Streptococcal pharyngitis—rapid diagnosis by Gram stain

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Summary
Group A β-haemolytic streptococci were isolated from 51 (9.8%) of 519 patients with pharyngitis. Throat culture results showed the mean sensitivity, specificity and predictive value of a positive Gram-stained smear of pharyngeal secretions as 70%, 89% and 69%. The data suggest that Gram staining of a smear directly from the swab is as accurate as and more speedy than a culture from the pharyngeal secretion for diagnosis of streptococcal pharyngitis.

Introduction
Upper respiratory infections are the most common cause of acute, short-term illness. The largest treatable subset of these infections are those attributable to Group A β-haemolytic streptococci (Streptococcus pyogenes) (Monto and Ullman, 1974). It is important to distinguish treatable streptococcal infections, which have serious sequelae, from other infections for which no specific therapy is available. Despite reports that the microbial aetiology of a sore throat can be ascertained only by laboratory diagnosis, many doctors in hospitals and general practice still do not heed this advice. There is evidence that the majority of patients presenting with upper respiratory tract infections do not have their throats swabbed, and some doctors still deny any real place for throat swabs in everyday medical practice, insisting that an experienced doctor can detect a streptococcal sore throat ‘immediately’. Clearly, in these circumstances, many patients will receive inappropriate antibiotic therapy—those with a sore throat of viral aetiology receiving antibiotics unnecessarily, and those with a streptococcal throat perhaps not having any specific therapy.

Despite difficulties in distinguishing streptococcal carriage from infection, the throat swab culture remains the recommended method of diagnosis (Kaplan et al., 1977). Because appropriate therapy based on the result of a culture is necessarily delayed, and return visits for treatment entail additional time and cost, many attempts have been made to develop criteria for the diagnosis of streptococcal sore throat on the initial presentation. Clinical criteria have been shown to be of no value in some reports, but other investigators have been able to assign patients into groups at significantly differing risks for streptococcal pharyngitis on the basis of clinical evaluation (Mernstein and Rogers, 1974; Walsh et al., 1975). Diagnosis by clinical criteria has suffered, however, from lack of sensitivity. Pharyngeal secretions can be evaluated by Gram stain for both cellular content and microbial flora. This technique has been supported (Brancato and Parker, 1966) and condemned (Keitel, 1965) as an aid to the diagnosis of streptococcal pharyngitis, further evidence to support the need for laboratory diagnosis of sore throat has been sought, and the authors now report a study of Gram-stained smears of pharyngitis.

Materials and methods
The study was conducted among out- and in-patients with pharyngitis at the Military Hospital, Jabalpur.

Requests for throat culture had to be accompanied by a smear of pharyngeal secretion from each patient, and a brief clinical summary in each case. There were 519 submissions for culture.

Obtaining specimens for culture followed a standard technique: a cotton swab rubbed on the area of maximal exudate (when present); or on the tonsillar pillars and posterior pharynx. The material was smeared on a clean glass slide which, on being received in the laboratory, was inoculated on to blood medium containing 5% human blood. The number of β-haemolytic colonies was recorded, as were details of observations made after 24 hr of incubation. No patient was excluded because of a technically inadequate smear or culture.

The air-dried smears were successively flooded with crystal violet, Gram's iodine, acetone-alcohol, and 0.1% basic fuchsin. The basic fuchsin counterstain was found to provide better definition of cellular detail for fusospirochaetal flora than did safranine or other counterstains. The slides were read by experienced observers. The Gram-stained smear was...
scanned at a magnification of $\times 100$ for the presence of leucocytes. If none was found, it was deemed negative. If leucocytes were present, the areas with the greatest proportion of polymorphonuclear cells or showing 'disruption' (loss of cytoplasmic integrity and cellular outlines) were located. The microbial flora associated with the polymorphonuclear cells in these areas were examined at a magnification of $\times 1000$. Spherical Gram-positive cocci occurring singly and in pairs (structure typical of S. pyogenes) were differentiated from other Gram-positive cocci, which may be more elongated or encapsulated (pneumococci), from chains or clumps. If Gram-positive cocci of typical structure were found associated with the polymorphonuclear cells, the slide was deemed positive.

