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Evaluation of a two-minute strep A direct swab test (SADST) on patients with pharyngitis at a primary care clinic

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SUMMARY

A two-minute strep A direct swab test (SADST) was used to detect the presence of Lancefield group A streptococci (GAS) from the throats of 207 patients with pharyngitis at a primary-care clinic. The results were compared with a standard culture method. Fifty-one specimens were positive and 156 specimens were negative for GAS by culture. The SADST had a sensitivity of 96 % (49 of 51) and specificity of 98·7 % (154 of 156). The predictive values of a positive and negative SADST, for GAS, were 96 % and 98·7 % respectively. The SADST showed negative reactions with five specimens containing beta-haemolytic streptococci other than GAS and 34 known stock cultures other than GAS. Our results indicate that SADST is a rapid, simple, convenient and reliable test to use for diagnosis of GAS pharyngitis at primary care clinics, physicians' offices and clinical laboratories.

INTRODUCTION

Early diagnosis and antibiotic treatment of Lancefield group A streptococcal (GAS) pharyngitis reduce the subsequent attack rate of acute rheumatic fever (ARF), minimize morbidity and decrease the risk of transmitting the organism (Moody, 1972; Stollerman, 1975; Wannamaker, 1958). The clinical diagnosis of GAS pharyngitis and its differentiation from other etiologies is difficult and unreliable (Breese & Disney, 1954; El-Batish, Mark & Majeed, 1985; Kaplan, 1980; McCracken, 1982). Therefore, throat swabs have been performed on patients with pharyngitis to confirm or exclude the presence of GAS by different methods. Most clinical laboratories use traditional throat culture methods, which take 24–48 h for isolation and identification of GAS and may result in a number of false-negatives due to interference of other microorganisms or if the patient is receiving antibiotics (Tagg, Dajani & Wannamaker, 1976; Taranta & Moody, 1971).

A new approach to rapid diagnostic methods was reported to identify GAS antigens directly from throat swabs using extraction procedures with nitrous acid

(El-Kholy *et al.* 1978; Gerber, 1983), trypsinization (Edwards, Phillips & Suiter, 1982) and lytic enzymes of *Streptomyces griseus* (Otero, Reyes & Noriega, 1983). These procedures have shown high specificity (86–100%), lower sensitivity (77–89%) and results were available in 30–90 min of testing. Subsequently several commercial kits were introduced for direct detection of GAS from throat swabs. These showed high specificity (98–100%) when tested with known culture strains (Facklam, 1985), and high sensitivity but their performance time varied. For example, the latex agglutination Directigen GAS test (Hynson, Wescott & Dunning, Baltimore Md.) takes 65–70 min. (McCusker *et al.* 1984; Miller *et al.* 1984) and it is 10 min. for the Culturette GAS ID (Marion Scientific, Kansas City Mo.) (Chang & Mohla, 1985; Slifkin & Gil, 1984). On the other hand a 2 min strep A direct swab test (SADST) (Pharmacia Diagnostic, Uppsala, Sweden) was reported using a 1 min extraction procedure of group specific polysaccharide antigen and identifying it by coagglutination technique (Ogay & Bille, 1985).

This study evaluates the performance of the 2 min strep A direct swab test (SADST) on patients with pharyngitis at a primary care clinic.

MATERIALS AND METHODS

Study location

This study was carried out at Midan Hawalli primary care clinic in Kuwait, from December 1984 through March 1985.

Patients

The criteria for selecting patients suspected of having GAS pharyngitis were previously reported (El-Batish, Mark & Majeed, 1985; El-Kholy *et al.* 1973); 207 patients with symptomatic pharyngitis were included in this study. A clinical diagnosis of pharyngitis was made in the presence of the following signs and symptoms; fever (38 °C), sore throat and signs of inflammation in the throat. The presence of enlarged cervical lymph glands, dysphagia or a pharyngeal exudate was not considered essential for the diagnosis, as the classical syndrome of exudative pharyngitis characterized by 'redness and oedema of the throat, presence of an exudate on the tonsils or tonsillar fossae and enlargement and tenderness of the anterior cervical nodes' (Stollerman, 1975) was rarely encountered in the studies of El-Kholy *et al.* (1973) in Egypt and Karoui *et al.* in Kuwait (1982).

