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Azithromycin Resistance Patterns in *Escherichia coli* and *Shigella* before and after COVID-19, Kenya

Appendix.

Appendix Table 1. Primer sequences used in multiplex PCR for detection of diarrheagenic *Escherichia coli*.

<i>E. coli</i> type	Target gene	Primer sequences (5' to 3')	Product size in bp
ETEC	<i>elt</i>	CACACGGAGCTCCTCAGTC CCCCCAGCCTAGCTTAGTTT	508
	<i>est</i>	GCTAAACCACTAGAGGTCT CCCGGTACAGAGCAGGATTACAACA	147
EHEC	<i>Stx1</i>	CAGTTAATGTGGTGGCGAAGG CACCAGACAATGTAACCGCTG	384
	<i>Stx2</i>	ATCCTATTCCCGGGAGTTACG GCGTCATCGTATACACAGGAGC	584
EPEC	<i>eae</i>	CCCGAATTCGGCACAAGCATAAGC CCCGGATCCGTCTCGCCAGTATTCG	881
	<i>bfpA</i>	GGAAGTCAAATTCATGGGGGTAT GGAATCAGACGCAGACTGGTAGT	300
EIEC	<i>ipaH</i>	TGGAAAACTCAGTGCCTCT CCAGTCCGTAATTCATTCT	423
EAEC	<i>aatA</i>	CTGGCGAAAGACTGTATCAT CAATGTATAGAAATCCGCTGTT	650
	<i>aaiC</i>	ATTGTCCTCAGGCATTTTAC ACGACACCCCTGATAAACAA	215

Appendix Table 2. Antibiotic susceptibility of *E. coli* cases among children in Kenya before and after the COVID-19 pandemic.

Characteristics	Overall	Pre-COVID	Post-COVID	p-value*
Children <5 years				
No. cases	47	27	20	--
% Resistant†				
CIP	2.2%	0.0%	5.0%	0.43
LEV	2.2%	0.0%	5.0%	0.43
AZM	21.3%	7.4%	40.0%	0.01
Children 5-17 years				
No. cases	28	17	11	--
% Resistant†				
CIP	0.0%	0.0%	0.0%	>0.99
LEV	0.0%	0.0%	0.0%	>0.99
AZM	25.0%	11.8%	45.5%	0.08
All children <18 years				
No. cases	75	44	31	--
% Resistant†				
CIP	1.4%	0.0%	3.2%	0.42
LEV	1.4%	0.0%	3.2%	0.42
AZM	22.7%	9.1%	41.9%	0.002

One CIP and LEV result removed from calculations of antibiotic susceptibility in children <5 years due to an inconclusive result.

CIP = Ciprofloxacin; LEV = Levofloxacin; AZM = Azithromycin

*Pearson's chi-square test used to measure differences in prevalence across COVID-19 periods. Fisher's exact test used to measure difference in resistance proportions between the pre-COVID-19 and post-COVID-19 periods.

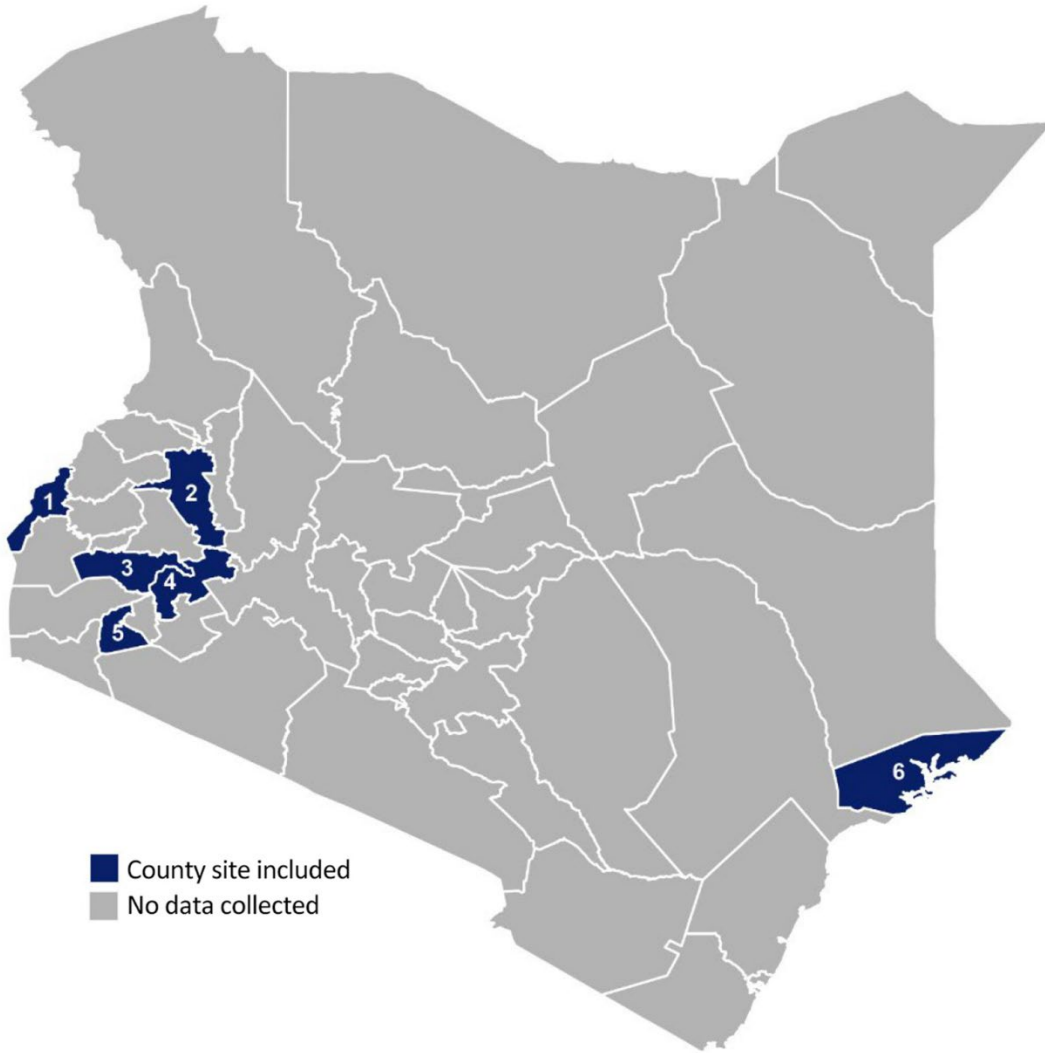
†Percent of all *E. coli* cases (No. cases column).

