PREVALENCE OF GROUP A BETA-HEMOLYTIC STREPTOCOCCAL PHARYNGITIS IN CHILDREN WITH ACUTE SORE THROAT, NARESUAN UNIVERSITY HOSPITAL, PHITSANULOK, THAILAND (MAY 2016 - APRIL 2017)

Sarunya Srijuntongsiri and Klaita Srisingh

Department of Pediatrics, Faculty of Medicine, Naresuan University Hospital, Phitsanulok Province, Thailand

Abstract. Streptococcal pharyngitis is a common bacterial pharyngotonsillitis in a pediatrics outpatient department setting. A rapid antigen detection test (RADT) was used in a prospective study in diagnosing group A beta-hemolytic streptococci (GAS) in children 1-15 years of age with fever and sore throat. Throat swabs were collected for culture and RADT from children (n = 134, median age = 5 years) experiencing fever and sore throat within the previous seven days of symptoms onset, who were treated at the Pediatric Outpatient Department, Naresuan University Hospital, Phitsanulok, Thailand from May 2016 to April 2017. All patients received regular care from physicians and information on patients' illness, diagnoses and antibiotic prescribed were collected. Acute pharyngitis was diagnosed in 43% of the children and GAS detected in 7% of throat swab cultures, with a prevalence of GAS pharyngitis of 7% (95% confidence interval (CI): 3-12). All GAS isolates were susceptible to chloramphenicol, penicillin and vancomycin, and 70% resistant to tetracycline. RADT demonstrated a sensitivity of 90% (95% CI: 85-95) and specificity of 91% (95% CI: 86-96), with positive and negative predictive value of 45 and 99%, respectively. In conclusion, the study shows low prevalence of GAS in children with acute pharyngitis and empirical antibiotic treatment should be avoided until a definitive diagnosis of GAS infection has been made.

Keywords: children, group A streptococcal pharyngitis, rapid antigen detection test, sore throat

INTRODUCTION

Fever with sore throat is one of the most frequently reported complaints among pediatric outpatients and is usually of viral origin (DuBose, 2002). However, it is difficult to diagnose pharyngitis stemming from viral or group A beta-hemolytic streptococci (GAS) infection just from signs and symptoms (Esposito *et al*, 2004). The majority of cases related bacterial pharyngotonsillitis is due to GAS, with prevalence depending on age group, namely, 20-30% in children and

Correspondence: Klaita Srisingh, Department of Pediatrics, Faculty of Medicine, Naresuan University, 99 Moo 9 Phitsanulok-Nakhon Sawan Road, Tha Pho Subdistrict, Phitsanulok 65000, Thailand
Tel: +66 (0) 5596 5515, +66 (08) 9703 6724;
Fax: +66 (0) 5596 5478
E-mail: sarnyachin@gmail.com, klaitas@nu.ac.th

5-15% in adults, and rarely due to group C and G streptococci, *Corynebacterium diphtheriae* or *Fusobacterium necrophorum* (Carapetis *et al*, 2005; Lindbaek *et al*, 2005; Shulman *et al*, 2012; Cohen *et al*, 2016). Clinical signs and symptoms of pharyngitis due to GAS are generally nonspecific and difficult to diagnose from viral causes such adenovirus and Epstein-Barr virus (Green, 1998). Infectious mononucleosis from Epstein-Barr virus infection presents with exudative tonsilitis, cervical lymphadenopathy and maculopapular rash (Johannsen and Kaye, 2015). Petechiae in palate and diffused scarlatiniform body rash are distinctive of streptococcal infection but otherwise are uncommon (Cohen *et al*, 2016). Appropriate treatment following detection of the causative agent can prevent subsequent suppurative and non-suppurative post-infection sequelae, such as acute rheumatic fever, post-streptococcal glomerulonephritis and other immunologically mediated pathologies (Gerber *et al*, 2009).

