The Journal of Laryngology and Otology
April 1993, Vol. **107**, pp. 309–312

Penetration of penicillin V to tonsillar surface fluid in healthy individuals and in patients with acute tonsillitis

A. STJERNQUIST-DESATNIK,* P. SAMUELSSON,* M. WALDER†

Abstract

In the treatment of group A streptococcal tonsillitis, as the bacteria are located on the epithelial surface, an important determinant of outcome is the concentration of penicillin in extracellular tonsillar surface fluid. Accordingly, we investigated the concentration of penicillin in serum, and penetration to tonsillar surface fluid and saliva in nine patients with acute group A streptococcal tonsillitis and in nine healthy controls.

Among the healthy subjects, despite high serum penicillin concentrations (mean, 2.04 μg/ml), there was no penetration to tonsillar surface fluid or to saliva, whereas erythromycin penetrated to tonsillar surface fluid in 3/6 cases.

Of the nine patients with acute tonsillitis, on the first day of treatment eight manifested high concentrations of penicillin in tonsillar surface fluid (mean, 0.34 μg/ml—i.e. well above the minimal inhibitory concentration (MIC) for group A streptococci), but penetration to saliva was found in only two patients. On the tenth day of treatment, penicillin was not present in the saliva of any of the patients and was present in the tonsillar surface fluid of only one. The results suggest that measurable concentrations of penicillin in tonsillar surface fluid can only be obtained in the presence of inflammation with fluid exudation through the tonsillar epithelium.

Key words: Tonsillitis; Penicillin, concentration; Streptococcus

Introduction

β-haemolytic group A streptococci are considered to be responsible for 30–50 per cent of all cases of acute tonsillitis (Wannamaker, 1972). Hitherto there have been no reports of penicillin-resistant group A streptococci, and the minimal inhibitory concentration (MIC) values for penicillin V are 0.005–0.02 μg/ml. Despite penicillin treatment, however, the bacterial and clinical treatment failure rates are as high as 10–25 per cent (Kaplan *et al.*, 1981; Roos *et al.*, 1985), and even higher if the course of treatment is reduced from 10 to 7 days (Schwarz *et al.*, 1981). Treatment failure in group A streptococcal tonsillitis has been ascribed to such factors as the inactivation of penicillin by β-lactamase-producing bacterial species present in the oral flora (Brook, 1985), the eradication by penicillin of α-haemolytic streptococci with inhibitory capacity against β-haemolytic streptococci (Roos *et al.*, 1986a), and the development of penicillin tolerance in group A streptococci (Kim and Kaplan, 1985) though the latter could not be confirmed in a recent study by us (Stjernquist-Desatnik *et al.*, 1991a). Re-infection from the patient's immediate surroundings might also explain the high frequencies of treatment failure (Stjernquist-Desatnik *et al.*, 1991a). Penicillin concentrations below the MIC value for group A streptococci at the site of infection is another possible explanation, though in several studies standard dosages of penicillin (i.e. 12.5 mg/kg body weight, *b.i.d.*) were found to yield penicillin concentrations in tonsillar tissue well above the MIC for group A streptococci (Holm and Ekedahl, 1982; Roos *et al.*, 1986b). However, as the bacteria are predominantly located in the extracellular tissue fluid, the penicillin concentration in this fluid would seem to be a more appropriate variable to study than whole or homogenized tissue concentrations. Strömberg *et al.* (1987), who measured the penicillin concentration in tonsillar surface fluid in patients tonsillectomized because of recurrent tonsillitis, found it to be higher than the concentration in tonsillar tissue and well above the MIC for group A streptococci.

Hitherto there have been no studies of antibiotic concentrations in tonsillar surface fluid in healthy individuals not suffering from recurrent tonsillitis, or in patients with acute tonsillitis.

The aim of the present study was to evaluate pencillin penetration to tonsillar surface fluid in healthy adults as well as in patients with acute group A streptococcal tonsillitis. Those studied were not given any anaesthesia. The penetration of erythromycin to tonsillar surface fluid was also studied in healthy adults.

Material and methods

Healthy persons

Ten adults (5 male and 5 female) aged 27–49 years were

From *Department of Oto-Rhino-laryngology, University Hospital, Lund, Sweden and †Department of Medical Microbiology, Malmö General Hospital, Malmö, Sweden.
Accepted for publication: 30 November 1992.