Appendix Table 3. Azithromycin susceptibility of *E. coli* and *Shigella* spp. isolates by water source and treatment

Water source/treatment	<i>E. coli</i>			<i>Shigella</i> spp.		
	Susceptible	Resistant	p-value	Susceptible	Resistant	p-value*
All ages	N=90	N=26		N=105	N=4	
Source						
Borehole	16 (17.8%)	6 (24.0%)	0.57	16 (15.2%)	2 (50.0%)	0.13
Rain	19 (21.1%)	3 (12.0%)	0.4	29 (27.6%)	1 (25.0%)	>0.99
Well	5 (5.6%)	2 (8.0%)	0.64	10 (9.5%)	1 (25.0%)	0.35
Bottle	3 (3.3%)	0 (0.0%)	>0.99	4 (3.8%)	0 (0.0%)	>0.99
Municipal	41 (45.6%)	13 (52.0%)	0.57	55 (52.4%)	1 (25.0%)	0.35
Spring	9 (10.0%)	0 (0.0%)	0.2	16 (15.2%)	2 (50.0%)	0.13
Stream	1 (1.1%)	0 (0.0%)	>0.99	0 (0.0%)	0 (0.0%)	>0.99
Tap	1 (1.1%)	0 (0.0%)	>0.99	0 (0.0%)	0 (0.0%)	>0.99
Other	0 (0.0%)	0 (0.0%)	>0.99	1 (1.0%)	0 (0.0%)	>0.99
Treatment						
Boil	15 (16.7%)	5 (20.0%)	0.77	20 (19.0%)	2 (50.0%)	0.18
Distillation	0 (0.0%)	0 (0.0%)	>0.99	1 (1.0%)	0 (0.0%)	>0.99
Chemical	1 (1.1%)	0 (0.0%)	>0.99	1 (1.0%)	0 (0.0%)	>0.99
Chlorine	0 (0.0%)	0 (0.0%)	>0.99	1 (1.0%)	0 (0.0%)	>0.99
Water guard	10 (11.1%)	4 (16.0%)	0.5	10 (9.5%)	0 (0.0%)	>0.99
No Tx	65 (72.2%)	16 (64.0%)	0.43	72 (68.6%)	2 (50.0%)	0.59
Adults ≥18 years						
Source	N=31	N=10		N=53	N=4	
Borehole	10 (32.3%)	1 (11.1%)	0.4	12 (22.6%)	2 (50.0%)	0.25
Rain	6 (19.4%)	3 (33.3%)	0.39	17 (32.1%)	1 (25.0%)	>0.99
Well	3 (9.7%)	2 (22.2%)	0.31	7 (13.2%)	1 (25.0%)	0.46
Bottle	1 (3.2%)	0 (0.0%)	>0.99	1 (1.9%)	0 (0.0%)	>0.99
Municipal	7 (22.6%)	4 (44.4%)	0.23	23 (43.4%)	1 (25.0%)	0.63
Spring	2 (6.5%)	0 (0.0%)	>0.99	8 (15.1%)	2 (50.0%)	0.14
Stream	0 (0.0%)	0 (0.0%)	>0.99	0 (0.0%)	0 (0.0%)	>0.99
Tap	1 (3.2%)	0 (0.0%)	>0.99	0 (0.0%)	0 (0.0%)	>0.99
Other	0 (0.0%)	0 (0.0%)	>0.99	0 (0.0%)	0 (0.0%)	>0.99
Treatment						
Boil	5 (16.1%)	1 (11.1%)	>0.99	8 (15.1%)	2 (50.0%)	0.14
Distillation	0 (0.0%)	0 (0.0%)	>0.99	1 (1.9%)	0 (0.0%)	>0.99
Chemical	0 (0.0%)	0 (0.0%)	>0.99	1 (1.9%)	0 (0.0%)	>0.99
Chlorine	0 (0.0%)	0 (0.0%)	>0.99	1 (1.9%)	0 (0.0%)	>0.99
Water guard	4 (12.9%)	0 (0.0%)	0.56	3 (5.7%)	0 (0.0%)	>0.99
No Tx	22 (71.0%)	8 (88.9%)	0.4	39 (73.6%)	2 (50.0%)	0.31
Children <18 years						
Source						
Borehole	6 (10.2%)	5 (31.3%)	0.05	4 (7.7%)	0 (NA%)	NA
Rain	13 (22.0%)	0 (0.0%)	0.058	12 (23.1%)	0 (NA%)	NA
Well	2 (3.4%)	0 (0.0%)	>0.99	3 (5.8%)	0 (NA%)	NA
Bottle	2 (3.4%)	0 (0.0%)	>0.99	3 (5.8%)	0 (NA%)	NA
Municipal	34 (57.6%)	9 (56.3%)	0.92	32 (61.5%)	0 (NA%)	NA
Spring	7 (11.9%)	0 (0.0%)	0.33	8 (15.4%)	0 (NA%)	NA
Stream	1 (1.7%)	0 (0.0%)	>0.99	0 (0.0%)	0 (NA%)	NA
Tap	0 (0.0%)	0 (0.0%)	>0.99	0 (0.0%)	0 (NA%)	NA
Other	0 (0.0%)	0 (0.0%)	>0.99	1 (1.9%)	0 (NA%)	NA
Treatment						
Boil	10 (16.9%)	4 (25.0%)	0.48	12 (23.1%)	0 (NA%)	NA
Distillation	0 (0.0%)	0 (0.0%)	>0.99	0 (0.0%)	0 (NA%)	NA
Chemical	1 (1.7%)	0 (0.0%)	>0.99	0 (0.0%)	0 (NA%)	NA
Chlorine	0 (0.0%)	0 (0.0%)	>0.99	0 (0.0%)	0 (NA%)	NA
Water guard	6 (10.2%)	4 (25.0%)	0.21	7 (13.5%)	0 (NA%)	NA
No Tx	43 (72.9%)	8 (50.0%)	0.082	33 (63.5%)	0 (NA%)	NA