Throat swab culture and rapid antigen detection test (RADT) are able to distinguish difference between GAS and viral pharyngitis (Shulman *et al*, 2012). Accurate diagnosis enables appropriate antibiotic treatment to prevent complications and transmission, and shortens course of the disease (Wessels, 2011). The gold standard in confirming GAS pharyngitis diagnosis is examining throat swab cultures for beta-hemolytic colonies (Colletti and Robinson, 2005). Recent antibiotic use, culture procedures and specimen integrity are common factors, which can affect outcome of bacterial culture tests (Felsenstein *et al*, 2014). Tests based on cultures take up to 48 hours to obtain results, thereby delaying appropriate antimicrobial treatment. RADT was introduced to provide a more rapid diagnosis of GAS pharyngotonsillitis, identifying group A bacterial cell wall carbohydrates from an immunological assay from throat swab sample within a few minutes (Armengol *et al*, 2004). RADTs have sensitivity of 70-90% and specificity >95%, allowing physicians to correctly and timely and prescribe antibiotics based on the positive results (Rimoin *et al*, 2010; Arnold and Nizet, 2012; Pelucchi *et al*, 2012). In children, overall sensitivity and specificity of RADTs is 86 and 96% respectively (Lean *et al*, 2014, Stewart *et al*, 2014). RADT has a negative predictive value of ~95%, which is sufficient to confidentially exclude GAS infection. Factors influencing RADT sensitivity include, but are not limited to, specimen integrity and personnel competency. On the other hand, a negative RADT prompts a culture test especially in regions with high prevalence of cardiac-related diseases (Leung *et al*, 2006).

Here, prevalence of children with group A streptococcal pharyngitis was determined employing both culture and a RADT (allowing validation) at Naresuan University Hospital, Phitsanulok Province, Thailand.

MATERIALS AND METHODS

Participants and study design

A prospective study involving children attending the Pediatric Outpatient Department, Naresuan University Hospital, Phitsanulok Province, Thailand, was carried out from May 2016 to April 2017. Inclusion criteria were children 1-15 years of age with fever and sore throat within the previous seven days. Exclusion criteria were children with lower respiratory tract infection, oral ulcer, herpangina, herpetic gingivostomatitis,

and immunocompromised status. Data were collected on the participants' illness, results of physical examination, diagnosis and type of prior antibiotic administered (if pertinent).

The study protocol was approved by the Institutional Review Board, Faculty of Medicine, Naresuan University Hospital (COA no. 222/2015). Prior written consent was obtained from parents or legal guardian of each participant and assent from children 7-15 years of age.

Sample collection and laboratory tests

Each participant had two throat swabs taken by rubbing sterile rayon swab over both tonsils, posterior pharynx or in areas with exudates without touching tongue and lips. Samples for culture were immediately transported to the microbiology laboratory, Naresuan University Hospital, where they were plated onto 5% sheep blood agar (Biomedia (Thailand) Co Ltd, Nonthaburi, Thailand) and incubated at 35±2ºC for 20-24 hours under a humidified atmosphere containing 5% CO_2. Beta-hemolytic colonies were identified by Gram staining, catalase test and bacitracin susceptibility test (CLSI, 2017). The second swab was used for GAS antigen detection employing a Quicknavi™-Strep A kit (DenkaSeiken, Niigata, Japan).

Antibiogram profiling

Antimicrobial susceptibility test was carried out using disk diffusion method on Mueller sheep blood agar plate by measuring the zone of inhibition diameter according to the guidelines of Clinical Laboratory and Standards Institute (CLSI, 2017). Antimicrobial disks (Oxoid, Hampshire, UK and BD BBL™ Sensi-Disc™, NSW, Australia) contained penicillin (10 units), vancomycin (30 μg), erythromycin (15 μg), tetracycline (30 μg), chloramphenicol (30 μg). The data were interpreted as sensitive (S), intermediate (I), or resistant (R) following CLSI criteria.

Statistical analysis

Data are presented as percent with 95% confidence interval (CI) and median with interquartile range (IQR). RADT accuracy was measured using an area under receiver operating characteristic (ROC) curve employing a STATA version 12.0 software (StataCorp, College Station, TX), with a p-value of 0.05 or less is considered statistically significant.

RESULTS

Children (n = 134), median age of 5 years (IQR = 3-8 years), 73 (54%) boys and 61 (46%) girls, were recruited (Table 1). Median duration of symptoms was 3 days (IQR = 2-3 days) and 27% of participants received antibiotic treatment prior to visiting the Hospital. Cough was the most common symptom (85%), followed by rhinorrhea (77%) and headache (45%). Physical examination revealed 84%, 28% and 27% of the children patients had tonsillar enlargement, tonsillar exudate or patch and palatal petechiae, with 43% diagnosed with acute pharyngitis (Table 1).

