THE SPECIFICITY AND SENSITIVITY RESULTS OF THE RAPID ANTIGEN TEST USED IN THE DIAGNOSIS OF GROUP A BETA HEMOLYTIC STREPTOCOCCAL TONSILLOPHARYNGITIS

HATICE ULUDAG ALTUN, TUBA MERAL, EMEL TURK ARIBAS
Turgut Ozal University, Faculty of Medicine, Department of Clinical Microbiology, Turkey

ABSTRACT

Aims: *The rapid antigen detection test and throat culture can be used for the diagnosis of group A beta hemolytic streptococcus (GABHS) infections. The aim of this study was to determine the sensitivity and specificity ratios of the rapid antigen test for GABHS in the laboratory setting.*

Materials and methods*: In this study, the throat culture and rapid antigen test results were evaluated for 5120 patients between the ages of 0-18 years, who were admitted between January and June 2014 to the pediatric outpatient clinic with a diagnosis of clinical exudative tonsillopharyngitis. The tests of these patients were performed at the microbiology laboratory of Turgut Ozal University Hospital. Patients with only a throat culture or a rapid antigen test result were excluded from the study. Thus, 1243 patients were included in the current study, in which both tests had been performed. Two throat swab samples were collected from these patients. Culture tests and rapid antigen tests were both performed for the patient samples. The Strep A Abon kit [Hangzhou, China] was used as the rapid antigen test.*

Results*: Nine hundred thirty-six patients had no bacterial growth in their throat cultures, while 307 throat cultures were positive for GABHS. The sensitivity and specificity of the rapid antigen test were 73% and 96.8%, respectively. The positive and negative predictive values for the rapid antigen test were 88.2% and 91.6%, respectively.*

Conclusion*: Although the specificity of the rapid antigen test used in this study was high (96.8%), its sensitivity was determined to be lower (73%). Therefore, for patients in whom negative test results are obtained, it would be appropriate to confirm the test results with throat cultures.*

Key words*: Group A beta hemolytic streptococcus, tonsillopharyngitis, rapid antigen test, sensitivity, specificity.*

Received *June 18, 2014;* ***Accepted*** *October 02, 2014*

Introduction

Acute tonsillopharyngitis is one of the most common infections in childhood. Group A beta hemolytic streptococcus (GABHS) causes 15-30% of cases of acute tonsillopharyngitis in pediatric cases[1]. GABHS tonsillopharyngitis is primarily a disease of school-aged children and adolescents. Under three years old, it is rare. The peak incidence in temperate climates occurs during winter and early spring[2]. Accurate diagnosis and treatment of GABHS tonsillopharyngitis provides various benefits, including the prevention of complications such as acute rheumatic fever and peritonsillar abscess, and the reduction of acute morbidity associated with the disease. Conversely, incorrect diagnosis, unnecessary antibiotic use, and the resulting increase in health care costs can lead to various negative consequences, including the development of bacterial resistance[1-7].

Throat culture is the gold standard for the laboratory diagnosis of GABHS infection. If the sample is collected appropriately, GABHS growth will be observed in the culture of 90-95% of patients with typical clinical symptoms[8]. However, the completion of throat culture tests may require as long as 24-48 hours. Given the importance of early diagnosis and early antibiotic therapy in life-threat-

ening infections caused by GABHS, the culture tests requiring 24-48 hours to provide results, as well as the possibility of obtaining negative results in patients who use antibiotics, represent significant disadvantages. On the other hand, the rapid antigen tests used to identify GABHS in throat swab samples can be performed rapidly, and the results of these tests can be obtained within a short period of time. There are a variety of commercially available rapid diagnostic kits. Tests that detect group A specific carbohydrates from bacterial cell wall in throat swabs generally employ the membrane-bound immunoassay method. The current study aimed to determine the sensitivity and specificity rates of the rapid antigen test used in the laboratory.

Materials and methods

In the current study, the throat culture and rapid antigen test results were evaluated for 5120 patients between the ages of 0-18 years, who were admitted between January and June 2014 to the pediatric outpatient clinic with a diagnosis of clinical exudative tonsillopharyngitis. The tests of these patients were performed at the microbiology laboratory of Turgut Ozal University Hospital. Patients with only a throat culture or a rapid antigen test result were excluded from the study. Thus, 1243 patients were included in the study, in which both tests had been performed. Two throat swab samples were collected from these patients. Culture tests and rapid antigen tests were both performed on the patient samples. Throat swab samples were taken using sterile swab tubes (Copan, Italy).