TABLE I
SERUM CONCENTRATIONS OF PENICILLIN V (μG/ML) IN HEALTHY SUBJECTS AFTER PERORAL INTAKE OF PENICILLIN V

Drug	Dose	Site	Time	*n*	Mean	SD	Range
pcV	1 g	Serum	1.5 h	9	2.04	1.39	0.9–4.2
			3 h	9	0.87	0.53	0.2–2.0
pcV	2 g	Serum	1.5 h	5	3.94	2.33	1.3–6.6
			3 h	5	2.56	1.17	1.0–3.7

included in the study. All 10 had normal serum creatinine concentrations, and none was suffering from recurrent acute tonsillitis or tonsillar hypertrophy or showed any signs of upper respiratory tract infection at the time of the study, or had received any antibiotic treatment during the preceding four-week period.

All ten adults were given oral penicillin V (T. Calciopen®), 1 g as a single dose. In a second trial, five of the adults received oral penicillin V (T. Calciopen®), 2 g as a single dose. Finally, in a third trial, six were given a single oral dose of 500 mg erythromycin (C. Erymax®). The interval between the trials was four weeks.

Patients

The patients included were nine 16 to 52-year-olds attending the ENT Department at University Hospital, Lund, because of sore throat. All nine had positive direct antigen test results for group A streptococci (Abbot testpack Strep A), manifested signs of acute tonsillitis at clinical examination and had normal serum creatinine concentrations. All patients were given oral penicillin V (T. Calciopen®), at a dosage of 12.5 mg/kg body weight *b.i.d.* for 10 days. None of the patients had received any antibiotic treatment during the preceding four-week period.

Sampling

At 1.5 and 3 hours after administration of the antibiotics, sampling was performed with the test subject awake and sitting in an upright position. No general or local anaesthetics were used. Three filter paper discs (AB Biodisk, Sweden) were placed on the surface of the tonsils to obtain tonsillar surface fluid and another three on the buccal mucosa to obtain saliva. The discs were left in place for one minute, during which time 10 μl fluid was usually absorbed. To avoid weight loss due to evaporation, the filter paper discs were immediately sealed in plastic tubes. A venous blood sample was taken simultaneously with the filter paper disc sampling. The procedure used was essentially that described by Strömberg *et al.* (1987), although in our study the test subjects were not anaesthetized. Ten filter paper discs were weighed before and after sampling. The weight gain of the discs were 10 μg ± 0.8 μg.

In the patients with acute streptococcal tonsillitis, sampling was performed as described above, 1.5 hours after administration of the first penicillin tablet on day 1, and 1.5 hours after administration of the penicillin tablet on day 10.

Antibiotic assays

The sampled sera and filter paper discs were kept frozen at −70°C until assayed (within 7 weeks). The concentrations were determined microbiologically with a routine agar-well diffusion technique. The agar used was DST (Oxid), pH 7.5, 125 ml agar in 24 × 24 cm disposable plastic plates. *Micrococcus luteus* ATCC 9341 was used as the indicator organism for the penicillin V assay, and *Micrococcus luteus* XCX for the erythromycin assay. Zones of inhibition were measured after overnight incubation at 36°C. Sterile, dry powder of penicillin (Astra, Sweden) and erythromycin (Astra, Sweden) of known potency were used to prepare standards. Serum standards were prepared by dissolving known concentrations of antibiotic in normal human serum. The serum assay limits were 8.0–0.01 μg/ml for penicillin V and 6.0–0.03 μg/ml for erythromycin. The discs were placed on the agar surface. Disc standards were prepared by impregnating filter paper discs (Biodisk, Sweden) with 10 μl of phosphate-buffered saline containing different concentrations of antibiotic. The assay limits were 5.0–0.03 μg/ml for penicillin V and 10.0–0.4 μg/ml for erythromycin. All sera, discs and standards were assayed in duplicate.

Results

Penicillin concentrations in healthy controls (Table I)

In the group given penicillin V, 1 g, one person's data was lost. In the remaining nine, the mean serum penicillin concentrations was 2.04 μg/ml (SD 1.39) at 1.5 hours after administration and 0.87 μg/ml (SD 0.53) at three hours. There were no measurable penicillin concentrations in any of the tonsillar surface fluid or saliva samples.