Following examination and diagnosis, children (n = 92) were treated with amoxicillin (n = 68; 74%), amoxicillin-clavulanate (n = 8; 9%), cephalosporins (n = 7; 8%), macrolides (n = 6; 6%), and other types of antibiotics (one person each received amoxicillin-clindamycin, cephalosporin-macrolide and meropenem).

Throat swab cultures indicated 21 patients (16%) with pathogenic growth, group A beta-hemolytic streptococci, *Staphylococcus aureus* and *Streptococcus pneumoniae* identified in 10 (7%), 6 (4%), and 5 (4%) of the samples, respectively.

Table 1
Demographic profiles, clinical presentations and diagnosis of children with acute sore throat attending the Pediatric Outpatient Department, Naresuan University Hospital, Phitsanulok Province, Thailand (May 2016 - April 2017).

Characteristics	Number (%) (n = 134)
Gender	
Male	73 (54)
Female	61 (46)
Age in years	
Median (IQR)	5 (3 to 8)
Age group	
1-5 years	72 (54)
>5-10 years	40 (30)
>10-15 years	22 (16)
Duration of symptoms in days, median (IQR)	3 (2 to 3)
History of antibiotic medication prior to hospital visit	36 (27)
Sign/symptom*	
Cough	114 (85)
Rhinorrhea	103 (77)
Headache	61 (45)
Malaise	51 (38)
Vomiting	37 (28)
Myalgia	36 (27)
Hoarseness of voice	35 (26)
Conjunctivitis	20 (15)
Diarrhea	12 (9)
Drooling	12 (9)
Tonsillar enlargement	113 (84)
Tonsillar exudates/patches	38 (28)
Palatal petechiae	36 (27)
Lymph node enlargement	20 (15)
Conjunctival injection	11 (8)
Scarlatiniform rash	9 (7)
Tender at cervical lymph nodes	8 (6)
Diagnosis	
Acute pharyngitis	57 (43)
Acute exudative tonsillitis/acute tonsillitis	54 (40)
Influenza	9 (7)
Upper respiratory tract infection/common cold	6 (4)
Scarlet fever	3 (2)
Infectious mononucleosis	2 (1)
Acute febrile illness	2 (1)
Acute lymphadenitis	1 (1)

*One child could have had more than one symptom; IQR: interquartile range.

Twenty (15%) throat swab samples produced positive GAS antigen RADT results, with one, nine and ten being from *Staphylococcus aureus*-positive, GAS-positive and pathogen-negative culture(s), respectively. Overall, prevalence of GAS in throat swabs was 7%. One of 10 children (10%) with positive throat swab culture and 5/20 (25%) with positive RADT results had a history of antibiotic use before the hospital visit, mainly treatment with amoxicillin (33%), amoxicillin-clavulanate (8%) and cephalosporins (8%).

Using a disc diffusion assay, among GAS isolates ($n = 10$), all were susceptible to chloramphenicol, penicillin, and vancomycin; seven were resistant to tetracycline and one was resistant to both erythromycin, and tetracycline. Sensitivity and specificity of RADT was 90% (95% CI: 85-95) and 91% (95% CI: 86-96), respectively, with positive and negative predictive value of 45% (95% CI: 37-53) and 99% (95% CI: 97-100), respectively. Area under ROC curve of RADT was 0.90 (Fig 1).

DISCUSSION

The study shows a prevalence of children diagnosed with GAS pharyngitis at Naresuan University Hospital from May 2016 to April 2017 was 7%, comparable with a prevalence (6.5%) observed among primary care patients with GAS pharyngitis in northern Thailand (Greer *et al*, 2020), while a prevalence of 3.3% was reported in Bangkok among adults presenting signs of upper respiratory

