Technical methods

A technique for bacteriological sampling of hair

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Bacteriological sampling of hair has been previously carried out by applying culture plates directly to the hair but the resulting heavy growth of mixed organisms could make rapid identification of individual colonies difficult and the use of selective media with this technique prevents a comparison of relative numbers of different organisms. The method described here, based on a technique used by MacKenzie (1963) to isolate ringworm fungi from schoolchildren, was compared with the direct plating method (Summers, Lynch, and Black, 1965) and a sampling method employing swabs.

Material and Methods

The brushes used were circular massage brushes of a synthetic material (see fig).¹ They were 3 in. in diameter and the teeth, arranged in concentric circles, were buffed down to the same level to enable even inoculation onto solid culture media. Since the outer four rings of 'teeth' had a total of 100 points it was possible, in samples where growth of discrete colonies was found, to calculate the relative percentages of different organisms present in the sample. Where confluent growth was obtained, as in figure, it was still possible to compare the proportions of organisms present in small numbers only by replicate inoculation onto selective media.

The brushes proved unsuitable for autoclaving and immersion in a 1 in 20 dilution of a solution of sodium hypochlorite containing 10-14% w/v available chlorine for approximately 18 hours was found to be the most satisfactory method of sterilization. This was followed by boiling in water containing 1% sodium carbonate for 10 minutes and rinsing in methylated spirit. The brushes were then placed in sterile polythene bags and allowed to dry at 45°C and the bags sealed with wire.

¹D.6 massage brushes obtained from Osborne, Garrett, Nagele Limited, 51-55 Frith Street, London W1A 2JN.

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Fig Brush used in the hair sampling technique with an inoculated blood agar plate after incubation.

Solid media were employed in disposable Petri dishes $3\frac{1}{2}$ in. in diameter. For the 'brush' technique the following media were used:

- 1 Ten per cent horse blood in Columbia agar base (Oxoid CM 331): this was also used as the only medium for the other two techniques
- 2 MacConkey agar no. 2 (Oxoid CM 109)
- 3 Mannitol salt agar (Oxoid CM 85)
- 4 1:500,000 dilution of Crystal violet in 10% horse blood agar

Method

A 'BRUSH' TECHNIQUE

The brush was pressed onto the scalp and drawn through the hair three times. Directly after sampling

Technical methods

the 'teeth' were pressed gently onto the surface of each of the media described, in turn, and finally onto a second blood agar plate to ensure that there was carry-over of the organisms (this plate was later dispensed with when it was found that there was satisfactory carry-over). The plates were incubated at 37°C for 36 to 48 hours and then examined. The organisms cultured were then identified by routine bacteriological methods.

With each batch of sterilized brushes a random sample was inoculated onto blood agar plates and incubated at 37°C for 48 hours to ensure that sterilization was effective.

B DIRECT PLATING TECHNIQUE

One blood agar plate was pressed onto the surface of the hair and the plate incubated as in A.

C SWABBING TECHNIQUE

A sterile cotton wool swab was rubbed against the scalp and through the hair and inoculated immediately onto a blood agar plate.

Results

A total of 141 persons (hospital personnel, inpatients, and outpatients) were examined by the 'brush' technique; of these 64 were also examined by the 'direct plating' method and the remaining 77 by the swab method. Since the percentage of Gram-negative bacilli was very low compared to the numbers of staphylococci in all cases and since there were no samples in which staphylococci failed to be isolated by all methods the isolation of Gram-negative bacilli from the blood agar plates only was taken as the criterion for the relative efficiency of the different methods.

The results obtained are shown in the table. These results show that the 'brush' technique and the direct plating method are equally effective in the isolation of Gram-negative bacilli. Each technique, however, 'missed' organisms which were isolated by the other technique. In addition Gram-negative bacilli were

Number of Samples from which Gram-negative Bacilli Isolated on Blood Agar Plates by:			
	Both Techniques used	'Brush' Technique Only	Other Method Only
Comparison of brush technique and direct plating method (64 samples)	7	4	5
Comparison of brush technique and swabbing method (77 samples)	5	13	2

Table Comparison of brush technique and direct plating and swabbing methods

isolated on selective media only, by the brush technique, in 10 cases.

The brush and plating techniques are, however, superior to the simple swabbing method for the isolation of Gram-negative bacilli and this method could not be recommended for routine use.

The method to be employed in bacteriological sampling of the hair would depend on the purpose of the examination. Selective media for the purpose of isolation of particular organisms could be employed with the direct plating method but comparison of the relative numbers of different organisms would not be possible.

The brush technique has been found to produce results comparable with the direct plating method in the isolation of organisms using a single culture medium. In addition, however, it was possible to facilitate the isolation and identification of Gram-negative bacilli, especially when present with a heavy growth of staphylococci, by replicate plating onto selective media.

References

- MacKenzie, D. W. R. (1963). 'Hairbrush diagnosis' in detection and eradication of non-fluorescent scalp ringworm. *Brit. med. J.*, 2, 363-365.
- Summers, M. M., Lynch, P. F., and Black, T. (1965). Hair as a reservoir of staphylococci. *J. clin. Pathol.*, 18, 13-15.