Although the mechanism of the effect of azathioprin is not known, the clinical effect has been so striking that a report is warranted.

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Cefuroxime Treatment of Urethritis Caused by a β-lactamase-producing Strain of *Neisseria Gonorrhoeae*

Staffan Normark,[1] Theodor Elmros,[3] Lena Norlander,[1] Erik Kjellberg,[4] Stig Holm,[3] Lars Å. Burman[2] and Sven Bergström[1]

Departments of [1]Microbiology, [2]Infectious Diseases, [3]Dermatology, and [4]Bacteriology, University of Umeå, Umeå, Sweden

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Abstract. A patient who contracted urethritis from a β-lactamase-producing strain of *Neisseria gonorrhoeae* was successfully treated with the cephalosporin derivative cefuroxime. As expected, neither cefuroxime nor cefamandole was hydrolysed by plasmid-coded gonococcal β-lactamase. Cefuroxime ought to be a valuable and efficacious substitute for penicillins in the treatment of gonhorrhoea due to β-lactamase-producing gonococcal strains.

Key words: Neisseria gonorrhoeae; β-Lactamase; Plasmids; Cefuroxime

Ever since its discovery, penicillin has been of the utmost importance in the treatment of gonorrhoea. Until 1976, the resistance of *Neisseria gonorrhoeae* to penicillins has rarely created serious therapeutical problems. However, β-lactamase-producing gonococcal strains were isolated for the first time in 1976 (9, 10). The β-lactamase was found to be a TEM-1 enzyme and is thus identical with the β-lactamase found in *Haemophilus influenzae* and many Gram-negative enterobacteria. It has been established that the β-lactamase of *Neisseria gonorrhoeae* is plasmid mediated (3). Two plasmids have been found, 3.3 Mdal and 4.4 Mdal in size, respectively.

It is likely that the 3.3 Mdal is derived from the 4.4 Mdal plasmid. The latter plasmid has probably been transmitted from a β-lactamase producing strain of *Haemophilus para-influenzae* (Sparling, unpublished information). *Neisseria gonorrhoeae* strains carrying the 4.4 Mdal resistance plasmid can transfer the plasmid to sensitive strains by conjugation (cell to cell contact) due to the presence of a large conjugative 24.5 Mdal plasmid (2, 12).

The fact that all β-lactamase-producing gonococci so far examined contain either of the above-mentioned resistant plasmids, makes it possible to predict the rate of β-lactam hydrolysis for a large number of different clinically relevant β-lactams (penicillins, cephalosporins and cefamycins). Thus the TEM-1 enzyme, a typical ampicillinase, is not very effective against most cephalosporins including the new β-lactamase-resistant derivatives, cefuroxime and cefamandole. It was therefore of interest to clinically evaluate one of these drugs, cefuroxime, in the treatment of a patient with urethritis caused by a β-lactamase-producing strain of *Neisseria gonorrhoeae*.

MATERIALS AND METHODS

Strain 1773 was a gonococcal isolate from the urethal discharge of the patient, whose case is described in the text. The β-lactamase-producing strain CDC67 was obtained from Stanley Falkow and is known to carry the 2.6, 4.4 and 24.5 Mdal plasmids. The solid medium used was GC medium base supplemented with Iso-Vitalex (BBC). Plates were incubated in 6% CO_2 in a CO_2 incubator. The complete liquid medium was identical with the solid medium except that agar was omitted. β-lactamase activity when using penicillins as substrates was assayed by the automatic iodometric method of Lindström & Nordström, (1). When cephalosporins were used as substrates the spectrophotometric method of Fu & Neu (5) was used. Specific activity in nanokatal is defined as

Table I. *Resistance pattern of Neisseria gonorrhoeae strain 1793*

Resistance was determined on GC medium plates using a multipoint inoculation technique.

Antibiotic	Minimum inhibitory concentration (μg/ml)	
	Small inoculum 4×10^4 CFU/ml	Large inoculum 4×10^8 CFU/ml
Cefuroxime	0.25	2.0
Cefamandole	0.15	1.5
Ampicillin	4.0	32.0
Penicillin G	2.0	32.0
Tetracycline	0.5	2.0
Spectinomycin	8.0	16.0
Erythromycin	0.25	2.0

the enzyme activity that hydrolysed 1 nanomole of β-lactam per second per milligram of protein. Protein was determined by the method of Lowry et al. (7). Resistance to antimicrobial agents was tested using a multipoint inoculation technique. Covalently closed circular DNA were prepared from clear lysates and subjected to agarose gel electrophoresis, as described by Bergström et al. (1).