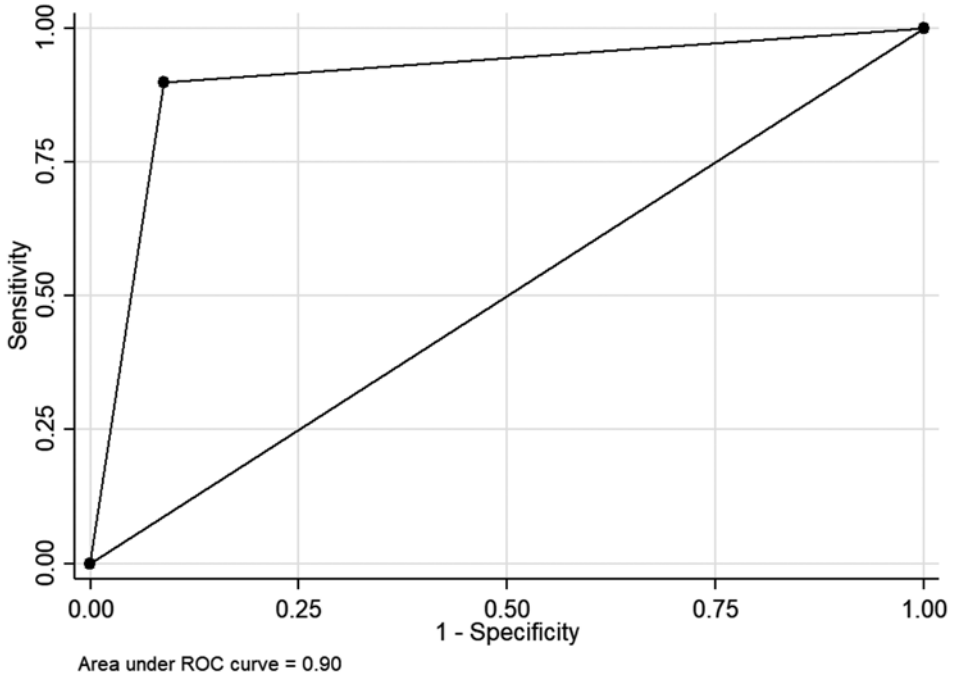

Fig 1-Receiver operating characteristic (ROC) curve for determination of accuracy of the rapid antigen detection test for group A streptococci used in the study.

X-axis: 1-specificity (false positive rate); Y-axis: sensitivity (true positive rate).

tract infection (Thamlikitkul *et al*, 2018). However, prevalence of GAS pharyngitis varies with country, region and age groups. A prevalence of 15% was reported among school-age children with GAS pharyngitis in developed nations (Carapetis *et al*, 2005) where there is an overall prevalence range 20-30% (Cohen *et al*, 2015). In a meta-analysis conducted in a previous study, prevalence of GAS pharyngitis is 37% among all children with sore throat and 24% among children <5 years of age (Shaikh *et al*, 2010).

The main reason for prescribing antibiotics is to treat GAS pharyngotonsillitis upon early diagnosis to prevent subsequent occurrence of suppurative and non-suppurative post-infection sequelae (Wessels, 2011). Although clinical manifestations are important for diagnosis of GAS pharyngotonsillitis, laboratory confirmation is necessary to clinically distinguish between viral and GAS pharyngotonsillitis (Kimberlin *et al*, 2018). In the present study, over half of the children presenting sore throat were prescribed antibiotics in the absence of any evidence of bacterial infection. Inappropriate antibiotic use leads to development of multi-drug resistant bacteria and consequently poses a serious challenge in pediatric practice (Tanz *et al*, 2004). A study conducted in Italy found GAS isolates resistant to clindamycin, erythromycin and tetracycline (Gherardi *et al*, 2015). In the present study, a high proportion of GAS isolates from children throat swabs were resistant to tetracycline but level of macrolide resistance was lower than that reported for erythromycin from China (>95% resistance) (Liu *et al*, 2009), France (22.4% resistance) (Bingen *et al*, 2004) and USA (48% resistance) (Martin *et al*, 2002).

Sensitivity and specificity of RADTs in the diagnosis of streptococcal pharyngitis ranges 83.3-87.6% and 94.5-96.2%, respectively (Arnold and Nizet, 2012; Pelucchi *et al*, 2012). Variability in RADT sensitivity might be due to differences in laboratory kits, quality of specimen collection and personnel experience. Throat culture is 90-95% positive in GAS patients with active symptoms (Wessels, 2011), but requires 1 to 2 days to obtain results. The positive predictive value of RADT used in the present study was lower than the range (67.9-88.6%) previously reported (Oliver *et al*, 2018) and conversely negative predictive value higher than that (88.0-95.7%) reported earlier (Rimoin *et al*, 2010); but these results depend on the type of RADT employed. Although the high accuracy of RADT provides a precise test results in screening patients for GAS infection and thereby reduces unnecessary prescription of antibiotics, in Thailand, RADT is not recommended in routine practice because it is more expensive than blood agar plate culture, and there are limited available data regarding its cost-effectiveness; nevertheless, in private practice a combination of clinical presentation and RADT result provides rapid decision whether antibiotic should be prescribed in an outpatient setting.