In the five subjects given 2 g penicillin V, the mean serum concentration was 3.94 μg/ml (SD 2.33) after 1.5 hours and 2.56 μg/ml (SD 1.17) at 3 hours. No penicillin could be detected in the tonsillar surface fluid or in the saliva.

To rule out the possibility that freezing of the samples or bad absorption might affect the results, two persons were given 1 g penicillin V and the filter paper discs were left for several minutes on the tonsils and buccal mucosa to get 'extra wet'. The samples were analysed immediately without freezing, but no measurable concentrations of penicillin could be detected in the tonsillar surface fluid or in the saliva, despite the serum concentrations being comparable with those in the first group (see above).

Erythromycin concentrations in healthy persons (Table II)

In the six healthy subjects given erythromycin (500 mg), the mean serum erythromycin concentration

TABLE II
CONCENTRATIONS OF ERYTHROMYCIN (μG/ML) IN SERUM, TONSILLAR SURFACE FLUID AND SALIVA IN HEALTHY SUBJECTS AFTER PERORAL INTAKE OF 500 MG ERYTHROMYCIN

Site	Time	*n*	Mean	SD	Range
Serum	1.5 h	6	1.12	0.65	0.05–2.0
	3 h	6	0.82	0.55	0 –1.4
Tonsillar Surface fluid	1.5 h	6	One sample 0.6		
	3 h	6	Three samples 0.5; 0.6; 0.9		
Saliva	1.5 h	6	Two samples 0.5; 0.8		
	3 h	6	Three samples 0.4; 0.5; 0.5		

https://doi.org/10.1017/S0022215100122893 Published online by Cambridge University Press

TABLE III

CONCENTRATIONS OF PENICILLIN V (μG/ML) IN SERUM AND TONSILLAR SURFACE FLUID IN PATIENTS WITH ACUTE TONSILLITIS 1.5 HOURS AFTER PERORAL INTAKE OF 1 G PENICILLIN V ON THE FIRST DAY OF TREATMENT

Site	*n*	Mean	SD	Range
Serum	9*	0.93	0.42	0.38–1.7
Tonsillar Surface fluid	9	0.34	0.52	0 –1.7

*Serum from two patients missing.

was 1.12 μg/ml (SD 0.65) at 1.5 hours after administration, and 0.82 μg/ml (SD 0.55) at 3 hours. At 1.5 hours after administration, erythromycin could be detected in saliva and tonsillar surface fluid in one person, and at 3 hours in both saliva and tonsillar surface fluid in three persons (including the subject with measurable concentrations, at 1.5 hours). In one subject erythromycin was estimated in saliva at 1.5 hours but not in tonsillar surface fluid, and no erythromycin could be detected in this person's disc samples at 3 hours after administration. Thus, erythromycin penetrated both to tonsillar surface fluid and to saliva in 3/6 persons.

In the two subjects who did not manifest measurable erythromycin concentrations in saliva or in tonsillar surface fluid, the serum concentration was much lower than in the other four, mean 0.425 μg/ml at 1.5 hours after administration and 0.2 μg/ml at 3 hours.

Penicillin concentrations in patients with acute tonsillitis (Table III)

Among the patients, the mean serum penicillin concentration was 0.93 μg/ml (SD 0.42) on day one and 1.58 μg/ml (SD 0.76) on day 10. The serum samples of two of the patients were lost. In 8/9 patients, penicillin could be detected in tonsillar surface fluid on day one, at a mean concentration of 0.34 μg/ml (SD 0.52). The patient with no measurable penicillin concentration in tonsillar surface fluid had a serum concentration of 1.05 μg/ml. Penicillin in saliva was detected in two patients only, 0.14 μg/ml in one case and 0.21 μg/ml in the other. On day 10, penicillin was detected in tonsillar surface fluid in one person only, 0.1 μg/ml; this patient had had the highest concentration in tonsillar surface fluid on day 1 (1.7 μg/ml). In none of the patients could penicillin be detected in saliva on day 10.

