REPLY: THE USE OF XYLENE (XYLOL) IN MEDICAL LABORATORIES

Sir,

I was interested in Dr A C McDougall's comments and questions concerning the deterioration of acid-fast colouration of mycobacteria during the examination of smears (*Lepr Rev*, 1989, **60:** 67).

In my experience *Mycobacterium leprae* in smears is irregular in its retention of acid-fast stain. On occasion the colouration may withstand immersion oil, xylene or mountant, but at other times it unaccountably fades. This unpredictability applies to all forms of bacilli, solid, fragmented and granular, and to bacilli from different sites in the same patient. For this reason I would recommend that a leprosy smear be examined as soon as it is stained; it can be left fixed and unstained until a suitable time.

Once the colouration is lost the only sure way to restore it is to restain the smear as for a tissue section, but using a slightly modified technique as follows. Immerse the smear in xylene for 15 min, rehydrate, stain in cold carbol fuchsin for 15 min, dry in air, apply a drop of turpentine (pinene) to one side of the smear and tilt the slide so that the turpentine flows over the smear, differentiate immediately without drying in 25% acetic acid, wash in water, dry, counterstain and examine. The staining and bacterial morphology are restored.

The foregoing remarks apply to smears from patients. In tissue homogenates, and in smears from some animals, *M. leprae* for some reason holds the acid-fast dye much more strongly. Similarly smears of *M. tuberculosis* and BCG hold acid-fast dye tenaciously, which is not due only to the higher temperatures at which these organisms are usually stained since *M. leprae* stained in the same way may not retain its colouration so well. The explanation presumably lies in the active principle of the component involved in the staining reaction, which is not yet clearly identified. The fatty products surrounding the bacilli in a smear may also play a role.

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