium periodate (British Drug Houses) and left for 15 minutes. The periodate was replaced with distilled water, and sulphur dioxide gas (SO_2 siphon—British Drug Houses) was passed into the water. The gel was left in the SO_2 water until the brown colour, which appeared initially, had disappeared when the gel was washed briefly in distilled water. At this stage the gel was stained with a 1 per cent solution of pararosaniline hydrochloride (Hopkin and Williams Ltd.) which had been decolorized by passing SO_2 gas through the solution. The gel was left in the stain for 20–30 minutes, after which it was washed in SO_2 water and re-dried quickly by placing in a film drying-cabinet. When dry, the gel was evenly sprayed with 'Quelspray' (clear, No. 418 A) a vinyl protective coating supplied by Fisons Scientific Apparatus Ltd., Loughborough.

In the stained, permanent preparation the gel was a light bluish-purple and the specific polysaccharide reaction was dark pink. There was no indication of the colour increasing in intensity after spraying with the protective coating.

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POLYACRYLAMIDE GELS

The technique may be used to stain polysaccharide bands separated electrophoretically in polyacrylamide gels but the times of exposure to the two reagents must be increased. There are several factors which may affect the times of exposure. Pore size of the polyacrylamide gels (7 per cent w./v. monomer) is much smaller than that of agar gels. Also polyacrylamide gel slices, which are stained in the wet state, are substantially thicker than dried agar gels. Hence penetration of the reagents is a slow process.

In this laboratory when staining gels 1·5 mm. in thickness the times of exposure were 45–60 minutes in 1 per cent sodium periodate and *at least* 24 hours in 1 per cent pararosaniline hydrochloride. After staining the gel was kept in a weak solution of SO_2 water.

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