RESULTS

A 32-year-old male contracted urethritis during a holiday in Thailand. After arrival at his home town in Sweden he consulted a general practitioner who, after microscopic investigation and bacteriological cultivation, made the diagnosis gonorrhoea. The patient was initially treated with ampicillin 0.5 g× 2 for 10 days. The symptoms were reduced in intensity, but returned after the end of the treatment. The patient was thereafter treated with lymecyclin (0.3 g×2) for 10 days with the same negative results. He was therefore referred for specialist treatment. Treatment with cefuroxime (1.5 g×3) intravenously was initiated and continued for 6 days. After 4 days the patient was free from symptoms. Three examinations were made after the end of the cure. The last control was performed 2 months after the termination of the treatment. Neither symptoms nor bacteriological evidence of gonococcal infection were found.

The minimum inhibitory concentration of the isolated gonococcal strain against 7 different antibiotics was sought (Table I). The gonococcal specimen was found to be resistant to ampicillin and benzylpenicillin when a large inoculum 4×10^8 CFU/ml) was used, but only moderately so when using a small inoculum (4×10^4 CFU/ml). This is a typical finding for β-lactamase-producing gonococci. No such large inoculum dependence was found for cefuroxime, cefamandole and the non-β-lactam compounds.

The presence of β-lactamase activity in the causative specimen of *N. gonorrhoeae* was demonstrated by chromogenic cephalosporin (8). Crude extracts of the patient strain 1793 were also analysed with respect to β-lactamase activity against benzylpenicillin, ampicillin, cephalotin, cefuroxime and cefamandole (Table II). The activity in a crude extract of the well characterized β-lactamase-producing strain CDC67 was included in the table. As expected, the latter two drugs were not hydrolysed by gonococcal β-lactamase. Cephalotin was also markedly resistant to hydrolysis.

Covalently closed circular DNA from strains 1793 and CDC67 were purified by cesium chloride ethidium bromide buoyant density gradient centrifugation. Both strains were found to contain the 4.4 Mdal resistance plasmid, the 24.5 Mdal conjugative plasmid as well as the small phenotypically cryptic 2.6 Mdal plasmid (Fig. 1).

DISCUSSION

Until 1971 ampicillin was the drug of choice in the treatment of infections caused by *Haemophilus in-*

Table II. *β-lactamase activity using five β-lactam compounds as substrates*

Strain	Specific β-lactamase[a]				
	Benzyl penicillin	Ampicillin	Cefalothin	Cefuroxime	Cefamandole
1793[b]	3.7	7.8	0.01	<0.01	<0.01
CDC67	4.4	7.4	0.01	<0.01	<0.01

[a] Expressed in nanokatals.
[b] The strain isolated from the patient.

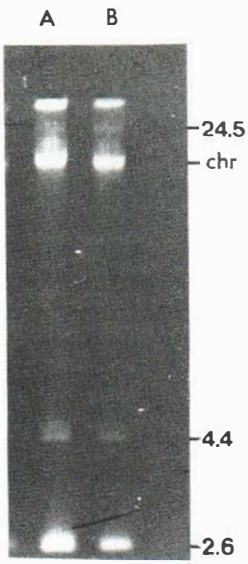

Fig. 1. Plasmids carried by strain 1793. Strain CDC67 (B), of known plasmid content, is used as reference. The figures represent the molecular weight in Mdal (10^6 daltons) of the plasmids. Chr marks the position for chromosomal material.

fluenzae. In 1972 β-lactamase-producing strains of *H. influenzae* were detected. This increase in antibiotic resistance was thereafter found to create obvious therapeutical problems. Therefore, in 1976 when a similar development was found in *N. gonorrhoeae* a discussion about alternative antibiotic treatment was started.

Spectinomycin and tetracycline have been suggested for use in the treatment of gonorrhoea caused by β-lactamase-producing gonococci. The gonococcal strain isolated from our patient showed a low resistance towards tetracycline in vitro. Despite this the patient was not cured even after 10 days of treatment with tetracycline.

In the past, relatively little interest has been paid to cephalosporins in the treatment of gonorrhoea. This is in part due to the relatively high in vitro tolerance of *N. gonorrhoeae* to these drugs as compared with penicillins. However, during recent years new cephalosporins have been developed, some of which may be given in a peroral form and some of which are resistant to most β-lactamases (6, 10, 11). The availability of these new cephalosporins emphasizes the need to re-evaluate the usefulness of cephalosporins in the treatment of gonorrhoea and especially of β-lactamase-producing derivatives. This has also recently been suggested by others (4, 11). The clinical case presented demonstrates this point and in particular indicates the usefulness of a β-lactamase-resistant cephalosporin (cefuroxime).

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