The samples were sent to the microbiology laboratory in appropriate conditions. The swab samples were cultivated on 5% sheep blood agar (Salubris, Turkey), and the plates were incubated at 37°C for 16-18 hours. Following the incubation period, the colonies were identified as beta hemolytic streptococci based on colony morphology, Gram staining, and the catalase test. The bacitracin susceptibilities of these colonies were then investigated using 0.04 bacitracin discs (BBL™, BD). The Strep A Abon kit [Hangzhou, China] was used as the rapid antigen test. In accordance with the manufacturer's recommendations, four full drops of Reagent A (approximately 240 µl) and four full drops of Reagent B (approximately 160 µl) were added to the extraction tube and stirred. The throat swab samples were added to the extraction tube. The mixture was vortexed, and then incubated at 25°C for 1 minute. After the removal of the swab, three full drops of the mixture (approximately 100 µl) were added to the sample compartment of the kit. The results were obtained and evaluated five minutes later. Because the intensity of the color in the test line region (T) can vary depending on the concentration of GABHS in the sample, pale color changes in the test line were considered to be positive results.

Results

All patients were between the ages of 0 and 18 years, while the mean age was 5.45 ± 3.07 years (mean age ± standard deviation). Of these patients, 640 were male, while 603 were female. In addition, 936 patients had no bacterial growth in their throat cultures, while 307 had positive throat cultures for GABHS. One thousand one hundred seventy-eight patients were referred from the general pediatrics outpatient clinic, 29 patients were referred from the pediatric metabolism clinic, 33 patients were referred from the pediatric nephrology clinic, one patient was referred from the pediatric cardiology clinic, and one patient was referred from the child and adolescent psychiatry clinic. The throat swab samples of 936 patients had no bacterial growth in the culture of the throat; the GABHS was isolated in 307 samples of the patients.

Of the 1243 throat swab samples administered with the rapid antigen test, negative results were obtained for 989 of the samples, while positive results were obtained for 254 of the samples (Table 1).

	Culture (+)	Culture (-)	Total
Rapid antigen test (+)	224	30	254
Rapid antigen test (-)	83	906	989
Total	307	936	1243

Table 1: Throat culture and rapid antigen test results for all the patients.
** Rapid test PPV: 88.2%*
*** Rapid test NPV: 91.6%*
(+):Positive, (-):Negative, PPV: Positive Predictive Value, NPV: Negative Predictive Value

Sensitivity and specificity of rapid antigen tests were 73% and 96.8%, respectively, while the positive and negative predictive values of the test were 88.2% and 91.6%, respectively.

Discussion

Throat culture is the gold standard for the laboratory diagnosis of GABHS tonsillopharyngitis. Rapid antigen tests used in the diagnosis of GABHS have high specificity rate. According to the results of previous studies conducted in Turkey, the specificity of several rapid antigen tests were determined to be within the 90-99% range, while the sensitivity was determined to be within the 60-70% range[9-12]. In a meta-analysis encompassing 24 studies conducted between the years 2000-2009, Ruiz-Aragon et al.[13] determined that the specificity and sensitivity range of the rapid antigen tests were 68.7-99.3% and 65.6-96.4%, respectively, while its positive and negative predictive value ranges were 87.8-98% and 59.4-97.4%, respectively. In the current study, the specificity, sensitivity, positive predictive value, and positive and negative predictive were determined as 96.8%, 73%, 88.2%, and 91.6%, respectively. Similar to the current study, the test sensitivities observed in previous studies[9-13] were lower than the specificities.

In a study conducted in Turkey, Coban et al.(10) evaluated throat cultures and rapid antigen tests in 2163 patients, and determined that the sensitivity and specificity of the rapid antigen tests were 68.1%, 92.2%, respectively. Although this research is valuable because of the sufficient number of cases included, the study was performed with two different rapid antigen test kits [Ecotest ® Strep A rapid test kit (Wellkang, UK) or SD ® rapid test kit (Standard Diagnostics, Korea)] without separately evaluating or distinguishing the sensitivity and specificity of each kit. Because the sensitivity and specificity of each kit may be different, the data collected for different kits should not be evaluated together. In the current study, all samples were evaluated with the same kit. If the same samples had been evaluated with a different rapid test kit, differing results would have likely been obtained.

