

Prevalence of quinolone resistance in *Enterobacteriaceae* from Sierra Leone and the detection of *qnrB* pseudogenes and modified LexA binding sites.

**TABLE S1.** PCR primers and cycling conditions used in this study.

Primer name	Sequence (5' → 3') <sup>1</sup>	Target	Reference	PCR conditions <sup>2</sup>		
				Annealing	Cycles	Extension
qnrAm-F	AGAGGATTTCTCACGCCAGG	<i>qnrA</i>	(1)	53	30	1
qnrAm-R	TGCCAGGCACAGATCTTGAC					
qnrBm-F	GGMATHGAAATTCGCCACTG	<i>qnrB</i>	(2)	53	30	1
qnrBm-R	TTTGCGYYCGCCAGTCGAA					
psp2	AAATTTAAYCAGAAAAAGC	<i>qnrB</i> -full ORF	(3)	53	30	1
sc3	GCTSARGAGAACAGCTATAC					
ds3	ATGGCTGAAGTTGAGATTAT					
qnrC-F	GGGTTGTACATTTATTGAATCG	<i>qnrC</i>	(4)	53	30	1
qnrC-R	CACCTACCCATTTATTTTCA					
qnrD-F	CGAGATCAATTTACGGGGAATA	<i>qnrD</i>	(5)	53	30	1
qnrD-R	AACAAGCTGAAGCGCCTG					
qnrSm-F	GCAAGTTCATTGAACAGGGT	<i>qnrS</i>	(1)	53	30	1
qnrSm-R	TCTAAACCGTCGAGTTCGGCG					
qnrS_ext_fwd1	GGGTTGTAATGTGTTGATGTAACAGG	<i>qnrS</i> - full ORF	this study	50	40	2
qnrS_ext_rev1	CCCTATGTCTATTATTGCAAGGTTG					
qnrVC-F	AATTTTAAGCGCTCAAACCTCCG	<i>qnrVC</i>	(6)	53	30	1
qnrVC-R	TCCTGTTGCCACGAGCATATTTT					
aac(6)-Ib	TTGCGATGCTCTATGAGTGGCTA	<i>aac(6')</i> -Ib-cr	(7)	53	30	1
aac(6)-Ib-ext-R	ACG TCC CCC TCG ATG GAA G		this study			
qepA-F	AACTGCTTGAGCCCGTAGAT	<i>qepA</i>	(8)	50	45	1
qepA-R	GTCTACGCCATGGACCTCAC					
oqxAB-F	CTCGCAGCGTCTGTTACAG	<i>oqxA/oqxB</i>	this study	50	40	1
oqxAB-R	GAGACTGCTTCTGGGTGGTGATC					
gyrA-F	GTACTTTACGCCATGAACGT	<i>gyrA</i> (QRDR)	(8)	50	40	1
gyrA-R	TACCGTCATAGTTATCCACGA					
gyrA-alt-R	ATCACTTCCGTCAGGTTGTGC		this study			
parC-F	AATGAGCGATATGGCAGAGC	<i>parC</i> (QRDR)	(8)	50	40	1
parC-R	TTGGCAGACGGGCAGGTAG					
parC-kleb1-R	GCCAATTACGCAGGTTGTG		this study	53	40	2

<sup>1</sup> M = A or C; H = A or C or T; Y = C or T

<sup>2</sup> For all PCR amplifications the following general cycling program was used: the samples were denatured at 95°C for 2 min, followed by 30 to 45 cycles of denaturation at 95°C for 30 s, annealing at 50 to 60°C for 30 s, and extension at 72°C for 1 or 2 min. The table provides the actual values for – the annealing temperature in degrees Celsius (annealing), the number of cycles conducted (cycles) and the duration of extension part of the cycle in minutes (extension).

