

Diagnosis of Streptococcal Pharyngitis by Detection of *Streptococcus pyogenes* in Posterior Pharyngeal versus Oral Cavity Specimens

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Carbohydrate antigen detection, nucleic acid probe detection, and bacterial culture are commonly used to confirm group A streptococcus (GAS) pharyngitis. Compared to standard throat swab specimens, the sensitivities of these tests with mouth specimens are poor. When testing for GAS pharyngitis, the throat remains the optimum site for sampling.

Streptococcus pyogenes (group A streptococcus [GAS]) causes approximately 15 to 30% of pediatric sore throats (1). Accurate diagnosis permits appropriate administration of antimicrobial therapy to prevent complications, hasten symptom resolution, and reduce the transmission of GAS in the community (7, 12, 13). Unfortunately, clinical evaluation and scoring systems cannot reliably identify patients with streptococcal pharyngitis, necessitating the utilization of laboratory tests (3, 6). While culture of GAS on sheep blood agar medium remains the gold standard for detection of GAS in throat swab specimens (2), newer and more rapid diagnostic tests, including direct carbohydrate antigen and nucleic acid probe detection, are now available.

Current methods of GAS pharyngitis testing rely on swab specimens obtained from the posterior pharynx and tonsils. This convention is based on recommendations published by the Infectious Diseases Society of America (IDSA), which defines specimens obtained from these sites to be the only adequate samples to test for the presence of *Streptococcus pyogenes* (2). It should be noted, however, that these recommendations are based on two small studies performed more than 20 years ago that evaluated only 32 patients (4, 9). In this study, we investigate the use of newer techniques of rapid direct carbohydrate antigen and nucleic acid probe detection to identify GAS in samples obtained purposively from the oral cavities and posterior pharynxes of pediatric patients with sore throats. This comparison is of practical significance because it is often difficult to obtain a well-collected posterior pharyngeal swab specimen from young children.

This study was approved by the Institutional Review Board of Columbus Children's Hospital and conducted with informed consent between September 2004 and February 2005. Fifty-three patients presenting with a chief complaint of sore throat to the Children's Hospital emergency department were enrolled as study subjects. Subjects ranged from 3 to 18 years of age; the only exclusion criterion was prior tonsillectomy.

Each subject underwent two swab collections with a double-

swab collection/transport system (COPAN Venturi Transystem; COPAN Diagnostics Inc., Corona, CA). The standard posterior pharyngeal (throat swab) collection technique involved vigorous swab sampling of the posterior pharynx and bilateral tonsillar tissue while avoiding contact with the tongue, buccal surfaces, and lips. The oral cavity (mouth swab) collection technique involved blind sampling of the subject's mouth, targeting the tongue and buccal mucosa. Each subject was asked to rate his/her pain with a validated age-appropriate pain scale after each swab collection (11).

Immediately after sample collection, one swab from each site was used for direct antigen detection with an Abbott Signify Rapid Strep A test (Abbott Laboratories, Abbott Park, IL). Within 24 h, a nucleic acid probe test (Gen-Probe Direct Strep test; Gen-Probe Inc., San Diego, CA) was performed on the remaining throat and mouth swabs according to the manufacturer's instructions, and a broth-enhanced culture for GAS was performed on the pledget from each collection/transport tube system. Briefly, the pledget was placed into LIM broth (Becton Dickinson, Sparks, MD) and incubated overnight at 35°C. A subculture of the broth was performed on SXT blood agar (Becton Dickinson), and the plate was incubated in 5% CO₂ at 35°C for 48 h. Beta-hemolytic colonies were identified as GAS by a latex agglutination test (Streptex GAS test; Remel Inc., Lexena, KS). The gold standard for diagnosing GAS pharyngitis was a positive culture or nucleic acid probe of the pharyngeal/tonsillar specimens.

