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The Extended Microbiology of Group A Streptococcal Pharyngitis. Observations During a Double-blind Controlled Study of Cephalexin Twice versus Four-times Daily

Summary: In a double-blind controlled study we compared the effectiveness of cephalexin b.i.d. versus q.i.d. in the treatment of group A streptococcal pharyngitis in 65 children. Clinical improvement was noted in 64 patients (98%) and bacteriologic cure in 60 (92%). Despite good compliance, three bacteriologic failures were noted in the q.i.d., and two in the b.i.d. treatment groups. Two of these five were carriers. Significant antibody responses were observed in 61% of the patients by at least one of three tests (ASO, anti-DNase B, Streptozyme). We also investigated the extended microbiology of streptococcal pharyngitis by looking for the presence of viruses, chlamydia and β-lactamase producing organisms in the pharynx. Respiratory viruses were isolated concomitantly with Streptococcus pyogenes in six patients. β-lactamase producing bacteria were present in the pharynx of 98% of the patients at the initiation of treatment and comprised 1–98% of the total bacterial flora. The β-lactamase producing flora did not significantly change with cephalexin therapy.


Introduction

Group A streptococcal pharyngotonsillitis is one of the most common infections in children. Eradication of the organism from the nasopharynx is important for epidemiological reasons and for the prevention of rheumatic fever (1). Penicillin is considered the treatment of choice for this infection; however, significant bacteriologic failure rates have been reported (2, 3). The reasons for these failures are unknown, since the organism has remained highly susceptible to penicillin in vitro (4, 5). During the last 10–15 years many other antibiotics have been evaluated for the treatment of group A streptococcal pharyngitis. Among the cephalosporins, cephalexin (2), cefadroxil (6) and cefaclor (7) have good in vitro activity and clinical efficacy. Stillerman et al. found that the bacteriologic failure rate was lower with cephalexin than with penicillin (2). Based on these reports, and the concept that a b.i.d. dose schedule might increase compliance, we designed a study to compare the effectiveness of cephalexin administered twice daily with the same total dose given four times daily. We also evaluated the serological responses to streptococcal antigens, and investigated the extended microbiology of streptococcal pharyngitis by looking for the concomitant presence of viruses, chlamydia, and β-lactamases in the pharynx of these patients.
Patients and Methods

The study population was comprised of children seen at the Oklahoma Children's Memorial Hospital outpatient department between October 1980 and April 1982, because of signs and symptoms of group A streptococcal pharyngitis. These include sore throat, fever and anterior cervical lymphadenopathy. Throat cultures were obtained and if group A Streptococcus was isolated, the parents were notified of the result and the study by telephone. The study nurse explained the purpose of the study, and, if verbal consent to participate in the study was obtained, the patient and a parent were asked to return to the Infectious Disease Clinic.

Study design: In the clinic, written, informed consent was obtained and throat cultures for respiratory viruses, chlamydia and \( \beta \)-lactamase producing bacteria were collected. A throat culture for Streptococcus pyogenes was repeated only if more than 24 hours had elapsed between the initial culture and the return visit. A urinalysis was performed, and blood for complete blood count, differential and serological studies, including anti-streptolysin O (ASO), anti-deoxyribonuclease B (anti-DNase B) titers and the Streptozyme test, was also obtained. Patients were assigned to the b.i.d. or q.i.d. groups according to a list of random numbers. Patients were started on oral suspension or capsule, depending on their weight. Patients weighing 20 kg or less received 500 mg oral suspension per day, while those over 20 kg received 1000 mg/day in capsule form in two or four equally divided doses for ten days. Each dose was numbered and packaged individually. For patients in the b.i.d. group, two of the four bottles or capsules in each daily treatment set contained placebo that was identical in appearance to those containing cephalaxin (prepared by the Eli Lilly Company). Written and verbal information was given to the patients and they were contacted by telephone daily during the first three days of therapy by the study nurse and questioned about the presence of fever, sore throat or other symptoms. Each patient returned to the clinic at the end of therapy and approximately 14 days later. On each visit, patients were examined for signs and symptoms of group A streptococcal pharyngitis. A throat culture for group A Streptococcus was repeated, and blood was obtained for serological studies.