The present study suffers from two major limitations. Firstly, the number of GAS isolates obtained was very small, and secondly, throat swab culture was not performed to detect other groups of streptococci, such as groups C and G, that could be the cause of sore throat in children.

In summary, the study shows a 7% prevalence of group A streptococcal pharyngitis among children with sore throat examined at an outpatient department, Naresuan University Hospital from May 2016 to April 2017.

All group A streptococci isolates were sensitive to chloramphenicol, penicillin and vancomycin, but most were resistant to tetracycline. While RADT for detection of group A streptococci is not routinely available in an outpatient setting, it provides valuable tool for ruling out group A streptococcal pharyngitis, thereby reducing unnecessary antibiotic treatment and potential development of drug resistance.

ACKNOWLEDGMENTS

The authors would like to thank the personnel of the Pediatrics Outpatient Department, the head and staff of the Microbiology Laboratory, Naresuan University Hospital for their kind support, Ms Kornthip Jeephet, Research Center, Faculty of Medicine, Naresuan University for her valuable assistance in the statistical analysis of the data, and Ms Judely Marish Cañete, Ms Ajarn Daisy and Mr Aaron Fey, International Relations Section, Faculty of Medicine, Naresuan University for revising and editing the manuscript.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

REFERENCES

Armengol CE, Schlager TA, Hendley JO. Sensitivity of a rapid antigen detection test for group A streptococci in a private pediatric office setting: answering the Red Book's request for validation. *Pediatrics* 2004; 113: 924-6.

Arnold JC, Nizet V. Pharyngitis. In: Long SS, Pickering LK, Prober CG, editors. Principle and practice of pediatric infectious disease. 4th ed. Philadelphia: Elsevier Saunders; 2012. p. 199-205.

Bingen E, Bidet P, Mihaila-Amrouche L, *et al*. Emergence of macrolide-resistant *Streptococcus pyogenes* strains in French children. *Antimicrob Agents Chemother* 2004; 48: 3559-62.

Carapetis JR, Steer AC, Mulholland EK, Weber M. The global burden of group A streptococcal diseases. *Lancet Infect Dis* 2005; 5: 685-94.

Clinical and Laboratory Standards Institute (CLSI). M100: Performance Standards for Antimicrobial Susceptibility Testing. Wayne, PA: Clinical and Laboratory Standards Institute; 2017.

Cohen JF, Bertille N, Cohen R, Chalumeau M. Rapid antigen detection test for group A streptococcus in children with pharyngitis. *Cochrane Database Syst Rev* 2016; 7: CD010502.

Cohen JF, Cohen R, Levy C, *et al*. Selective testing strategies for diagnosing group A streptococcal infection in children with pharyngitis: a systematic review and prospective multicentre external validation study. *CMAJ* 2015; 187: 23-32.

Colletti T, Robinson P. Strep throat: guidelines for diagnosis and treatment. *JAAPA* 2005; 18: 38-46.

DuBose KC. Group A streptococcal pharyngitis. *Prim Care Update Ob Gyns* 2002; 9: 222-5.

Esposito S, Blasi F, Bosis S, *et al*. Aetiology of acute pharyngitis: the role of atypical bacteria. *J Med Microbiol* 2004; 53: 645-51.

Felsenstein S, Faddoul D, Sposto R, Batoon K, Polanco CM, Dien Bard J. Molecular and clinical diagnosis of group A streptococcal pharyngitis in children. *J Clin Microbiol* 2014; 52: 3884-9.

Green M. Nonstreptococcal pharyngitis. *Semin Pediatr Infect Dis* 1998; 9: 56-9.

Greer R, Althaus T, Ling C, *et al*. Prevalence of group A *Streptococcus* in primary care patients and the utility of C-reactive protein and clinical scores for its identification in Thailand. *Am J Trop Med Hyg* 2020; 102: 377-83.

Gerber MA, Baltimore RS, Eaton CB, *et al*. Prevention of rheumatic fever and diagnosis and treatment of acute Streptococcal pharyngitis: a scientific statement from the American Heart Association Rheumatic Fever, Endocarditis, and Kawasaki Disease Committee of the Council on Cardiovascular Disease in the Young, the Interdisciplinary Council on Functional Genomics and Translational Biology, and the Interdisciplinary Council on Quality of Care and Outcomes Research: endorsed by the American Academy of Pediatrics. *Circulation* 2009; 119: 1541-51.

