Letter to the Editor

Third-generation cephalosporin resistance in clinical isolate of *Shigella sonnei* in Andaman & Nicobar Islands, India

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Diarrhoea is an important cause of morbidity and mortality in all regions of the world and among all ages [1]. It has been estimated that 91 million individuals worldwide contract shigellosis each year and among them 1.1 million die [2]. About, 410,000 (40%) of these deaths occur among Asian children [3]. Appropriate antibiotic treatment of shigellosis depends on identifying resistance patterns [4]. Rapid emergence of resistance warrants the need for continuous monitoring of sensitivity patterns [5]. The emergence of multiple-drug resistant (MDR) strains of *Shigella* sp., especially over the last two decades, has made the treatment for shigellosis more difficult. Currently ciprofloxacin (or other fluoroquinolones) is recommended as the drug of choice by the World Health Organization for the therapy of *Shigella* infections in both adults and children [6]. In addition, ceftriaxone, pivmecillinam (amdinocillin pivoxil) and azithromycin are considered as alternative drugs suitable for treatment of shigellosis [6]. *Shigella* species expressing extended-spectrum beta-lactamases (ESBLs) have emerged globally and this situation has limited the treatment strategies available for shigellosis. The archipelago of Andaman and Nicobar (92-94ºE; 6-14ºN), a chain of more than 500 islands situated about 1,200 km south-east of Indian peninsula in the Bay of Bengal, is a Union Territory of India. These islands are the home to 350,000 people including six indigenous tribes and settlers from mainland India. Health care is almost entirely provided by the government. G. B. Pant Hospital, located at Port Blair, the capital of the Union Territory, is the only referral hospital in the islands. Hospital-based bacteriological surveillance has identified shigellosis as endemic and a major cause of acute childhood diarrhoea [7,8] with *S. flexneri 2a* as the commonest isolate.

A stool sample was collected from a male child aged one and half years who was admitted in G. B. Pant Hospital prior to the administration of antimicrobials. He was admitted with high-grade fever (for three days), vomiting, and severe watery stool with blood. The patient was initially treated with intravenous fluid and powergyl (metronidazole 500 mg, norfloxacin 400 mg/5 mL), to which he did not respond. The sample was processed following standard techniques [9]. *S. sonnei* was isolated on Hektoen Enteric Agar (Difco, Detroit, USA) and confirmed by biotyping and serotyping. Antibiotic susceptibility testing was conducted using the disc diffusion method, according to Clinical and Laboratory Standards Institute guidelines [9] using the following antibiotic discs (Hi-Media, Mumbai, India): ampicillin (AMP, 10 µg), carbenicillin (CAR, 100 µg), imipenem (IMP, 30 µg), amoxicillin-clavulanic acid (AMC,20/10 µg), cefixime (CFM, 30 µg), cefuroxime (CXM, 5 µg), cephalothin (CEF, 30 µg), ceftriaxone (CRO, 30 µg), cefotaxime (CTX, 30 µg), ceftazidime (CAZ, 30 µg), tetracycline (TET, 30 µg), co-trimoxazole (CoT, 20 µg), nalidixic acid (NAL, 30 µg), ciprofloxacin (CIP, 30 µg), norfloxacin (NOR, 10 µg), ofloxacin (OFX, 5 µg), gatifloxacin (GAT, 5 µg), gentamicin (GEN, 10 µg), amikacin (AMK, 30 µg), nitrofurantoin (NIT, 300 µg), azithromycin (AZM, 30 µg), and chloramphenicol (CHL, 30 µg).

Control strains *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 were included in each test. The MICs of the third-generation cephalosporins including ceftriaxone, cefotaxime and ceftazidime were tested using Etest (AB Biodisk, Solna, Sweden).

The isolate displayed an ESBL phenotype and was co-resistant to co-trimoxazole, chloramphenicol, gentamicin, azithromycin and fluoroquinolones. The *S. sonnei* isolate exhibited resistance to amoxicillin-clavulanic acid with minimum inhibitory concentrations (MIC) > 256 µg/mL and > 256 µg/mL for ceftriaxone, 30 µg/mL for cefotaxime and > 256 µg/mL for ceftazidime. All these values are above the breakpoint for reduced susceptibility per the CLSI guidelines [9] and therefore were considered resistant to these drugs. The patient recovered completely without any complication after treatment with amikacin (40 mg) for a period of five days. Our laboratory test confirmed the sensitivity of the *S. sonnei* isolate to amikacin and nitrofurantoin.

The third-generation cephalosporin resistant *S. sonnei* strains were confirmed to produce ESBL due to an increase of 5 mm in zone diameter around ceftazidime-clavulanic acid disc compared to the zone around the ceftazidime disc, using the combination disc test. PCR based detection of the ESBL gene was performed according to standard protocol [10]. The isolate harbored the plasmid borne CTX-M3 gene.

Changing patterns of antimicrobial susceptibilities among *Shigella* isolates pose major difficulties in selecting an appropriate drug for the treatment of shigellosis [11]. Currently in India, third-generation cephalosporins are used as an alternative in patients who do not respond to fluoroquinolone treatment. Emergence of resistance to third-generation cephalosporins has been observed in these islands among other *Shigella* sp. except *S. sonnei* [12]. Resistance to third-generation cephalosporin in *S. sonnei* is very rarely reported from India. These findings portend the spread of serious resistance in *Shigella* throughout these Islands and beyond. The acquisition of resistance by enteric pathogens to an increasing number of antibacterial drugs is becoming a grave concern, particularly in developing countries where shigellosis is common. The options for effective and inexpensive antibacterial therapy for shigellosis are shrinking. A network of laboratories for real-time monitoring of antibiotic resistance among enteric pathogens and timely dissemination of such information to the clinicians for modification of treatment strategy are gravely needed.

Ethical approval

The study was approved by the Regional Medical Research Centre (Indian Council of Medical Research) ethics committee for research on humans.

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