In their study, Kurtz et al.[14] observed that increasing the quantity of worked material in order to increase the number of bacteria in the samples, such as processing together two separately taken throat swabs (as if they were one swab), had the effect of significantly improving the sensitivity of the direct diagnostic assay. In another study, it was observed that the sensitivity of the rapid antigen test varied significantly between patients with light and according to the number of GABHS colonies in their throat culture plates.

In the same study, light inoculum was identified in 65.8% of false-negative rapid antigen test patients, and in 6.9% of true-positive patients[15].

These studies suggest that increasing number of GABHS colonies in the throat swab samples of the patients also increases the sensitivity of the rapid tests. Ezike et al.[16] selected two groups consisting of 117 and 186 patients between the ages of 5-18. In the first group, one throat swab sample was taken from the patients, which was used both for throat culture cultivation and for rapid antigen testing. In the second group, two throat swab samples were taken from the patients, and each patient's samples was evaluated separately with the rapid antigen test and the throat culture method. The researchers found that the use of two throat swabs instead of one swab did not increase the sensitivity of the rapid antigen test. The effect of the number of bacteria colonies and the number of swab samples taken from patients on the sensitivity of the rapid antigen test was not evaluated in the current study.

The evaluation of rapid diagnostic tests by trained personnel in a laboratory environment is important. In a study conducted by Fox et al.[17], the sensitivity of rapid antigen tests were higher when they were performed by laboratory personnel compared to those performed by non-laboratory personnel. In a study evaluating rapid antigen tests and throat culture results in hospital laboratories and community pediatric offices, it was observed that 41 patients had positive throat culture results in the offices and negative culture results in the laboratories, while 105 had negative throat culture results in the offices and positive throat culture results in the laboratories[18]. Therefore, the quality of the swab samples, proper sample collection, and the experience of the personnel performing the test are all important for the sensitivity of the rapid antigen test and for accurate evaluations. In the current study, trained laboratory personnel performed the rapid antigen tests. Therefore, the authors believe that the current study results had high accuracy.

Although clinical signs are important for the diagnosis of GABHS tonsillopharyngitis, laboratory confirmation is necessary to clinically distinguish viral tonsillopharyngitis and GABHS[19]. However, it should not be ignored that throat cultures and rapid antigen tests that can also be used in the diagnosis of laboratory can not distinguish GABHS carriers with viral tonsillopharyngitis[19].

Furthermore symptomatic patients with asymptomatic carriers cannot be distinguished with these tests[20,21]. Therefore, the evaluation of laboratory results with clinical findings is important for the correct diagnosis of these patients.

In conclusion, cultures are still the gold standard in the laboratory diagnosis of GABHS tonsillopharyngitis. Although rapid antigen tests are used recently in the diagnosis of GABHS, many of these tests have low sensitivities. Therefore, it is important to confirm negative rapid antigen test results with cultures.

