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High-Level Resistance to Fluoroquinolones Linked to Mutations in *gyrA*, *parC*, and *parE* in *Salmonella enterica* Serovar Schwarzengrund Isolates from Humans in Taiwan

Recent reports suggest high-level fluoroquinolone (FQ) resistance is emerging in *Salmonella enterica* serovar Typhimurium, *S. enterica* serovar Choleraesuis, and *S. enterica* serovar Schwarzengrund in different parts of the world (1–9).

In this study we analyzed high-level FQ resistance mechanisms in four *S. enterica* serovar Schwarzengrund strains which showed resistance levels to ciprofloxacin (Cip) MICs of 16 or 64 μg/ml and were isolated from stools or blood from patients in Taiwan (Table 1). Three of them were also multidrug resistant (Table 1).

Strains CGST2, CGST3, CGST4, and CGST5 carried up to five mutations in the quinolone target genes relative to those from *S. enterica* serovar Typhimurium, i.e., all carried a double mutation in the quinolone resistance-determining region (QRDR) of *gyrA*, leading to amino acid changes Ser83Phe and Asp87Asn or Asp87Gly, and a double mutation in the QRDR of *parC*, leading to amino acid changes Thr57Ser and Ser80Arg or Glu84Lys. Two strains displaying the highest levels of resistance to Cip carried an additional single mutation in the QRDR of *parE*, leading to amino acid change Ser458Pro. The ParC Thr57Ser amino acid change is likely not involved in quinolone resistance, because it was also identified in quinolone-susceptible *S. enterica* serovar Schwarzengrund control strain 383SA99 (Table 1). This amino acid change was, however, recently reported to be involved in decreased FQ susceptibility of *S. enterica* strains isolated in Hong Kong (7). Because we also found this amino acid change in a quinolone-susceptible *S. enterica* serovar Hadar strain (Table 1), it is probably the result of genetic divergence between the *parC* genes of *S. enterica* serovar Typhimurium, serovar Schwarzengrund, and serovar Hadar rather than being related to quinolone resistance. The ParC Ser80Arg and Glu84Lys amino acid changes found in the FQ-resistant *S. enterica* serovar Schwarzengrund strains have also been recently reported in FQ-resistant serovar Typhimurium isolates from patients in France, Hong Kong, Japan, and Taiwan (3, 6, 7, 8). The ParE Ser458Pro amino acid change found in strains CGST2 and CGST4 was also recently reported for FQ-resistant *S. enterica* serovar Typhimurium isolates from patients in Hong Kong (7). According to our data, this amino acid change, which is located at a same position as that found in the homologous GyrB protein (amino acid change Ser464Phe in FQ-resistant *S. enterica* serovar Typhimurium DT204 [2]), could account for a two- to fourfold increase in resistance levels to FQs (Table 1). The use of the efflux pump inhibitor Phe-Arg-β-naphthylamide (PAβN) decreased the FQ resistance levels 2- to 64-fold depending on FQ and the strain, suggesting participation of efflux in high-level FQ resistance (Table 1). The combination PAβN-enrofloxacin was the most effective in decreasing the resistance levels, as previously demonstrated (2).

In conclusion, high-level FQ resistance in *S. enterica* serovar Schwarzengrund strains appears to be linked to multiple target gene mutations at codon positions 83 and 87 for *gyrA*, codon positions 80 and 84 for *parC*, and codon position 458 for *parE*, as well as being linked to active efflux.

TABLE 1. Characteristics of the *S. enterica* serovar Schwarzengrund strains studied

Serovar	Strain	Origin[a]	Year of isolation	Antibiotic resistance profile[b]	MICs quinolones (μg/ml)[b]					Substitutions in QRDR[c]		
					NAL	FLU	ENR	MAR	CIP	GyrA	ParC	ParE
Schwarzengrund	CGST2	T	2000	Ap Cm Sm Sp Su Tc Tm	>1,024 [512]	>1,024 [64]	256 [4]	32 [4]	64 [16]	Ser83Phe; Asp87Gly	Thr57Ser, Ser80Arg	Ser458Pro
Schwarzengrund	CGST3	T	2000	Ap Cm Sm Sp Su Tc Tm	>1,024 [512]	1,024 [64]	64 [4]	16 [4]	16 [8]	Ser83Phe; Asp87Asn	Thr57Ser; Glu84Lys	
Schwarzengrund	CGST4	T	2001	Ap Cm Sm Sp Su Tc Tm	>1,024 [512]	>1,024 [64]	256 [4]	32 [4]	64 [16]	Ser83Phe; Asp87Gly	Thr57Ser; Ser80Arg	Ser458Pro
Schwarzengrund	CGST5	T	2002		>1,024 [512]	>1,024 [128]	64 [4]	16 [2]	16 [4]	Ser83Phe; Asp87Gly	Thr57Ser; Ser80Arg	
Controls												
Schwarzengrund	383SA99	B	1999		4	1	0.060	0.030	0.030		Thr57Ser	
Hadar	13SA02	B	2002		4	1	0.125	0.060	0.030		Thr57Ser	
Typhimurium	S/921495	S	1992		4	0.5	0.125	0.060	0.030			

[a] T, Taiwan; B, Belgium; S, Scotland.
[b] Antibiotics are ampicillin (Ap), chloramphenicol (Cm), streptomycin (Sm), spectinomycin (Sp), sulfonamide (Su), tetracycline (Tc), trimethoprim (Tm), nalidixic acid (NAL), flumequine (FLU), enrofloxacin (ENR), marbofloxacin (MAR), and ciprofloxacin (CIP). Numbers in brackets are the MICs in the presence of PABN at 80 μg/ml.
[c] No GyrB substitutions were detected.

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