



Article

Burden, antibiotic resistance, and clonality of *Shigella flexneri* implicated in community-acquired acute diarrhoea in Lilongwe, Malawi

Abel F.N.D. Phiri ^{1,2}, Akebe Luther King Abia ¹, Daniel Gyamfi Amoako ¹, Rajab Mkakosya ³,
Sabiha Y. Essack ¹ and Gunnar Skov Simonsen ^{4,5*}



Table S1. Primer sequences for detection of ampicillin, sulfonamide and trimethoprim resistant genes in *Shigella* spp.

Gene target	Primer	Sequence (5' to 3')	Annealing Temp (°C)	Reference
OXA-1	OXA-1F	ACACAATAACATATCAACTTCGC	55	Ouellette & Bissonnette, 1987
group	OXA-1R	AGTGTGTTAGAACATGGTGATC		
OXA-2	OXA-2F	ATGGCAATCCGAATCTTCG	55	Chmelnitsky et al., 2005
group	OXA-2R	TTATCGCGCAGCGTCCGAG		
dfrA1	dfrA1F	ACGGATCCTGGCTGTTGGTGGACGC	58	Lombardo et al., 2016
	dfrA1R	CGGAATTCACCTTCCGGCTCGATGTC		
dfrA12	dfrA12F	GTTGCGGTCCAGACATAC	58	Thungpathra et al., 2002
	dfrA12R	CCGCCACCAAGACACTA		
dfrA17	dfrA17F	TCGAGCTTCATGCCATT	58	Al-Assil, 2013
	dfrA17R	TCTTCCATGCCATTCTGC		
sul1	sul1F	CTTCGATGAGAGCCGGCGC	*	Ruiz, 2002
	sul1R	GCAAGGCGGAAACCCCGCGC		
sul2	sul2F	TCGTCAACATAACCTCGGACAG	*	Byne-Bailey, 2009
	sul2R	GTTGCGTTGATACCGGCAC		
sul3	sul3F	GAGCAAGATTTTGGAAATCG	*	Perreten, 2003
	sul3R	CATCTGCAGCTAACCTAGGGCTTGGA		

* Annealing temperatures were optimised at 62 °C as reported in the manuscript

**Table S2.** Primer sequences for detecting virulence genes in *Shigella* spp. isolates.

Gene target	Primer	Sequence (5' to 3')	Product size (bp)	Tm (°C)
<i>Set 1A</i>	<i>ShET-1A</i>	F: TCACGCTACCATCAAAGA R: TATCCCCCTTGTTGGTGGTA	309	55
<i>Set 1B</i>	<i>ShET-1B</i>	F: GTGAACCTGCTGCCGATATC R: ATTTGTGGATAAAAATGACG	147	55
<i>sat</i>	<i>Sat1</i>	F: ACTGGCGGACTCATGCTGT R: AACCCCTGTAAGAAGACTGAGC	387	55
<i>Ial</i>	<i>Ial1</i>	F: CTGGATGGTATGGTGAGG R: GGAGGCCAACAAATTATTCC	320	58
<i>ipaH</i>	<i>Shig1</i>	F: TGGAAAAACTCAGTCGCCTCT R: CCAGTCCGTAAATTCAATTCT	423	58
<i>virA</i>	<i>virA</i>	F: CTGCATTCTGGCAATCTCTTCACATC R-TGATGAGCTAACCTCGTAAGCCCTCC	215	58
<i>Stx</i>	<i>Stx1</i>	F: CAGTTAATGTGGTTGCGAAG R: CTGCTAACATAGTTCTGCGCATC	895	60
<i>Sen</i>	<i>ShET2</i>	F: ATGTGCCTGCTATTATTAT R: CATAATAATAAGCGGTCAAGC	799	60
<i>ipaBCD</i>	<i>ipaBCD</i>	F: GCTATAGCAGTGACATGG R: ACGAGTTCGAAGCACTC	612	60

Source: Yaghoubi, et al., 2017

**Table S3.** Detection of sul, dfrA and OXA genes in phenotypically resistant isolates.

Isolate number	Location	Resistance genes							
		sul1	sul2	sul3	dfrA1	dfrA12	dfrA17	OXA-1	OXA-2
1	Area 36	+	+	+	-	+	+	-	-
2	Area 46	+	+	+	-	+	+	-	-
3	Area 25	+	+	-	-	+	+	-	-
4	Chinsapo	+	+	+	-	+	+	-	-
5	Kawale	+	+	+	-	+	+	-	-
6	Mchesi	+	+	+	-	+	+	-	-
7	Area 24	+	+	+	-	+	+	-	-
8	Kawale	+	+	+	-	+	+	-	-
9	Mtsiliza	+	+	+	-	+	+	-	-
10	Chinsapo	+	+	-	-	-	-	-	-
11	Area 23	+	+	-	-	-	-	-	-
12	Chinsapo	-	-	-	-	-	-	-	-
13	Kawale 2	+	+	+	-	+	+	-	-
14	Mchesi	+	+	-	-	+	-	-	-
15	Likuni	+	+	+	-	+	+	-	-
16	Area 24	+	+	-	-	+	+	-	-
17	Chinsapo	+	+	-	-	+	-	-	-
18	Area 36	+	+	+	-	+	+	-	-
19	Chinsapo	-	+	-	-	+	+	-	-
20	Biwi	+	+	-	-	-	-	-	-
21	Area 24	-	-	-	-	-	-	-	-
22	Area 36	-	-	-	-	-	-	-	-
23	Area 36	+	+	+	-	+	+	-	-
24	Chinsapo	+	+	-	-	+	-	-	-
25	Area 23	+	+	+	-	-	+	-	-
26	Chinsapo	+	+	+	-	+	+	-	-
27	Area 36	+	+	-	-	+	+	-	-
28	Area 1	-	+	+	-	+	+	-	-
29	Mtsiliza	+	+	-	-	+	-	-	-
30	Area 46	+	+	-	-	+	-	-	-
31	Area 36	-	-	-	-	-	-	-	-
32	Chigwirizano	+	-	-	-	-	-	-	-
33	Area 46	+	+	+	-	+	-	-	-
34	Mtandile	-	-	-	-	-	-	-	-
Total		27	28	7	0	24	19	0	0

+ = gene detected, - = gene not detected