Discussion

Despite high serum concentrations, penicillin could not be detected in tonsillar surface fluid or saliva from healthy individuals in the present study. Strömberg *et al.* (1987) reported penicillin concentrations in tonsillar surface fluid to be above the MIC value for group A streptococci, and higher than concentrations in tonsillar tissue. However, the patients in Strömberg's study were suffering from recurrent tonsillitis, and thus manifested chronic inflammation of the tonsils with increased plasma leakage and exudation of fluid through the epithelium. The tonsils in such patients are probably characterized by constant antigen stimulation, as in an earlier study of ours we found the isolation rate of such aerobic bacteria as *H. influenzae* and group A streptococci in tonsillar core tissue from patients with recurrent tonsillitis to be higher than that in normal tonsils from patients tonsillectomized because of sleep apnoea (Stjernquist-Desatnik *et al.*, 1991b). The sampling in the Strömberg study was performed with the subject under general anaesthesia, which might have affected tonsillar blood flow, while our test subjects were not anaesthetized.

In the patients with acute group A streptococcal tonsillitis, penicillin could be detected in tonsillar surface fluid on day 1 of the treatment, the mean concentration being well above the MIC for group A streptococci. On the tenth day of treatment, penicillin could be detected in tonsillar surface fluid in only one person. This suggests that the concentration of penicillin decreases as the inflammation abates, an interpretation consistent with the finding by Ingvarsson *et al.* (1980) in patients with acute otitis media that the penicillin concentration in middle ear exudate decreased as the inflammation in the middle ear abated.

Prophylactic penicillin treatment of children with acute otitis media has been studied by Persico *et al.* (1985). Our findings provide no support for the efficacy of prophylactic penicillin treatment of patients with recurrent group A streptococcal tonsillitis.

Most pharmacokinetic studies on antibiotics are performed in healthy individuals, both serum and tissue being analysed for antibiotic content. However, microscopic studies of tonsils from patients with recurrent tonsillitis have shown the bacteria to be located in the crypts and on the surface of the tonsils, but not to penetrate the surface epithelium (Engquist and Lundberg, 1987); and as it is well known that most bacterial infections are located in the extracellular fluid, there would seem to be little point in studying antibiotic concentrations in the tissues. Since exudation of fluid occurs as an inflammatory response to infection, the antibiotic concentration at the site of acute infection would seem to be the most logical variable to study.

In the present study, as penicillin could be detected in saliva in only two of the nine patients with acute group A streptococcal tonsillitis, the antibiotic concentration detected in tonsillar surface fluid cannot have originated from saliva reaching the tonsillar surface. Erythromycin could be detected in tonsillar surface fluid in 3/6 healthy persons. Sundberg *et al.* (1981) found the erythromycin concentration to be the same in tonsillar tissue as in plasma in patients tonsillectomized because of recurrent tonsillitis. Regarding penicillin, the concentration in tonsillar tissue has been found to be only 20 per cent of that in serum (Roos *et al.*, 1986b). Thus, in healthy persons, the penetration of erythromycin to tonsillar tissue and surface fluid seems to be better than that of penicillin. However, as an increasing resistance to erythromycin has been found among group A streptococci both in Finland (Nissinen *et al.*, 1991) and in the south of Sweden (Schalén, C., personal communication), erythromycin would not seem to be suitable for treating group A streptococcal infections.

To sum up, this is the first study of penicillin penetration to tonsillar surface fluid in patients with acute tonsillitis. No penicillin could be detected in tonsillar surface fluid in healthy individuals, while in patients with acute tonsillitis the concentrations were well above the MIC for group A streptococci on the first day of treatment. On the tenth day, penicillin could be detected in tonsillar surface fluid in only 1/ 9 patients. The findings suggest that detectable

concentrations of penicillin in tonsillar surface fluid (where the bacteria are predominantly located) can only be obtained in the presence of inflammation with exudation of fluid through the tonsillar epithelium.

Acknowledgements

This work has been supported by grants from the Medical Faculty of the University of Lund, the Royal Physiographical Society at Lund, and the Swedish Medical Society.

References

Brook, I. (1985) Role of beta-lactamase producing bacteria in the failure of penicillin to eradicate group A streptococci. *Pediatric Infectious Disease Journal* **4:** 491–495.

