ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Aug. 1981, p. 265-266
0066-4804/81/080265-02$02.00/0

Vol. 20, No. 2

Comparative In Vitro Activity of Mk-0366 and Other Selected Oral Antimicrobial Agents Against *Neisseria gonorrhoeae*

MOHAMMED Y. KHAN,* YOUSUF SIDDIQUI, AND ROBERT P. GRUNINGER

Section of Infectious Disease, Department of Medicine, Hennepin County Medical Center, and University of Minnesota Medical School, Minneapolis, Minnesota 55415

Received 14 May 1981/Accepted 27 May 1981

The in vitro activity of a new oral antimicrobial agent, Mk-0366 (AM-715), was compared with those of rosoxacin, ampicillin, erythromycin, and tetracycline against *Neisseria gonorrhoeae*. Mk-0366 was as active as rosoxacin and more active than the other three antimicrobial agents. It inhibited all isolates, regardless of β-lactamase activity, at a concentration of 0.03 μg/ml.

Mk-0366 (AM-715) is an antibacterial organic acid structurally related to nalidixic acid. Unlike nalidixic acid, Mk-0366 exhibits a very broad spectrum of antibacterial activity against both gram-negative and gram-positive bacteria (5). In experimental systemic and urinary bladder-kidney infections in mice, Mk-0366 was found to be five times as active as pipemidic acid and nalidixic acid (4). This study was conducted to evaluate the in vitro activity of Mk-0366 against β-lactamase-negative and β-lactamase-positive *Neisseria gonorrhoeae* strains. As Mk-0366 is an oral antimicrobial agent, we therefore compared it with rosoxacin, a related investigational agent active against *N. gonorrhoeae*, and with ampicillin, erythromycin, and tetracycline, currently used orally in gonococcal infections.

Mk-0366 was obtained from Merck Institute for Therapeutic Research, Rahway, N.J.; rosoxacin was provided by Sterling-Winthrop Research Institute, Rensselaer, N.Y.; erythromycin was obtained from Abbott Laboratories, North Chicago, Ill.; and tetracycline was obtained from Pfizer Inc., New York, N.Y.

A total of 76 strains of *N. gonorrhoeae* were tested. Fifty-six β-lactamase-negative strains were collected during 1980 from patients with anogenital infection at the Hennepin County Medical Center, Minneapolis, Minn. Sixteen β-lactamase-positive strains were obtained from the following sources: W. Hall, Veterans Administration Medical Center, Minneapolis, Minn.; Minnesota Department of Health, Minneapolis; and Centers for Disease Control, Atlanta, Ga. The identity of the isolates was confirmed by growth on Thayer-Martin agar, Gram stain, a positive oxidase reaction, and acidification of glucose, but not maltose, lactose, or sucrose. The organisms were frozen in tryptic soy broth (Difco Laboratories, Detroit, Mich.) containing 20% glycerol and stored at −80°C. *N. gonorrhoeae* isolates were tested for β-lactamase activity by an acidometric method with phenol red indicator (7).

The minimum inhibitory concentrations (MICs) of Mk-0366, rosoxacin, ampicillin, erythromycin, and tetracycline were determined by an agar dilution technique (6). Twofold dilutions of the antimicrobial agents, from 8 to 0.015 μg/ml, were distributed into Mueller-Hinton agar supplemented with 2% hemoglobin and 1% IsoVitaleX. The frozen gonococcal isolates were thawed, grown overnight on chocolate agar, and then suspended in tryptic soy broth until the turbidity matched that of a 0.5 McFarland standard. One microliter of a 1:10 dilution of the adjusted suspension (10^4 colony-forming units) was inoculated onto the antimicrobial agent-containing plates with a Steers replicator. The plates were incubated for 18 to 24 h at 35°C in a CO_2 atmosphere. The MIC was the lowest concentration of antimicrobial agent that inhibited visible growth.

The MICs for five antimicrobial agents against the β-lactamase-negative *N. gonorrhoeae* strains are given in Table 1. Mk-0366 inhibited 90% of the isolates in this group at a concentration of ≤0.015 μg/ml. All isolates were inhibited at a concentration of 0.03 μg/ml.

Rosoxacin was essentially equal to Mk-0366 in its activity against this group. It inhibited 90% of the isolates at a concentration of 0.03 μg/ml and 100% of the isolates at a concentration of 0.06 μg/ml. The currently available antimicrobial agents ampicillin, erythromycin, and tetracycline were less active than Mk-0366 and rosoxacin against the β-lactamase-negative *N. gonorrhoeae* strains.

Table 2 shows the MICs of the five antimicrobial agents against 16 β-lactamase-positive *N. gonorrhoeae* strains. Mk-0366 was effective against this group. All strains were inhibited at

TABLE 1. *Antimicrobial susceptibility of 56 β-lactamase-negative N. gonorrhoeae strains*

Antimicrobial agent	MIC (μg/ml)			
	Mean	For % strains		
		50	90	100
Mk-0366	0.015	≤0.015	≤0.015	0.03
Rosoxacin	0.018	≤0.015	0.03	0.06
Ampicillin	0.070	0.06	0.125	0.25
Erythromycin	0.425	0.25	1.0	2.0
Tetracycline	0.415	0.25	1.0	1.0

TABLE 2. *Antimicrobial susceptibility of 16 β-lactamase-positive N. gonorrhoeae strains*

Antimicrobial agent	MIC (μg/ml)			
	Mean	For % strains		
		50	90	100
Mk-0366	0.017	≤0.015	0.03	0.03
Rosoxacin	0.019	≤0.015	0.03	0.03
Ampicillin	7.312	≥8.0	≥8.0	≥8.0
Erythromycin	0.945	1.0	2.0	2.0
Tetracycline	2.250	2.0	4.0	4.0

a concentration of 0.03 μg/ml; rosoxacin was equally effective, with MICs similar to those of Mk-0366 against these strains. As expected, all isolates were highly resistant to ampicillin. Against erythromycin and tetracycline, all strains were susceptible at concentrations of 2.0 and 4.0 μg/ml, respectively.