**TABLE S2.** Quinolone sensitivity phenotypes, PMQR genes and QRDR mutations.

Isolate <sup>1</sup>	Identification	Quinolone MIC <sup>2</sup>			PMQR genes detected <sup>3</sup>	QRDR mutations <sup>4</sup>			
		ND	CIP	MOX		GyrA		ParC	
						Ser83	Asp87	Ser80	Glu84
SL121	<i>Escherichia coli</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>qepA1</i>	Leu	Asn	Ile	
SL158	<i>Escherichia coli</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>qepA1</i>	Leu	Asn	Ile	
SL176	<i>Escherichia coli</i>	<u>≥256</u>	<u>≥32</u>	<b>24</b>	<i>qepA1</i>	Leu	Asn	Ile	Gly
SL152	<i>Escherichia coli</i>	<u>≥256</u>	0.75	0.75		Val		Arg	
SL169	<i>Escherichia coli</i>	<u>≥256</u>	0.38	0.5		Leu			
SL171	<i>Escherichia coli</i>	<u>≥256</u>	0.38	0.5		Leu			
SL178	<i>Escherichia coli</i>	<u>≥256</u>	0.25	0.25		Leu			
SL188	<i>Escherichia coli</i>	<u>≥256</u>	0.25	0.38		Leu			
SL115	<i>Escherichia coli</i>	2	0.016	0.064					
SL125	<i>Escherichia coli</i>	3	0.016	0.094					
SL133	<i>Escherichia coli</i>	2	0.016	0.047					
SL137	<i>Escherichia coli</i>	3	0.016	0.064					
SL159	<i>Escherichia coli</i>	3	0.012	0.19					
SL162	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>12</b>	<u>≥32</u>	<i>oqxAB</i>	Phe	Ala	Ile	
SL108	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>4</b>	<b>8</b>	<i>oqxAB</i>	Ile		Ile	
SL116	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>4</b>	<b>8</b>	<i>oqxAB</i>	Ile		Ile	
SL163	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>3</b>	<b>6</b>	<i>oqxAB</i>	Ile		Ile	
SL164	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>2</b>	<b>4</b>	<i>oqxAB</i>	Ile		Ile	
SL165	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>3</b>	<b>4</b>	<i>oqxAB</i>	Ile		Ile	
SL185	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<u>≥32</u>	<b>12</b>	<i>oqxAB, aac(6)-1b-cr, qnrB1d</i>	Ile		Ile	
SL186	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<b>4</b>	1.4	<i>oqxAB</i>	Ile		Ile	
SL190	<i>Klebsiella pneumoniae</i>	<u>≥256</u>	<u>≥32</u>	<b>16</b>	<i>oqxAB, aac(6)-1b-cr</i>	Tyr	Ala	Ile	
SL113	<i>Klebsiella pneumoniae</i>	<b>24</b>	<b>2</b>	<b>4</b>	<i>oqxAB, aac(6)-1b-cr, qnrB1d</i>				
SL136	<i>Klebsiella pneumoniae</i>	3	0.094	0.25	<i>oqxAB</i>				
SL141	<i>Klebsiella pneumoniae</i>	6	0.064	0.5	<i>oqxAB</i>				
SL180	<i>Klebsiella pneumoniae</i>	8	0.75	0.75	<i>oqxAB, aac(6)-1b-cr, qnrB1d</i>				

SL183	<i>Klebsiella pneumoniae</i>	3	0.032	0.094	<i>oqxAB</i>		
SL196	<i>Klebsiella pneumoniae</i>	12	<b>3</b>	1	<i>oqxAB, aac(6')-Ib-cr, qnrB1d</i>		
SL513 (e)	<i>Klebsiella pneumoniae</i>	3	0.032	0.064	<i>oqxAB</i>		
SL531 (e)	<i>Klebsiella pneumoniae</i>	3	0.032	0.094	<i>oqxAB</i>		
SL179	<i>Enterobacter cloacae</i>	16	<b>3</b>	1	<i>aac(6')-Ib-cr, qnrB1a</i>		
SL182	<i>Enterobacter cloacae</i>	3	<b>1.5</b>	0.75	<i>aac(6')-Ib-cr, qnrB1a</i>		
SL189	<i>Enterobacter cloacae</i>	12	1	0.75	<i>aac(6')-Ib-cr, qnrB1a</i>		
SL199	<i>Enterobacter cloacae</i>	16	<b>1.5</b>	0.75	<i>aac(6')-Ib-cr, qnrB1a</i>		
SL167	<i>Enterobacter cloacae</i>	<b>24</b>	0.75	<b>4</b>	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL168	<i>Enterobacter cloacae</i>	<b>24</b>	1	<b>8</b>	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL174	<i>Enterobacter cloacae</i>	12	0.75	0.75	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL175	<i>Enterobacter cloacae</i>	12	0.75	0.75	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL181	<i>Enterobacter cloacae</i>	12	0.5	0.75	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL195	<i>Enterobacter cloacae</i>	12	0.38	1	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL198	<i>Enterobacter cloacae</i>	12	0.75	0.75	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL203	<i>Enterobacter cloacae</i>	12	1	0.75	<i>aac(6')-Ib-cr, qnrB1i</i>		
SL161	<i>Enterobacter cloacae</i>	<b>24</b>	<b>3</b>	<b>12</b>	<i>oqxAB, aac(6')-Ib-cr, qnrB6</i>		
SL160	<i>Enterobacter cloacae</i>	6	0.125	2	<i>qnrS1</i>	Thr	
SL192	<i>Enterobacter cloacae</i>	<u>≥256</u>	<b>1.5</b>	<b>16</b>	<i>qnrS1</i>	Ile	Ile
SL512 (e)	<i>Enterobacter cloacae</i>	6	0.19	0.5	<i>qnrS1</i>		
SL184	<i>Enterobacter sp./Leclercia sp.</i>	<u>≥256</u>	<u>≥32</u>	<b>6</b>	<i>aac(6')-Ib-cr, qnrB1a</i>	Ile	Ile
SL191	<i>Enterobacter sp./Leclercia sp.</i>	<u>≥256</u>	<u>≥32</u>	<b>6</b>	<i>aac(6')-Ib-cr, qnrB1a</i>	Ile	Ile
SL201	<i>Enterobacter sp./Leclercia sp.</i>	<u>≥256</u>	<u>≥32</u>	<b>12</b>	<i>aac(6')-Ib-cr, qnrB1a</i>	Ile	Ile
SL204	<i>Enterobacter sp./Leclercia sp.</i>	<u>≥256</u>	<u>≥32</u>	<b>6</b>	<i>aac(6')-Ib-cr, qnrB1a</i>	Ile	Ile
SL129	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrA1, Δ<sup>1</sup>qnrB<sup>5</sup></i>	Ile	Ile
SL156	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB82, Δ<sup>3</sup>qnrB, qnrS1</i>	Ile	Ile
SL157	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB81, Δ<sup>3</sup>qnrB, qnrS1</i>	Ile	Ile
SL187	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<b>12</b>	<i>aac(6')-Ib-cr, qnrB1a, Δ<sup>3</sup>qnrB</i>	Ile	Ile
SL197	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<b>16</b>	<i>aac(6')-Ib-cr, qnrB1a, Δ<sup>3</sup>qnrB</i>	Ile	Ile
SL150	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB1d, Δ<sup>1</sup>qnrB</i>	Ile	Ile
SL172	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB1d, Δ<sup>1</sup>qnrB</i>	Ile	Ile