References

1) Vicedomini D, Lalinga G, Lugli N, D'Avino A. *Diagnosis and management of acute pharyngotonsillitis in the primary care pediatrician's office*. Minerva Pediatr 2014; 66: *69-76*.
2) Chiappini E, Regoli M, Bonsignori F, Sollai S, Parretti A, Galli L, de Martino M. *Analysis of different recommendations from international guidelines for the management of acute pharyngitis in adults and children*. Clin Ther 2011; 33: *48-58*.
3) Ozkaya Parlakay A, Uysal M, Kara A. G*roup A streptococcal tonsillopharyngitis burden in a tertiary Turkish hospital*. Turk J Pediatr 2012; 54: *474-7*.
4) Mazur E, Bochyńska E, Kochańska E, Kozioł-Montewka M. *Utility of clinical differentiation and microbiological examination for appropriate antibiotic therapy of acute pharyngotonsillitis in children*. B.Pol Merkur Lekarski 2011; 31: *31-6*.
5) Brook I, Dohar JE. *Management of group A beta-hemolytic streptococcal pharyngotonsillitis in children*. J Fam Pract 2006; 55: *1-11*.
6) Steinhoff MC, Abd el Khalek MK, Khallaf N, Hamza HS, el Ayadi A, et al. *Effectiveness of clinical guidelines for the presumptive treatment of streptococcal pharyngitis in Egyptian children*. Lancet 1997; 350: *918-21*.
7) Mazur E, Bochyńska E, Juda M, Kozioł-Montewka M. *Empirical validation of Polish guidelines for the management of acute streptococcal pharyngitis in children*. Int J Pediatr Otorhinolaryngol 2014; 78: *102-6*.
8) Bisno AL, Gerber MA, Gwaltney JM, Gwaltney Jr, Kaplan EL, Schwartz RH. *Infectious Diseases Society of America: Practice guidelines for the diagnosis and management of group A streptococcal pharyngitis*. Clin Infect Dis 2002; 35: *113-25*.
9) Gurol Y, Akan H, Izbirak G, Tekkanat ZT, Gunduz TS, et al. *The sensitivity and specificity of rapid antigen test in streptococcal upper respiratoryinfections*. Int J Pediatr Otorhinolaryngol 2010; 74: *591-3*.
10) Coban B, Kaplan H, Topal B, Ulku N. *The sensisivity and specificity of rapid antigen test in group A streptococcal tonsillopharyngitis*. J Pediatr Inf 2013; 7: *143-6*.
11) Sarikaya S, Aktas C, Ay D, Cetin A, Celikmen F. *Sensitivity and specificity of rapid antigen detection testing for diagnosing pharyngitis in the emergency department*. Ear Nose Throat J 2010; 89: *180-2*.
12) Gozukucuk R, Gocmen I, Nas Y, Yılmazer F, Unuvar E. *The Effectiveness of Rapid Strep A Test in the Diagnosis of Streptococcal Tonsillopharyngitis*. J Child 2011; 11: *157-9*.
13) Ruiz-Aragón J, Rodríguez López R, Molina Linde JM. *Evaluation of rapid methods for detecting Streptococcus pyogenes. Systematic review and meta-analysis*. JM. An Pediatr (Barc). 2010; 72: *391-402*.
14) Kurtz B, Kurtz M, Roe M, Todd J. *Importance of inoculum size and sampling effect in rapid antigen detection for diagnosis of Streptococcus pyogenes pharyngitis*. J Clin Microbiol 2000; 38: *279-81*.
15) Cohen JF, Chalumeau M, Levy C, Bidet P, Thollot F, et al. *Spectrum and inoculum size effect of a rapid antigen detection test for group A streptococcus in children with pharyngitis*. PLoS One 2012; 7: *e39085*.
16) Ezike EN, Rongkavilit C, Fairfax MR, Thomas RL, Asmar BI. *Effect of using 2 throat swabs vs 1 throat swab on detection of group A streptococcus by a rapid antigen detection test*. Arch Pediatr Adolesc Med 2005; 159: *486-90*.
17) Fox JW, Cohen DM, Marcon MC, Cotton WH, Bonsu BK. *Performance of Rapid Streptococcal Antigen Testing Varies by Personnel*. J Clin Microbiol 2006; 44: *3918-22*.
18) Tanz RR, Gerber MA, Kabat W, Rippe J, Seshadri R, Shulman ST. *Performance of a rapid antigen-detection test and throat culture in community pediatric offices: implications for management of pharyngitis*. Pediatrics 2009; 123: *437-44*.
19) *Report of the Committee on Infectious Diseases, Group A Streptococcal Infections*, In: Pickering LK, ed. American Academy of Pediatrics 2012 Red Book. Elk Grove Village, 2012: *668-80*.
20) Yilmaz Ciftdogan D, Vardar F. *Rapid diagnostic tests for infectious diseases*. Turkish Pediatric Journal 2009; 52: *159-66*.
21) Mazur E, Bochyńska E, Juda M, Kozioł-Montewka M. *Empirical validation of Polish guidelines for the management of acute streptococcal pharyngitis in children*. Int J Pediatr Otorhinolaryngol 2014; 78: *102-6*.

Correspoding author
Assist. Prof. HATICE ULUDAG ALTUN
Turgut Ozal University, Faculty of Medicine, Department of Clinical Microbiology
Bestepe, 06510, Ankara
(Turkey)

View publication stats