Early Detection of Streptococci in Swabs by Latex Agglutination
Before Culture

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A commercial streptococcal grouping system was used to demonstrate streptococcal antigen in swabs before culture. The method detected 81% of the streptococci of groups A, B, C, and G subsequently isolated in culture. The method offers a sensitive and specific method for the early detection of beta-hemolytic streptococci.

When traditional techniques for the detection of streptococci are used, at least 18 h elapse between the receipt of a specimen and the issuance of a culture report; however, there are some circumstances such as major burns for which the early detection of Streptococcus pyogenes may be important. Various methods for the earlier detection of S. pyogenes and other streptococci have been described, including short incubation of swabs in selective enrichment broths followed by fluorescent-antibody staining (1, 9), staphylococcal coagglutination (10, 11), latex agglutination (7, 9), or counterimmunoelectrophoresis (4) to detect streptococcal antigens in the broth. Methods involving the direct examination of the specimen include the use of fluorescent-antibody staining smears (6, 8), the extraction of throat scrapings with nitric acid and precipitation detection of antigen (5), and the detection of streptococcal antigen in throat gaggles with latex agglutination (3).

Streptex (Wellcome Diagnostics, Temple Hill, Dartford, England) is a sensitive and reliable method for grouping streptococci of groups A, B, C, and G (2). This note reports the use of Streptex in a new method for the detection of streptococcal antigen in original swab specimens. In addition to the detection of S. pyogenes, streptococci of groups B, C, and G were also sought.

Altogether 153 swabs were investigated. These were received in Cary-Blair transport medium (CM519; Oxoid Ltd., Basingstoke, Hampshire, England) and were from patients with skin lesions, including burns, decubitis ulcers, skin grafts, and wounds. The investigation was carried out in three stages. Extracts from 52 swabs from which beta-hemolytic streptococci of groups A, B, C, or G had previously been isolated were examined to verify that the method was capable of detecting these streptococci. An additional 101 swabs were examined upon receipt and before the culture results were known. The sensitivity of the method was determined by diluting overnight broth cultures of one wild strain each of streptococcal groups A, B, C, and G. Logarithmic dilutions of these broths were prepared in sterile distilled water to give dilutions of 10^{-1} to 10^{-9}. Viable counts were performed on these dilutions. Sterile swabs were also placed into each dilution, allowed to absorb fluid, removed, drained, and then treated as the clinical swabs. The average volume of fluid absorbed by this batch of swabs was determined by dipping 10 preweighed swabs into distilled water. These were removed, drained, and reweighed, and the volume of water adsorbed was calculated.

Streptex reagents were used for the extraction and detection of the streptococcal antigens. The extraction enzyme and the latex suspensions were prepared and stored according to the instructions of the manufacturer. The enzyme was dispensed in 0.4-cm³ volumes in polystyrene tubes (75 by 10 mm). Swabs were placed in the enzyme, and the tube was vortexed to ensure impregnation of the swabs. The tubes were then placed at 37°C for 1 h. After extraction the swabs were removed from the tubes, and the absorbed fluid was expressed from the swabs by squeezing the swabs against the inside of the tube. The tubes were then centrifuged to deposit any precipitate present. One drop (25 µl) of the supernatant was added to each of the four wells on the glass tile provided with the test kit. Latex suspensions for streptococci of groups A, B, C, and G were added to the appropriate wells. The contents of the wells were mixed, and the tile was gently rocked. Strong agglutination of the latex was recorded as a positive reaction.

The initial study of 52 swabs indicated that the method was capable of detecting streptococci of groups A, B, C, and G. The results obtained with the total 153 swabs examined are shown in Table 1.

The broth cultures used for the quantitative studies contained ca. (per cm³): 20 × 10^6 CFU of group A streptococci, 90 × 10^5 CFU of group B streptococci, 30 × 10^5 CFU of group C streptococci, and 50 × 10^5 CFU of group G streptococci. These all gave positive reactions at dilutions of 10^{-2}. The swabs were found to absorb ca. 40 µl of distilled water. Assuming that the rate of absorption and retention of streptococcal cells by the swabs was the same as that measured for distilled water, the smallest number of streptococci detectable by this method is 8 × 10^3 CFU of group A, 3.6 × 10^3 CFU of group B, 1.2 × 10^4 CFU of group C, and 2 × 10^5 CFU of group G.