Several new cephalosporins and cephamycin antimicrobial agents have excellent in vitro activity against *N. gonorrhoeae*, including β-lactamase-positive strains (1, 8). Recent clinical studies concerning the use of these new antimicrobial agents in gonococcal infections have shown encouraging results (2, 3). However, these antimicrobial agents are available only in parenteral form and are not preferred by some patients with gonorrhea. It is desirable to have alternate oral therapy available for infections due to *N. gonorrhoeae*, including β-lactamase-positive strains.

Rosoxacin is an oral antimicrobial agent related to nalidixic acid. It is active against *N. gonorrhoeae*, including β-lactamase-positive strains, and is currently undergoing clinical trials in gonococcal infections. Preliminary results have shown that it is effective in uncomplicated gonococcal infections, but side effects have prompted further trials with lower doses (B. Lutz, B. Pauling, and W. J. Mogabgab, Program Abstr. Intersci. Conf. Antimicrob. Agents Chemother. 20th, New Orleans, La., abstr. no. 314, 1980; H. H. Handsfield, F. N. Judson, and K. K. Holmes, 20th ICAAC, abstr. no. 316).

Mk-0366 is another new orally active antimicrobial agent which is also related to nalidixic acid. Our results indicate that Mk-0366 is as active as rosoxacin against both β-lactamase-positive and β-lactamase-negative strains of *N. gonorrhoeae*. It is more active in vitro against *N. gonorrhoeae* than are the currently available oral antimicrobial agents ampicillin, erythromycin, and tetracycline. After a single oral dose of 200 mg, Mk-0366 gave a peak serum level of 1.11 μg/ml, with a half-life of 2.43 h (J. Shimada, Y. Ueda, and T. Yamaji, 20th ICAAC, abstr. no. 75). This level is 30-fold greater than the MIC of Mk-0366 required to inhibit all *N. gonorrhoeae* strains tested in our study. A preliminary report from Japan has shown that it is effective in gonococcal urethritis. Of the 14 patients treated with Mk-0366, 93% were cured (Y. Nishimura, H. Kishi, O. Tsukada, T. Tominaga, and T. Niijima, 20th ICAAC, abstr. no. 76).

On the basis of our in vitro data and the reported successful treatment of a few gonorrhea cases, further clinical studies are needed to define the efficacy of Mk-0366 as an oral antimicrobial agent for gonococcal infections.

We thank Sue Counter for typing this manuscript.

LITERATURE CITED

1. **Baker, C. N., C. Thornsberry, and R. N. Jones.** 1980. In vitro antimicrobial activity of cefoperazone, cefotaxime, moxalactam, azlocillin, mezlocillin, and other β-lactam antibiotics against *Neisseria gonorrhoeae* and *Haemophilus influenzae*, including β-lactamase-producing strains. Antimicrob. Agents Chemother. **17**:757-761.
2. **Berg, W. S., M. E. Kilpatrick, W. O. Harrison, and J. A. McCutchan.** 1979. Cefoxitin as a single-dose treatment for urethritis caused by penicillinase-producing Neisseria gonorrhoeae. N. Engl. J. Med. **301**:509-511.
3. **Fowler, W., G. Rahim, and J. D. Brow.** 1978. Clinical experience in the use of cefuroxime in gonorrhoeae. Br. J. Vener. Dis. **54**:400-402.
4. **Hirai, K., A. Ito, Y. Abe, S. Suzue, T. Irikura, M. Inoue, and S. Mitsuhashi.** 1981. Comparative activities of AM-715 and pipemidic and nalidixic acids against experimentally induced systemic and urinary tract infections. Antimicrob. Agents Chemother. **19**:188-189.
5. **Ito, A., K. Hirai, M. Inoue, H. Koga, S. Suzue, T. Irikura, and S. Mitsuhashi.** 1980. In vitro antibacterial activity of AM-715, a new nalidixic acid analog. Antimicrob. Agents Chemother. **17**:103-108.
6. **Jaffee, H. W., J. W. Biddle, C. Thornsberry, R. E. Johnson, R. E. Kaufman, G. H. Reynolds, P. J. Wiesner, and the Cooperative Study Group.** 1976. National gonorrhea therapy monitoring study: in vitro antibiotic susceptibility and its correlation with treatment results. N. Engl. J. Med. **294**:5-9.
7. **Philips, C. W., R. D. Allen, and S. N. Cohen.** 1976. Penicillinase-producing Neisseria gonorrhoeae. Lancet **ii**:960.
8. **Philips, I.** 1978. The susceptibility of Neisseria gonorrhoeae to cefoxitin sodium. J. Antimicrob. Chemother. **4**:61-64.