ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Jan. 1998, p. 206–207 Vol. 42, No. 1
0066-4804/98/$04.00+0

Letter to the Editor

Emergence of Fluoroquinolone Resistance in Strains of *Vibrio cholerae* Isolated from Hospitalized Patients with Acute Diarrhea in Calcutta, India

The problem of antimicrobial resistance in microorganisms causing diarrheal diseases in both developed and developing countries continues to be alarming (6). Toxigenic *Vibrio cholerae* is the causative agent of the life-threatening disease cholera. Plasmid-encoded high-level resistance to ampicillin, kanamycin, streptomycin, sulfonamide, tetracycline, trimethoprim, and gentamicin has been demonstrated among strains of *V. cholerae* O1 isolated from Bangladesh, India, and Latin America (7, 9). Fluoroquinolones have excellent activity against all pathogenic *Vibrio* species, and clinical trials have found norfloxacin to be effective for the treatment of cholera in adults and in children (2, 3).

Antimicrobial susceptibility analysis of 212 strains of *V. cholerae* isolated from patients with acute watery diarrhea admitted to the Infectious Diseases Hospital, Calcutta, India, between March 1996 and September 1996 was performed by the disk diffusion technique (1) with commercial discs (Hi-Media, Bombay, India). Characterization of strains as susceptible, intermediately resistant, or resistant was based on the size of the inhibition zones around each disc according to the manufacturer's instructions, which matched the interpretive criteria recommended by World Health Organization (8). Strains showing intermediate zones of inhibition were interpreted as resistant on the basis of previous MIC studies conducted with *V. cholerae* (9). MICs of norfloxacin and ciprofloxacin were determined by the agar dilution method with Mueller-Hinton agar according to standard procedures (8). *V. cholerae* strains resistant to norfloxacin and cirprofloxacin began appearing in late 1995 (5), and strains resistant to fluoroquinolones started increasing from July 1996, with a steady decrease in the zone of inhibition. To date, resistance to norfloxacin and ciprofloxacin is restricted to the non-O1 non-O139 serogroups of *V. cholerae* (Table 1). This prompted us to examine the activity of three more fluoroquinolones against *V. cholerae* non-O1 non-O139 strains (Table 1). Of the 69 strains examined, 14.5, 10.1, and 11.6% showed reistance to lomefloxacin, pefloxacin, and ofloxacin, respectively. Among the O1 and O139 serogroups isolated in 1996, a contrasting pattern of resistance to different antibiotics was observed, with the most common (71%) drug resistance in *V. cholerae* O1 strains being to ampicillin, chloramphenicol, co-trimoxazole, furazolidone, neomycin, nalidixic acid, and streptomycin, while the most common (66%) resistance pattern in *V. cholerae* O139 was ampicillin, furazolidone, neomycin, and streptomycin.

TABLE 1. Percentage distribution of drug-resistant *V. cholerae* strains isolated from hospitalized patients in 1996[a]

Drug	Percent resistance in *V. cholerae*:		
	O1 (n = 79)	O139 (n = 64)	Non-O1, non-O139 (n = 69)
Ampicillin	100	100	100
Chloramphenicol	75.9	9.4	21.7
Ciprofloxacin	0	0	31.9
Co-trimoxazole	100	0	44.9
Furazolidone	100	100	100
Gentamicin	7.6	1.6	18.8
Neomycin	93.7	100	100
Nalidixic acid	100	4.7	36.2
Norfloxacin	0	0	18.8
Streptomycin	100	93.8	63.8
Tetracycline	0	6.3	21.7

[a] Antibiotic susceptibility was performed by the dry disk diffusion technique (1) with commercial discs, and strains were characterized as resistant as described in the text.

