



Discovery of a Fluoroquinolone-Resistant *Serratia marcescens* Clinical Isolate without Quinolone Resistance-Determining Region Mutations

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Sir,
Serratia marcescens, once considered to be an innocuous and non-pathogenic organism, is now an important cause of hospital-acquired infections. This organism is associated with respiratory tract infections, urinary tract infections, septicemia, meningitis, and wound infections [1, 2]. *S. marcescens* infections are difficult to treat because of high resistance to a wide variety of antibiotics, including cephalosporins, fluoroquinolones, and aztreonam [2].

Fluoroquinolones are broad-spectrum bactericidal antimicrobial agents that are used to treat various bacterial infections. Although *S. marcescens* infections are frequently treated with fluoroquinolones, the incidence of fluoroquinolone resistance continues to increase in clinical settings in China [2]. Fluoroquinolone resistance is mainly caused by chromosomal mutations affecting the quinolone resistance-determining region (QRDR) of *gyrA* and *gyrB*, which encode DNA gyrase subunits, and *parC* and *parE*, which encode topoisomerase IV subunits [3]. More-

over, plasmid-mediated quinolone resistance (PMQR) genes have been reported in gram-negative bacteria, including *S. marcescens*, and include the *qnr*, *qep*, and *oqx* systems [4]. The major cause of fluoroquinolone resistance is chromosomal mutation. The acquisition of PMQR genes alone results in a low level of fluoroquinolone resistance and does not lead to minimum inhibitory concentrations (MICs) exceeding the threshold of these agents [3].

We isolated a *S. marcescens* strain, designated as GN0780, from the wound drainage fluid of a 66-yr-old male patient who had femoral fractures and was admitted to the Department of Orthopedics at the People's Hospital of Huangshan (Huangshan, China) in 2011. The MICs of ciprofloxacin (CIP), levofloxacin (LVX), gatifloxacin (GAT), and nalidixic acid (NAL) exceeded the resistance thresholds proposed by the Clinical and Laboratory Standards Institute (2012) (Table 1) [5]. Surprisingly, direct DNA sequencing of the QRDRs did not reveal any mutations in

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Table 1. Fluoroquinolone susceptibility, QRDR mutations, and PMQRs in *Serratia marcescens* GN0780

Strain	QRDR mutations				PMQRs			MIC (mg/L)			
	<i>gyrA</i>	<i>gyrB</i>	<i>parC</i>	<i>parE</i>	<i>qnr</i>	<i>qep</i>	<i>oqx</i>	CIP	LVX	GAT	NAL
GN0780	WT	WT	WT	WT	–*	–	–	16	4	8	32

*None of the PMQR genes were identified in GN0780.

Abbreviations: QRDR, quinolone-resistance determining region; PMQR, plasmid-mediated quinolone resistance; MIC, minimum inhibitory concentration; WT, wild type; CIP, ciprofloxacin; LVX, levofloxacin; GAT, gatifloxacin; NAL, nalidixic acid.

gyrA, *gyrB*, *parC*, or *parE* when compared with the wild-type strain (Table 1). We then screened for the PMQR genes *qnrA*, *qnrB*, *qnrS*, *qnrC*, *qnrD*, *aac(6′)-Ib-cr*, *qepA*, and *oqxAB* by PCR. None of the PMQR genes were identified in *S. marcescens* GN0780 (Table 1).

Quinolones were introduced into clinical practice in the late 1960s. Although quinolone resistance was described soon after their introduction, the transmission mechanism of quinolone resistance was confirmed only in 1998. To date, five different PMQR mechanisms have been described in the literature, including target protection (Qnr), quinolone modification (AAC(6′)-Ib-cr), plasmid-encoded efflux systems (e.g., QepA or OqxAB), effect on bacterial growth rates, and natural transformation. Although PMQRs usually result in only a slight increase in the MICs of quinolones, they show an additive effect and may thus facilitate the acquisition of full quinolone resistance [6].

Several clinical bacterial isolates that have been reported to express phenotypic resistance do not exhibit corresponding genotypic mutations. This phenomenon has recently been studied in a clinical isolate of *Escherichia coli* HUE1 from Japan [7]. The authors suggested that the fluoroquinolone resistance in this HUE1 isolate, which does not have mutations in the QRDR, is caused by the coexistence of *oqxAB* and *qnrS*. OqxAB and QnrS increase the MIC of CIP by approximately 32-fold and 64-fold, respectively. However, other mechanisms may also be associated with fluoroquinolone resistance in HUE1.

Chopra and Galande [8] isolated an *Acinetobacter baumannii* mutant, designated as strain AB-7, which exhibited a CIP MIC of 16 mg/L. However, no mutation was detected in QRDRs, and no PMQR genes were present in AB-7. Our findings are consistent with the case of AB-7; *S. marcescens* GN0780 showed no mutation in QRDRs, and no PMQR gene was detected. We speculate that other mechanisms may be associated with fluoroquinolone resistance in GN0780; therefore, further investigations are needed. Our findings suggest an exception to the well-accepted mechanism of resistance to fluoroquinolones. These results also underscore the need of achieving deeper understanding of the mechanisms of action and evolution of resis-

tance to conventional antibiotics. In conclusion, we present the first report of fluoroquinolone resistance in *S. marcescens* lacking PMQR genes and mutations in QRDRs.

Authors' Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

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REFERENCES

- Mahlen SD. *Serratia* infections: from military experiments to current practice. Clin Microbiol Rev 2011;24:755-91.
- Yang HF, Cheng J, Hu LF, Ye Y, Li JB. Plasmid-mediated quinolone resistance in extended-spectrum-β-lactamase- and AmpC β-lactamase-producing *Serratia marcescens* in China. Antimicrob Agents Chemother 2012;56:4529-31.
- Ruiz J. Mechanisms of resistance to quinolones: target alterations, decreased accumulation and DNA gyrase protection. J Antimicrob Chemother 2003;51:1109-17.
- Strahilevitz J, Jacoby GA, Hooper DC, Robicsek A. Plasmid-mediated quinolone resistance: a multifaceted threat. Clin Microbiol Rev 2009;22:664-89.
- Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing, Twenty-Second Informational supplement, M100-S22. Wayne, PA: CLSI, 2012.
- Ruiz J, Pons MJ, Gomes C. Transferable mechanisms of quinolone resistance. Int J Antimicrob Agents 2012;40:196-203.
- Sato T, Yokota S, Uchida I, Okubo T, Ishihara K, Fujii N, et al. A fluoroquinolone-resistant *Escherichia coli* clinical isolate without quinolone resistance-determining region mutations found in Japan. Antimicrob Agents Chemother 2011;55:3964-5.
- Chopra S and Galande A. A fluoroquinolone-resistant *Acinetobacter baumannii* without the quinolone resistance-determining region mutations. J Antimicrob Chemother 2011;66:2668-70.