It was determined that 58% (31/53) of subjects had GAS pharyngitis. Only one patient had discordant throat probe and culture results. For testing of throat swab specimens, the sensitivity and specificity were 80.6% and 100%, respectively, for the direct antigen test and 93.3% and 95.7%, respectively, for the nucleic acid probe test. For testing of mouth swab specimens, the sensitivity and specificity were 19.4% and 100%, respectively, for the direct antigen test and 41.9% and 100%, respectively, for the probe test. The sensitivity of enhanced culture performed on mouth swab specimens was 80.6% (Table 1). By pairwise comparison, the sensitivity of tests performed on mouth swabs was lower than the sensitivity of the same tests performed on throat swabs ($P < 0.001$, analysis of variance for proportions). The level of subject discomfort during collection of mouth swabs was lower than that during collection of throat

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TABLE 1. Performance characteristics of culture, direct carbohydrate antigen test, and DNA probe test^a

Test ^b	Site	% Sensitivity	% Specificity	% PPV	% NPV
Rapid Strep*	Throat	80.6 (62.5–92.5)	100 (85.1–100)	100 (86.3–100)	78.6 (59.0–91.7)
DNA probe†	Throat	93.3 (77.9–99.2)	95.7 (78.1–99.9)	96.6 (82.2–99.9)	91.7 (73.0–98.9)
Rapid Strep*	Mouth	19.4 (7.5–37.5)	100 (84.6–100)	100 (54.1–100)	46.8 (32.1–61.9)
DNA probe*	Mouth	41.9 (23.9–60.9)	100 (84.6–100)	100 (75.3–100)	55.0 (38.5–70.7)
Culture*	Mouth	80.6 (62.5–92.5)	100 (84.6–100)	100 (86.3–100)	78.6 (59.0–91.7)

^a Specimens were obtained from the posterior pharynxes and mouths of 53 study subjects. Values shown are means (95% confidence intervals). PPV, positive predictive value; NPV, negative predictive value. A *P* value of <0.001 was determined for all pairwise comparisons of sensitivities of the different streptococcal tests performed on specimens obtained from the mouth versus the posterior pharynx/throat.

^b *, gold standard reference is positive culture or DNA probe of posterior pharynx/tonsils. †, gold standard reference is positive culture of the posterior pharynx/tonsils.

swabs (data not shown; *P* < 0.0001, Wilcoxon signed rank sum test).

Throat swab sampling demonstrated superior sensitivity for all GAS detection techniques investigated in this study, substantiating the importance of a well-collected sample from the posterior pharynx when attempting to diagnose GAS pharyngitis. The performances of the direct antigen and nucleic acid probe tests on throat swabs in this study were similar to previously reported data (5, 8, 10). However, the sensitivity of each test (direct antigen, probe, and culture) on mouth swabs was unsatisfactory compared to the recommended throat swab. Notably, the prevalence of GAS pharyngitis in this study population (58%) was higher than that from previously published reports (1). While the positive and negative predictive values of tests may be affected, prevalence does not affect a test's sensitivity and specificity. Although throat sampling is less tolerable to patients, medical personnel should make every effort to obtain samples from the posterior pharynxes and tonsils of children with sore throats.

Despite this recommendation, our data suggest that there may be some utility in special circumstances to performing direct antigen tests on swab specimens believed to arise from nonpharyngeal/nontonsillar sites. One such circumstance involves the uncooperative child for whom a "clean" throat swab without oral secretion contamination is impossible to obtain. With excellent specificity, positive direct antigen tests of such samples—collected inadvertently/purposely from the oral cavity—would be sufficient to confirm GAS pharyngitis. A negative test in this situation, however, would not exclude this infection and should prompt additional attempts to sample the posterior pharynx for further testing, empirical treatment with antibiotics, or performance of an enhanced culture on the remaining mouth swab.

In conclusion, our study validates recommendations by the IDSA to obtain a sample from the posterior pharynxes and tonsils of patients suspected of having GAS pharyngitis. Traditional as well as modern tests used to detect the presence of

GAS in symptomatic children (direct carbohydrate antigen, DNA probe, and sheep blood agar culture) all perform significantly better on specimens obtained from the throat.

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