In this study, the agar dilution breakpoints adopted by the Centers for Disease Control and Prevention, Atlanta, Ga. (MICs of $\geq$1.0 μg of ciprofloxacin and norfloxacin per ml), were used for interpretation of resistance to these agents (4). Among the fluoroquinolones, MICs of the widely used norfloxacin and ciprofloxacin for the resistant strains of *V. cholerae* non-O1 non-O139 ranged from 3.125 to >100 and 3.125 to 75 μg/ml, respectively; MICs of norfloxacin were >100 μg/ml for 5 strains, and the MIC of ciprofloxacin was about 50 μg/ml. MICs for the susceptible strains of *V. cholerae* non-O1 non-O139 ranged from $\leq$0.0245 to 0.39 μg/ml for ciprofloxacin and 0.049 to 0.78 μg/ml for norfloxacin. A trend of increasing MICs for susceptible strains isolated after June 1996 was observed. All strains, with the exception of one in each case, were concurrently resistant to both norfloxacin and ciprofloxacin. The emergence of fluoroquinolone-resistant *V. cholerae* is ominous and may be a prelude to other pathogenic *Vibro* species acquiring resistance, which will create major problems in treating severe cases of diarrhea when an antibiotic intervention is deemed necessary.

REFERENCES

1. **Barry, A. L., and C. Thornsberry.** 1991. Susceptibility tests: diffusion test procedures, p. 1117–1125. *In* A. Balows, W. J. Hausler, Jr., K. L. Herrmann, H. D. Isenberg, and H. J. Shadomy (ed.), Manual of Clinical Microbiology, 5th ed. American Society for Microbiology, Washington, D.C.
2. **Bhattacharya, S. K., M. K. Bhattacharya, P. Dutta, D. Dutta, S. P. De, S. N. Sikdar, A. Maitra, A. Dutta, and S. C. Pal.** 1990. Double-blind, randomized, controlled clinical trial of norfloxacin for cholera. Antimicrob. Agents Chemother. **34:**939–940.
3. **Dutta, D., S. K. Bhattacharya, M. K. Bhattacharya, A. Deb. M. Deb, B. Manna, A. Moitra, A. K. Mukhopadhyay, and G. B. Nair.** 1996. Efficacy of norfloxacin and doxycycline for treatment of *Vibrio cholerae* O139 infection. J. Antimicrob. Chemother. **37:**575–581.
4. **Knapp, J. S., J. A. Hale, S. W. Neal, K. Wintersheid, R. J. Rice, and W. L. Whittington.** 1995. Proposed criteria for interpretation of susceptibilities of strains of *Neisseria gonorrhoeae* to ciprofloxacin, ofloxacin, enoxacin, lomefloxacin, and norfloxacin. Antimicrob. Agents Chemother. **39:**2442–2445.
5. **Mukhopadhyay, A. K., S. Garg, R. Mitra, A. Basu, D. Dutta, S. K. Bhattacharya, T. Shimada, T. Takeda, Y. Takeda, and G. B. Nair.** 1996. Temporal shifts in traits of *Vibrio cholerae* strains isolated from hospitalized patients in Calcutta: a 3-year (1993–1995) analysis. J. Clin. Microbiol. **34:**2537–2543.
6. **Murray, B. E.** 1994. Can antibiotic resistance be controlled? (Editorial.) N. Engl. J. Med. **330:**1229–1230.
7. **Threlfall, E. J., and B. Rowe.** 1982. *Vibrio cholerae* El Tor acquires plasmid-encoded reistance to gentamicin. Lancet **i:**42.
8. **World Health Organization.** 1993. Guidelines for cholera control. World Health Organization, Geneva.
9. **Yamamoto, T., G. B. Nair, M. J. Albert, C. C. Parodi, and Y. Takeda.** 1995.

Survey of in vitro susceptibilities of *Vibrio cholerae* O1 and O139 to antimicrobial agents. Antimicrob. Agents Chemother. **39:**241–244.

Asish Kumar Mukhopadhyay
Indira Basu
S. K. Bhattacharya
M. K. Bhattacharya
G. Balakrish Nair
National Institute of Cholera and Enteric Diseases
P-33, CIT Road, Scheme XM, Beliaghata
Calcutta 700 010
India