Engquist, S., Lundberg, C. (1987) Bakteriers topografiska relation till slemhinnestrukturer vid slemhinneinfektioner (in Swedish). *Svensk Otolaryngologisk Förenings Förhandlingar* **2**.

Holm, S. E., Ekedahl, C. (1982) Comparative study of the penetration of penicillin V and cefadroxil into tonsils in man. *Journal of Antimicrobial Chemotherapy* **10:** *(Suppl. B)*, 137–142.

Ingvarsson, L., Kamme, C., Lundgren, K. (1980) Concentration of penicillin V in serum and middle ear exudate during treatment of acute otitis media. *Annals of Otology, Rhinology and Laryngology* **89:** *(Suppl. 68)*, 59–61.

Kaplan, E. L., Gastandjy, A., Huwe, B. (1981) The role of the carrier in treatment failures after antibiotic therapy for group A streptococci in the upper respiratory tract. *Journal of Laboratory and Clinical Medicine* **98:** 326–335.

Kim, K. S., Kaplan, E. L. (1985) Association of penicillin tolerance with failure to eradicate group A streptococci from patients with pharyngitis. *Journal of Pediatrics* **107:** 681–684.

Nissinen, A., Kontinen, S., Huovinen, P., Järvinen, H., Oinonen, S., Liimatainen, O., Katila, M.-L., Herva, E. (1991) Erythromycin resistance in group A streptococcus in Finland 1990. *II Scandinavian Meeting of Bacteriology, Turku, Finland*, **Abstract** p. 14.

Persico, M., Podoshin, L., Fradis, M., Grushka, M., Golan, D., Foltin, V., Wellisch, G., Cahana, Z., Kolin, A., Winter, S. (1985) Recurrent acute otitis media—prophylactic penicillin treatment: a prospective study. Part I. *International Journal of Pediatric Otorhinolaryngology* **10:** 37–46.

Roos, K., Holm, S. E., Ekedahl, C. (1985) Treatment failure in acute streptococcal tonsillitis in children over the age of 10 and in adults. *Scandinavian Journal of Infectious Diseases* **17:** 357–365.

Roos, K., Grahn, E., Holm, S. E. (1986a) Evaluation of beta-lactamase activity and microbial interference in treatment failures of acute streptococcal tonsillitis. *Scandinavian Journal of Infectious Diseases* **18:** 313–319.

Roos, K., Grahn, E., Ekedahl, C., Holm, S. E. (1986b) Pharmacokinetics of phenoxymethylpenicillin in tonsils. *Scandinavian Journal of Infectious Diseases* **18:** 125–130.

Schwarz, R. H., Wientzen Jr., R. L., Pedreira, F., Feroli, E. J., Mella, G. W., Guandolo, V. L. (1981) Penicillin V for group A streptococcal pharyngotonsillitis. A randomized trial of seven vs 10 days' therapy. *JAMA* **246:** 1790–1795.

Stjernquist-Desatnik, A., Orrling, A., Schalén, C., Kamme, C. (1991a) Penicillin tolerance in group A streptococci and treatment failure in streptococcal tonsillitis. *Acta Oto-Laryngologica (Stockholm), (Suppl)* **492:** 68–71.

Stjernquist-Desatnik, A., Prellner, K., Schalén, C. (1991b) High recovery of *Haemophilus influenzae* and group A streptococci in recurrent tonsillar infection or hypertrophy as compared with normal tonsils. *Journal of Laryngology and Otology* **105:** 439–441.

Strömberg, A., Friberg, U., Cars, O. (1987) Concentrations of phenoxymethylpenicillin and cefadroxil in tonsillar tissue and tonsillar surface fluid. *European Journal of Clinical Microbiology* **6:** 525–529.

Sundberg, L., Edén, T., Ernstson, S. (1981) Penetration of erythromycin in Waldeyer's ring—tonsil tissue. *Acta Oto-Laryngologica (Stockholm), (Suppl)* **384:** 10–17.

Wannamaker, L. W. (1972) Perplexity and precision in the diagnosis of streptococcal pharyngitis. *American Journal of Diseases of Children* **124:** 352–358.

Address for correspondence:
Anna Stjernquist-Desatnik, M.D., Ph.D.,
Department of Oto-Rhino-Laryngology,
University Hospital,
S-221 85 Lund,
Sweden.