Enteric fever perspective in India: emergence of high-level ciprofloxacin resistance and rising MIC to cephalosporins

The emergence of multidrug-resistant enteric fever led to the use of ciprofloxacin as the mainstay of therapy. Furthermore, nalidixic acid-resistant *Salmonella typhi* (NARST) with decreased susceptibility to ciprofloxacin causing therapeutic failure emerged and became endemic (Threlfall & Ward, 2001). Consequently, there have been several isolated reports, including from India, on ciprofloxacin resistance (Adachi *et al.*, 2005; Capoor *et al.*, 2006; Cooke *et al.*, 2006; Joshi & Amarnath, 2006; Kownhar *et al.*, 2007; Mohanty *et al.*, 2006; Nair *et al.*, 2006; Renuka *et al.*, 2005; Saha *et al.*, 2006). Reports of cephalosporin resistance have also emerged (Saha *et al.*, 1999; Marano *et al.*, 1999).

The index case of ciprofloxacin-resistant *Salmonella enterica* serovar Paratyphi A (S. Paratyphi A) from our hospital was detected in December 2004 in the blood culture of a 7-year-old boy with septicaemia and bilateral cellulitis of the lower limbs of 2 months duration. He was treated for staphylococcal cellulitis with several broad-spectrum antimicrobials, including fluoroquinolones, in another hospital. In our hospital, his blood for culture grew S. Paratyphi A on the third day that was resistant to ciprofloxacin (MIC=256 µg ml⁻¹) and he was successfully treated with ceftriaxone. This study was initiated to evaluate the emerging resistance in enteric fever isolates over a span of 5 years (2001 and August 2005–August 2006).

The study was conducted in a 1570-bed tertiary care hospital in New Delhi. Patients from rural and urban areas of northern India are referred there for further management. In 2001, 178 isolates were collected, and 198 isolates were recovered between August 2005–August 2006 and included in the study. These isolates were compared for phenotypic traits including epidemiological parameters and antimicrobial susceptibility pattern.

Table 1. Antimicrobial profile of S. Typhi and S. Paratyphi A

Year	Organism	Antimicrobial* (% resistance)							
		A	C	Co	Na	Cf	Cfx	Ci	Cpm
2001	S. Typhi	32	41.4	57	51	0.6	0	0	0
	S. Paratyphi A	—	—	—	—	0	0	0	0
2005–2006	S. Typhi	7	7.4	11.1	92	15.4	0	0	0
	S. Paratyphi A	5.5	11.1	8.3	95	13.8	0	0	0

*A, Ampicillin; C, chloramphenicol; Co, trimethoprim/sulphamethoxazole; Na, nalidixic acid; Cf, ciprofloxacin; Cfx, cefixime; Ci, ceftriaxone; Cpm, cefepime.

Biochemical identification and serotyping with specific antisera (Central Research Institute, Kasauli, India) were performed to confirm all isolates. The antimicrobial susceptibility of the isolates was

determined by the disc diffusion method of Kirby Bauer on Mueller–Hinton agar using ampicillin (10 µg), chloramphenicol (30 µg), trimethoprim/sulphamethoxazole (1.25/23.75 µg), nalidixic acid (30 µg),

Table 2. Agar dilution MIC of ciprofloxacin, cefotaxime and cefepime for S. Typhi and S. Paratyphi A

Interpretive criteria (CLSI, 2006) of sensitive, intermediate and resistant strains, respectively, were as follows. Ciprofloxacin: ≤1 µg ml⁻¹, =2 µg ml⁻¹, ≥4 µg ml⁻¹; cefotaxime: ≤8 µg ml⁻¹, 16–32 µg ml⁻¹, ≥64 µg ml⁻¹; cefepime: ≤8 µg ml⁻¹, =16 µg ml⁻¹, ≥32 µg ml⁻¹.