Throat-swab specimens

A rayon-tipped swab (Culturette, Marion Scientific Kansas City, Mo.) was used to swab both tonsils and oropharynx from each of the 207 patients. Each swab was first used to inoculate a sheep-blood agar plate and then tested immediately with the SADST kit reagents according to the manufacturers instructions.

Swab culture

The inoculum on trypticase soy agar (BBL Microbiology systems Cockeysville, Md., plate containing 5% sheep blood) was spread by loop and stabbed into the agar (Facklam, 1980) for isolation of single colonies; it was incubated at 35 °C in 5% CO_2. Each plate was examined at 24 and 48 h for the presence of beta-haemolytic

streptococci. The number of beta-haemolytic colonies per plate was estimated as follows: 1–10 colonies; 11–50 colonies; 51–100 colonies; and more than 100 colonies. The person who read the culture did not know of the antigen-detection test results.

Beta-haemolytic streptococci were grouped serologically by the coagglutination technique (Christensen, Kahlemter & Johnson, 1979) using a Phadebact test kit (Pharmacia).

Direct GAS antigen detection test

After inoculation of the blood agar plate, the swab was placed in a glass tube containing sodium nitrite solution and glycine HCl buffer and heated at 100 °C for 1 min. Equal volumes of extraction solution and GAS specific antibodies bound to protein A of staphylococci (Reagent A) were mixed for 1 min and observed for coagglutination. Control reagents, negative antibodies similarly bound to staphylococci, were included in each test. Positive results (presence of streptococcus group A antigen) were recorded when coagglutination occurred in the strep A reagent with no or only a very weak reaction in the control reagent. Negative results (streptococcus group A antigen not present) were recorded when a very weak reaction occurred in the strep A reagent together with a very weak reaction in the control reagent or no reaction in either strep A and or control reagent.

Specificity study

Known bacterial stock cultures were used to test the SADST for specificity. These cultures included 36 streptococci (12 group A, 12 group B, 2 group C, 7 group D, 1 group F, 2 group G) and 10 *Staphylococcus aureus*. A suspension in physiological saline equivalent to McFarland Density Tube No. 1 was made from each culture. A rayon-tipped swab was soaked in each suspension and pressed against the edge of the tube while being removed. The same direct GAS antigen detection test was done on each swab.

RESULTS

Two hundred and seven throat swabs were evaluated by culture and SADST. Fifty-one specimens (24·6 %) were positive for GAS by culture, 49 (23·6 %) were positive by SADST. The SADST had a sensitivity of 96 % (49 of 51). There were two false-positive reactions; the specificity of SADST was 98·7 % (154 of 156). The predictive values of a positive and negative SADST were 96 % (49 of 51) and 98·7 % (154 of 156) respectively.

The difference in agglutination reaction between reagent A (test) and control was very clear for the true positive and false positives. Clear negative reactions between reagent A and control was observed for the two false negative specimens and 122 of 154 (79 %) true negative specimens. Among the remaining true negative specimens, 13 (8 %) showed weak positive reactions in both the test and control reagents while 19 (12·3 %) showed thread-like reactions in both the test and control reagents.

Distribution of SADST results according to GAS colony count is summarized in Table 1. The GAS grew in pure culture from 10 specimens and were predominant

Table 1. *SADST results according to quantitative distribution of group A streptococci (GAS)*

Colony quantitation (no. of GAS colonies)	SADST results Number (percent) +ve		−ve	
0	2*	(1·3)	154	(98·7)
1–10	13	(87)	2†	(13)
11–50	8	(100)	—	
51–100	5	(100)	—	
> 100	23	(100)	—	

* Patients received antibiotics before coming to the clinic.
† One culture showed two colonies and the second culture showed seven colonies.

in culture for 15 specimens. The remaining specimens (24) showed mixed growth of GAS with other micro-organisms. One of the two false negative SADST had yielded two colonies on culture while the other yielded seven colonies.