Compliance was evaluated by the detection of antibiotic in the urine on Days 3 and 8 using a Sarcina lutea bioassay, and by medication counts at the return visit. For the urine antibiotic assay, parents were supplied with filter paper strips and pre-addressed stamped envelopes. They were instructed to wet the filter paper with the patient's urine to "study the body's response to infection". The strip was mailed to our laboratory where it was applied to a Mueller-Hinton agar plate seeded with a preconfluent growth of S. lutea. Any zone of inhibition was considered indicative of the presence of antibiotic.

Compliance was judged "good" if all the medicine was taken and both urine strips showed the presence of antibiotic; "poor" if there was no antibiotic detected in both urine samples and/or more than four doses were not taken during the ten-day period; "questionable" if the urine strips or medicine bottles were not returned, or if one of the urines showed absence of antibiotic. The clinical response was divided into cures and failures. Patients who were asymptomatic on both return visits were defined as clinical cures, while those with failure showed no improvement or had recurrence of signs and symptoms of pharyngitis within the follow-up period. Patients with satisfactory bacteriologic responses had negative throat cultures on both return visits, while those with bacteriologic failure had group A Streptococcus present in the pharynx on either reculture.

Laboratory procedures: Throat swabs were processed within 30 min by streaking on Columbia agar with 5% sheep blood and incubation for 18 h in 5% CO₂. Group A beta-hemolytic Streptococcus was identified by a fluorescent antibody technique (8) (BBL Microbiology Systems). Viral cultures were obtained by gargling with viral transport media and immediate inoculation of rhesus monkey kidney, human embryonic lung and Hep 2 cell lines (9). Swabs for chlamydia culture were placed in chlamydia transport media and inoculated onto McCoy cells (10).

\( \beta \)-lactamase producing aerobic and anaerobic bacteria were detected by the modification of the method described by Baldwin et al. (11). A throat swab obtained prior to and after completion of antibiotic therapy was placed in Schaedler's broth and processed immediately. The swab was rinsed in the broth and all excess fluid was removed by rotating it several times against the wall of the tube. From this suspension, serial ten-fold dilutions were made in Schaedler's broth and 0.1 ml was cultured on brain heart infusion agar supplemented with vitamin K and hemin. The plates were incubated in an anaerobic atmosphere for 48 h at 35°C, after which colonies were counted (total bacterial flora). Plates were then overlayed with 5 ml of melted starch agar containing 10,000 U of penicillin G and reincubated in ambient air at 35°C for 1 h. Finally, the cultures were flooded with Gram iodine solution and observed for the presence of clearing around colonies. The number of clear colonies representing \( \beta \)-lactamase producing organisms (\( \beta \)-lactamase producing flora) was compared to the total number of colonies observed after 48 h of incubation.

Serological titrations were performed as recommended by the manufacturers for ASO (Fisher Scientific Co.) and anti-DNase B titers (Wampole Lab). In order to increase the sensitivity of the Streptozyme test, we used more closely spaced dilutions as described by Kaplan and Huwe (12). A significant rise in antibody titers was defined as a two tube dilutional difference between the acute and convalescent sera. All isolates were tested in vitro by the Kirby-Bauer disk diffusion method for susceptibility to penicillin, erythromycin and cephalaxin (13). Group A Streptococcus isolates were considered susceptible to penicillin, erythromycin and cephalaxin if the zone of inhibition was \( \geq 22 \), \( \geq 18 \) and \( \geq 18 \) mm, respectively.

Results

Therapeutic Outcome

There were 81 patients, 3 to 20 years of age, enrolled in the study. Of these, 65 returned for both visits, seven for one, and nine did not return for follow-up. Sixty of the 65 (92%) children who returned for both of their follow-up visits had negative throat cultures for group A Streptococcus (Table 1). There were five bacteriologic failures, three in the q.i.d. and two in the b.i.d. treatment groups. All five patients had negative pharyngeal cultures for group A Streptococcus at the end of therapy, but positive cultures 14 days later. Only one was symptomatic; he developed sore throat and fever 16 days after discontinuation of cephalaxin. He was treated with penicillin but did not return for follow-up. The other four patients were observed without therapy and remained asymptomatic. Two of the five had no change in their streptococcal antibody.