SL200	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB1d, Δ<sup>1</sup>qnrB</i>	Ile		Ile
SL202	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB1d, Δ<sup>1</sup>qnrB</i>	Ile		Ile
SL205	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB1d, Δ<sup>1</sup>qnrB</i>	Ile		Ile
SL154	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>qnrB12</i>	Ile	Tyr	Ile
SL155	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>qnrB12</i>	Ile	Tyr	Ile
SL194	<i>Citrobacter freundii</i>	<u>≥256</u>	<u>≥32</u>	<u>≥32</u>	<i>qnrB12</i>	Ile	Tyr	Ile
SL124	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>12</b>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL127	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>12</b>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL151	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>16</b>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL153	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>12</b>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL170	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>12</b>	<u>≥32</u>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL173	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>12</b>	<b>12</b>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL177	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>16</b>	<b>6</b>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL193	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>16</b>	<b>12</b>	<i>aac(6')-Ib-cr, qnrB6, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL122	<i>Citrobacter freundii</i>	<u>≥256</u>	<b>8</b>	<b>16</b>	<i>aac(6')-Ib-cr, Δ<sup>2</sup>qnrB, qnrS1</i>	Ile		
SL166	<i>Escherichia hermannii</i>	12	<b>3</b>	2	<i>aac(6')-Ib-cr, qnrB1a, qnrS1</i>			
SL522 (e)	<i>Pantoea dispersa</i>	2	0.016	0.047		ND	ND	ND

<sup>1</sup> All of the isolates were cultured from outpatient urine samples collected at Mercy Hospital, Bo Sierra Leone with the exception of SL513, SL531, SL512 and SL522 that were recovered from the hospital environment (e).

<sup>2</sup> Minimum inhibitory concentrations (MIC) were measured using E-test assays. CIP – ciprofloxacin, ND – nalidixic acid, MOX – moxifloxacin. The values in bold font only designate intermediate resistance whereas those that are in bold font and underlined denote high clinical resistance. All MIC values in regular font denote clinical sensitivity. The isolates in the table were listed in the order of decreasing MIC values.

<sup>3</sup> The presence of the following PMQR genes was tested: *qnrA, qnrB, qnrC, qnrD, qnrS, qnrVC, aac(6')-Ib-cr, qepA*, and *oqxAB*.

<sup>4</sup> Substitutions were deduced via *in silico* translation of the obtained DNA sequence. The position numbers are based on *E. coli* GyrA and ParC protein sequences.

<sup>5</sup> The  $\Delta^1$ *qnrB* sequence is identical to that previously described in the chromosome of *C. freundii* ATCC 43864 (9) and the sequences of  $\Delta^2$ *qnrB* and  $\Delta^3$ *qnrB* differ from  $\Delta^1$ *qnrB* by several point mutations.

## References

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