A total of five cultures (2·4 %) showed beta-haemolytic streptococci other than GAS: one was group B, one was group C, and three were group G. All were negative by SADST.

In the specificity study only the 12 GAS were SADST-positive. None of the other tested bacteria reacted with the SADST.

DISCUSSION

A SADST is one of the tests employed for detection of GAS directly from throat swabs. Such tests fall within the rapid diagnostic methodologies that are changing approaches to clinical microbiology (Edwards, Phillips & Suiter, 1982; Petts, 1984).

Our study showed that SADST enables the report of detection or exclusion of GAS, directly from throat swabs, within 2·5–3 min. of receipt of specimens. The SADST is accurate since it showed high specificity (98·7 %) and sensitivity (96 %). It was able to detect the presence of GAS in mixed cultures even at very few colony counts (1–10). In addition, no reaction was observed with the five specimens containing beta-haemolytic streptococci other than GAS. The SADST reliability was also confirmed in our study with different groups of streptococci and staphylococci. Reading and interpreting the SADST results for the test and control reagents was easily done. However, one has to be careful in reading and interpreting those tests which show thread-like reactions or weak positive reactions in the test and control reagents. In this study 15 % of the 207 swabs gave such reactions. Without using the control reagents these specimens could be falsely read as positive.

The SADST showed very low (0·9 %) false-positive and false-negative reactions. The two specimens which were negative by SADST and positive by culture (false-negative) are difficult to explain. These represented 2 of 15 specimens from which fewer than 10 GAS colonies were isolated. Since one swab was used to perform both tests, it is probable that the remaining streptococci were not sufficient for detection by SADST. On the other hand the two specimens which

were positive by SADST and negative by culture (false-positives) could be due to the antibiotics received (penicillin for 2 days) by those two patients before visiting the clinic. If this was the reason, then such a feature would be advantageous for direct antigen detection systems since they do not rely on viable bacteria. On the other hand these two specimens may have had small amounts of GAS or may have contained non-haemolytic GAS mutants (James & McFarland, 1971) which were overgrown or inhibited by normal throat flora (Tagg, Dajani & Wannamaker, 1976).

In this study, GAS was the causative agent in about 25% of pharyngeal infections. Such incidence is similar to that previously reported from Kuwait (El-Batish, Mark & Majeed, 1985) and slightly higher than those reported from other parts of the world, e.g. India (Koshi & Myers, 1971), Egypt (El-Kholy *et al.* 1973) and the USA (Dillon & Derrick, 1974). The majority of pharyngeal infections were usually due to viruses which require symptomatic treatment only.

Clinically, it is difficult to differentiate GAS from other aetiologies of pharyngitis (Breese & Disney, 1954; Kaplan, 1980; McCracken, 1982; Stollerman, 1975; Wannamaker, 1958). A previous study from the same primary-care clinic reported a high false-positive (76%) and false-negative (46%) diagnosis of GAS pharyngitis based on clinical findings alone (El-Batish, Mark & Majeed, 1985). This meant that the majority of patients with pharyngitis would have been given antibiotics unnecessarily while almost half of the patients with GAS pharyngitis would have been denied antibiotic treatment and left with the possibility of developing its non-suppurative sequelae. Similar findings were also reported in other studies (Breese & Disney, 1954; Kaplan, 1980; McCracken, 1982; Stollerman, 1975; Wannmaker, 1958). The SADST would help diagnosis since it is easy to perform and suitable for use in primary care clinics, physicians' offices and clinical laboratories. Its rapid results would allow the physician to treat promptly and appropriately and thus save the patient from the inconvenience of a second visit and delay of treatment while waiting for culture results. This would also be effective in programmes for the primary prevention of ARF and rheumatic heart disease as well as antibiotic use in primary care (Moody, 1972).

Cost analysis was not done in this study. The SADST cost may be greater than that of culture alone. However, SADST would save on man-power and kits used for grouping of isolates and ultimately may prove to be cost-effective in care of patients with pharyngitis.

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