MIC range (µg ml ⁻¹)	Ciprofloxacin		Cefotaxime		Cefepime	
	2001	2005–2006	2001	2005–2006	2001	2005–2006
≤0.0313	9	4	101	7	106	11
0.063	78	10	77*	16	72*	18
0.125	75*	16	0	106	0	117
0.25	11	26	0	51*	0	31
0.05	2	53	0	1	0	9
1	2	59*	0	7	0	6
2	0	2	—	4	—	7
4	1	0	—	6	—	0
8	0	5	—	—	—	—
16	0	1	—	—	—	—
32	0	9	—	—	—	—
64	0	5	—	—	—	—
128	0	1	—	—	—	—
256	0	1	—	—	—	—
≥512	0	6	—	—	—	—
Total	178	198	178	198	178	198

*MIC₉₀.

ciprofloxacin (5 µg), ceftriaxone (30 µg), cefixime (5 µg) and cefepime (30 µg). The MIC of ciprofloxacin, cefotaxime, and cefepime was determined by agar dilution, in accordance with CLSI guidelines (CLSI, 2006). The MIC experiments were repeated twice and the mean was taken.

In 2001, 59.6 % of enteric fever cases occurred in male patients, the median age of the patients was 22 years, and the majority (74 %) of cases occurred in the outpatient department. In 2005–2006, the respective figures were 64.7 %, 12 years and 70.2 %. There was an increase of 3.8 % in patients requiring hospitalization. The summer and monsoon months (April–September) uniformly showed the maximum rate of isolation (70 %). In 2001, 98.3 % of isolates were *S. Typhi* and 1.7 % were *S. Paratyphi A*, whereas in 2005–2006, 71.9 % were *S. Typhi* and 18 % were *S. Paratyphi A*. Table 1 depicts the comparative antimicrobial resistance profiles of enteric fever isolates in 2001 and August 2005–August 2006. Table 2 shows the comparative MICs of ciprofloxacin, cefotaxime and cefepime for the isolates.

Over a 5-year period, it was noted that the median age for enteric fever decreased significantly. There was an increase of 3.8 % in patients requiring hospitalization. This can be attributed to a commensurate increase in the numbers of NARST and instances of ciprofloxacin-resistant enteric fever, with treatment failures and morbidity. Emergence of *S. Paratyphi A* (18 % compared with 2 %) as a significant cause of enteric fever is in agreement with previous studies (Mandal *et al.*, 2004; Nair *et al.*, 2006). This is perhaps due to the reduction in *S. Typhi* isolation because of the typhoid-specific vaccination in this region. A sustained increased rate of isolation during summer and monsoon months implies that water conditions and sanitation remain the same or that a large number of carriers are present in the community (Gautam *et al.*, 2002).

Over a 5-year period, the numbers of NARST increased from 51 to 93 % and instances of ciprofloxacin-resistant enteric fever further increased from 0.6 to 15.2 %. The MIC₉₀ for ciprofloxacin increased from 0.125 µg ml⁻¹ to 1 µg ml⁻¹. Decreased therapeutic efficacy of quinolones is attributed to their

widespread indiscriminate use in human and veterinary medicine (Capoor *et al.*, 2006; Saha *et al.*, 2006). This is further corroborated by the fact that recent reports of ciprofloxacin resistance in enteric fever emerged from the Indian subcontinent, where NARST is endemic (Capoor *et al.*, 2006; Joshi & Amarnath, 2006; Kownhar *et al.*, 2007; Mohanty *et al.*, 2006; Saha *et al.*, 2006). This is perhaps the consequence of their indiscriminate prescription owing to their oral administration, easy availability and affordability. Multidrug resistance (ACCO) decreased from 30 to 7 %. Re-emergence of sensitivity to chloramphenicol in the current study and recent studies (Gautam *et al.*, 2002; Mandal *et al.*, 2004) is a direct consequence of its restricted use in the last decade. The rise in MIC₉₀ for third- and fourth-generation cephalosporins from 0.063 µg ml⁻¹ to 0.25 µg ml⁻¹ warrants their cautious use. Although ceftriaxone is well-established, cefixime and cefepime appear promising. These agents are prohibitively expensive for routine use in developing nations (Saha *et al.*, 2006). Moreover, there is also a concern that their extensive use in the outpatient setting will select β -lactamases, as has been observed in the hospital setting.

Resistance to quinolones and, more recently, increase in MIC levels for third- and fourth-generation cephalosporins have varying geographical patterns and re-emphasize the importance of continued surveillance in revision of enteric fever protocols.