Gherardi G, Petrelli D, Di Luca MC, *et al*. Decline in macrolide resistance rates among *Streptococcus pyogenes* causing pharyngitis in children isolated in Italy. *Eur J Clin Microbiol Infect Dis* 2015; 34: 1797-802.

Johannsen EC, Kaye KM. Epstein-bar virus (infectious mononucleosis, Epstein-Barr virus-associated malignant diseases, and other diseases). In: Bennett JE, Dolin R, Blaser MJ, editors. Mandell, Douglas, and Bennett's principles and practice of infectious diseases. 8th ed. Philadelphia, PA: Churchill Livingstone; 2015. p. 1754-71.

Kimberlin DW, Long SS, Brady MT, Jackson MA, editors. Group A Streptococcal infections. Red Book 2018-2021: Report of the Committee on Infectious Diseases. 31st ed. Elk Grove Village, IL: American Academy of Pediatrics; 2018. p. 748-62.

Lean WL, Arnup S, Danchin M, Steer AC. Rapid diagnostic tests for group A streptococcal pharyngitis: a meta-analysis. *Pediatrics* 2014; 134: 771-81.

Leung AK, Newman R, Kumar A, Davies HD. Rapid antigen detection testing in diagnosing group A beta-hemolytic streptococcal pharyngitis. *Expert Rev Mol Diagn* 2006; 6: 761-6.

Lindbaek M, Hoiby EA, Lermark G, Steinsholt IM, Hjortdahl P. Clinical symptoms and signs in sore throat patients with large colony variant beta-haemolytic streptococci groups C or G versus group A. *Br J Gen Pract* 2005; 55: 615-9.

Liu X, Shen X, Chang H, *et al*. High macrolide resistance in *Streptococcus pyogenes* strains isolated from children with pharyngitis in China. *Pediatr Pulmonol* 2009; 44: 436-41.

Martin JM, Green M, Barbadora KA, Wald ER. Erythromycin-resistant group A Streptococci in schoolchildren in Pittsburgh. *N Engl J Med* 2002; 346: 1200-6.

Oliver J, Wadu EM, Pierse N, Moreland NJ, Williamson DA, Bake MG. Group A *Streptococcus pharyngitis* and pharyngeal carriage: a meta-analysis. *PLoS Negl Trop Dis* 2018; 12: e0006335.

Pelucchi C, Grigoryan L, Galeone C, *et al*. Guideline for the management of acute sore throat. *Clin Microbiol Infect* 2012; 18 (Suppl 1): 1-28.

Rimoin AW, Walker CL, Hamza HS, *et al*. The utility of rapid antigen detection testing for the diagnosis of streptococcal pharyngitis in low-resource settings. *Int J Infect Dis* 2010; 14: e1048-53.

Shaikh N, Leonard E, Martin JM. Prevalence of streptococcal pharyngitis and streptococcal carriage in children: a meta- analysis. *Pediatrics* 2010; 126: e557-64.

Shulman ST, Bisno AL, Clegg HW, *et al*. Clinical practice guideline for the diagnosis and management of group A streptococcal pharyngitis: 2012 update by the Infectious Diseases Society of America. *Clin Inf Dis* 2012; 55: e86-102.

Stewart EH, Davis B, Clemans-Taylor BL, Littenberg B, Estrada CA, Centor RM. Rapid antigen group A streptococcus test to diagnose pharyngitis: a systematic review and meta-analysis. *PLoS One* 2014; 9: e111727.

Tanz RR, Shulman ST, Shortridge VD, *et al*. Community-based surveillance in the united states of macrolide-resistant pediatric pharyngeal group A streptococci during 3 respiratory disease seasons. *Clin*

Infect Dis 2004; 39: 1794-801.

Thamlikitkul V, Rachata T, Popum S, *et al*. Accuracy and utility of rapid antigen detection tests for group A beta-hemolytic *Streptococcus* on ambulatory adult patients with sore throat associated with acute respiratory infections at Siriraj Hospital. *J Med Assoc Thai* 2018; 101: 441-9.

Wessels MR. Clinical practice. Streptococcal pharyngitis. *N Engl J Med* 2011; 364: 648-55.