Extracts from six swabs gave a positive latex reaction when streptococci of the corresponding groups were not isolated from the swab. All of these false-positive reactions were with the group C latex and were associated with the isolations of a Klebsiella pneumoniae strain in large numbers from the specimen. No other false-positive latex results were obtained. In addition to beta-hemolytic streptococci of groups A, B, C, and G other organisms isolated from the specimens included Staphylococcus aureus, coagulase-negative staphylococci, Corynebacterium sp., Escherichia coli, Klebsiella spp., Enterobacter spp., Proteus spp., Pseudomonas aeruginosa, and anaerobic gram-positive cocci. With the exception of the K. pneumoniae strain already noted there were no cross-reactions attributable to these organisms. With the exception noted above, the positive latex reactions were specific for the group of streptococci isolated.

The Streptex method has been shown to be reliable for
TABLE 1. Swabs in which streptococci were detected by isolation and latex agglutination, isolation, or latex agglutination

<table>
<thead>
<tr>
<th>Streptococcal group</th>
<th>No. of swabs in which streptococci were detected by*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Isolation and latex agglutination</td>
</tr>
<tr>
<td>A</td>
<td>29 (87.9)</td>
</tr>
<tr>
<td>B</td>
<td>19 (76)</td>
</tr>
<tr>
<td>C</td>
<td>7 (87.5)</td>
</tr>
<tr>
<td>G</td>
<td>26 (76.5)</td>
</tr>
</tbody>
</table>

* Values in parentheses are the percentages of total positive isolations. The average percentages were: 81% (isolation and latex agglutination), 19% (isolation), and 6% (latex agglutination).

grouping streptococci of groups A, B, C, and G (2) and for the detection of these streptococci in enrichment broth cultures (D. N. Petts, M.S. thesis, University of Surrey, Guildford, England, 1981). It has also been shown to be more sensitive than precipitation or coagglutination for the detection of streptococcal group A antigen in throat gargles (3). In this investigation, 81% of the swabs from which streptococci of groups A, B, C, or G were isolated gave a positive agglutination with the appropriate group latex after enzyme extraction. The sensitivity study suggests that only $10^{6}$ CFU of the streptococci sought need to be present in the swab before a positive reaction can be obtained. Similar results have been reported in a study on broth containing swabs (Petts, M.S. thesis). However, colony counts may not be a valid method of assessing the sensitivity of a streptococcal antigen detection method. With streptococci a CFU may be a single cell or a long chain of cells and the length of the chains will vary considerably within and between species. The sensitivity of a streptococcal antigen detection method would therefore be best related to the number of cells rather than to the number of CFUs.

Swabs which gave a positive reaction with group C latex but from which a streptococcus of this group was not grown all grew K. pneumoniae type 54. The instructional literature for Streptex states that occasional false-positive streptococcal grouping results may occur with organisms from unrelated genera, including Klebsiella. It is suggested that this is due to the nonspecific stickiness often found with Klebsiella capsules. As this false-positive reaction occurred only with swabs containing this particular strain of K. pneumoniae and a Streptex grouping on a broth culture of this organism gave a positive result with group C latex, it is likely that this is a cross-reaction with Klebsiella antigen. It would appear from the results obtained in this investigation that enzyme extraction of swabs and latex agglutination detection of streptococcal antigen is a sensitive method for the detection of streptococci of groups A, B, C, and G. Results can be obtained within 90 min of receiving the specimen. The results obtained show a high degree of specificity, but it must be remembered that as in all immunological tests cross-reactions with other organisms may occur. Also, because a negative result does not exclude the presence of the streptococci sought, this method should only be viewed as a preliminary investigation which needs confirmation by culture. Nevertheless, a positive result has a good predictive value.

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LITERATURE CITED