Malini R. Capoor, Deepthi Nair, Monorama Deb and Pushpa Aggarwal

Department of Microbiology, Vardhman Mahavir Medical College & Safdarjung Hospital, New Delhi-110029, India

Correspondence: Deepthi Nair (deepthinair2@gmail.com)

Adachi, T., Sagara, H., Hirose, K. & Watanabe, H. (2005). Fluoroquinolone resistant *Salmonella enterica* serovar Paratyphi A. *Emerg Infect Dis* **11**, 172–174.

Capoor, M. R., Nair, D., Hasan, A. S., Aggarwal, P. & Gupta, B. (2006). Narrowing therapeutic options in typhoid fever, India. *Southeast Asian J Trop Med Public Health* **37**, 1170–1174.

CLSI (2006). *Performance Standards for Antimicrobial Susceptibility Testing*, 16th Informational Supplement, vol. 26. CLSI Document M100–S16. Wayne, PA: Clinical and Laboratory Standards Institute.

Cooke, F. J., Wain, J. & Threlfall, E. J. (2006). Fluoroquinolone resistance in *Salmonella* Typhi. *BMJ* **333**, 353–354.

Gautam, V., Gupta, N. K., Chaudhary, U. & Arora, D. R. (2002). Sensitivity pattern of salmonella serotypes in Northern India. *Braz J Infect Dis* **6**, 281–287.

Joshi, S. & Amarnath, S. K. (2006). Fluoroquinolone resistance in *Salmonella* Typhi and *S. Paratyphi A* in Bangalore, India. *Trans R Soc Trop Med Hyg* **101**, 308–310.

Kownhar, H., Shankar, E. M., Rajan, R. & Rao, G. A. (2007). Emergence of nalidixic acid-resistant *Salmonella enterica* serovar Typhi resistant to ciprofloxacin in India. *J Med Microbiol* **56**, 136–137.

Mandal, S., Mandal, M. D. & Pal, N. K. (2004). Reduced minimum inhibitory concentration of chloramphenicol for *Salmonella enterica* serovar Typhi. *Indian J Med Sci* **58**, 16–23.

Marano, N., Stamey, K., Jarrett, B. T., Bopp, C., Dabney, P., Angulo, F. J. & the NARMS Working Group (1999). Emerging quinolone and extended spectrum cephalosporin resistant salmonella in the United States. In *Proceedings of the 99th General Meeting of the American Society for Microbiology*, Chicago, IL, USA. Washington, DC: American Society for Microbiology.

Mohanty, S., Renuka, K., Sood, S., Das, B. K. & Kapil, A. (2006). Antibigram pattern and seasonality of *Salmonella* in a North Indian tertiary care hospital. *Epidemiol Infect* **134**, 961–966.

Nair, S., Unnikrishnan, M., Turner, K., Parija, S. C., Churcher, C., Wain, J. & Harish, B. N. (2006). Molecular analysis of fluoroquinolone resistance *Salmonella enterica* serovar Paratyphi A isolate, India. *Emerg Infect Dis* **12**, 489–491.

Renuka, K., Sood, S., Das, B. K. & Kapil, A. (2005). High-level ciprofloxacin resistance in *Salmonella enterica* serovar Typhi in India. *J Med Microbiol* **54**, 999–1000.

Saha, S. K., Talukder, S. Y., Islam, M. & Saha, S. (1999). A highly ceftriaxone resistant *Salmonella typhi* in Bangladesh. *Pediatr Infect Dis J* **18**, 387.

Saha, S. K., Darmstadt, G. L., Baqui, A. H., Crook, D. W., Islam, M. N., Hossain, M., Arifeen, S. E., Santoshan, M. & Black, R. E. (2006). Molecular basis of resistance displayed by highly ciprofloxacin resistant *Salmonella enterica* serovar Typhi in Bangladesh. *J Clin Microbiol* **44**, 3811–3813.

Threlfall, E. J. & Ward, L. R. (2001). Decreased susceptibility to ciprofloxacin in *Salmonella enterica* serovar Typhi, United Kingdom. *Emerg Infect Dis* **7**, 448–450.