The clinical summary for each patient included the presence or absence of cervical adenopathy, sore throat, pain on swallowing, tonsillar exudate, nasal discharge, pyrexia of 38.5°C+, vomiting, or a history of recent exposure to streptococcal pharyngitis. Examining physicians were also encouraged to give their clinical assessment and to indicate the probable cause, e.g. whether streptococcal or viral.

Results

β-haemolytic streptococci were isolated from 51 of 519 patients; of these, 12 showed 10 or fewer colonies on primary isolation, 39 showed 11 or more colonies.

Patients’ ages ranged from 1 to 75 years (mean 15-2 years); 54% were aged 6–15 years. The highest proportion of positive cultures (28%) was in the 6- to 11-year-old group (78 patients). The lowest proportion of positive cultures (9%) was in the group aged 30 years or more (73 patients). These figures accord with those of other workers (Glezen et al., 1967).

The sensitivity of the technique (positive culture and positive Gram stain), the specificity (negative culture and negative Gram stain), and predictive value (positive Gram stain with positive cultures) was determined. The mean sensitivity, specificity and predictive value were 70%, 89% and 69% respectively. The agreement of the Gram stain interpretation and the culture results were highly significant ($P < 0.001$). The sensitivity of the Gram stain was 76% for patients with 11 or more colonies on primary isolation but only 38% for those having 11 ($P < 0.001$).

Table 1 contrasts the sensitivity, specificity and predictive values of the Gram stain with those of the individual criteria. S. pyogenes was isolated significantly more often from patients with a history of exposure, tonsillar exudate, pain on swallowing, fever of 38.5°C or greater, or cervical adenopathy and less often from patients with only cough. The sensitivity, specificity and predictive values of positive Gram stain were higher than those of any individual criteria. The percentage of patients with signs and symptoms in the streptococcal-positive group, non-streptococcal, and 'no-isolation' group are given in Table 2; and the analysis of accuracy of clinical diagnosis in Table 3.

Discussion

Expertise in the interpretation of the Gram stain requires experience. The results show that Gram staining of throat swabs may be a reliable method for the detection of streptococcal infections (Table 1). There were appreciable differences in the signs and symptoms of the streptococcal, non-streptococcal and no-isolation groups (Table 2). Many of the cases in the no-isolation group could have been viral in origin in that there was a low incidence of tonsillar exudate and a high incidence of adenopathy. There was a low incidence of pyrexia (58%) in the cases of streptococcal infection. The presence or absence of the tonsils did not alter the clinical picture appreciably in any group. On clinical examination alone, a 47.3% accuracy in diagnosing streptococcal pharyngitis and 60.6% accuracy in diagnosing non-streptococcal throat were recorded (Table 3). The data emphasize the importance of laboratory study in all cases. It is suggested that signs and symptoms of streptococcal, non-streptococcal and no-isolation groups are so similar that an accurate diagnosis of the aetiology cannot be made on clinical grounds alone, and that Gram staining is a reliable alternative for arriving at a correct diagnosis and for instituting appropriate therapy.

Although delay in therapy until culture results

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<th>Table 1. Sensitivity, specificity and predictive value of various criteria from the Gram-stain examination relative to throat culture results</th>
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<tr>
<td><strong>Fever ≥ 38.5°C</strong></td>
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become available does not compromise the prevention of rheumatic fever, there are several potential benefits of treatment on initial presentation of the patient. Some studies have shown a small decrease in the morbidity of streptococcal pharyngitis if treatment is given early (Brink et al., 1951). Treatment of the patient within the first 24–48 hr of illness may minimize the chance of spread of streptococcal infection to other members of a family. The necessity of a return visit for treatment adds cost and inconvenience for the patient and, in some populations, follow-up may not be assured. In such groups with uncertain follow-up, a reliable method of early diagnosis of streptococcal pharyngitis should increase the treatment rate. The findings suggest that the Gram-stain study of the throat swab itself may have a role as an alternative to throat swab culture.

References
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