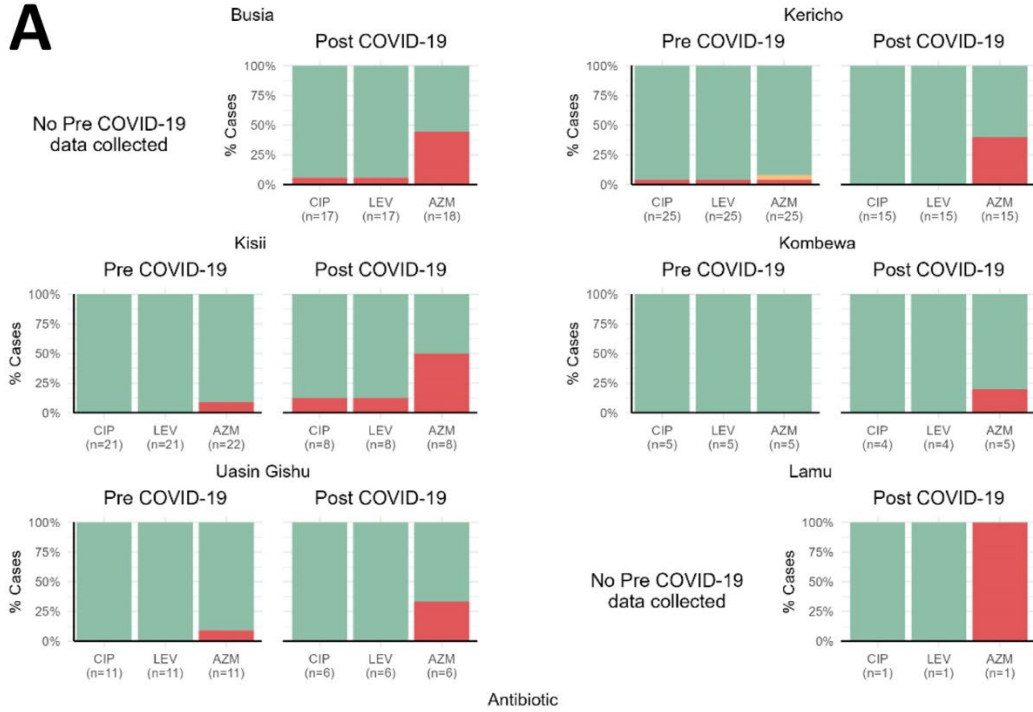
Azithromycin susceptibility profile of *E. coli* and *Shigella* spp. isolates by water source and treatment, stratified by age group.

*Fisher's exact test

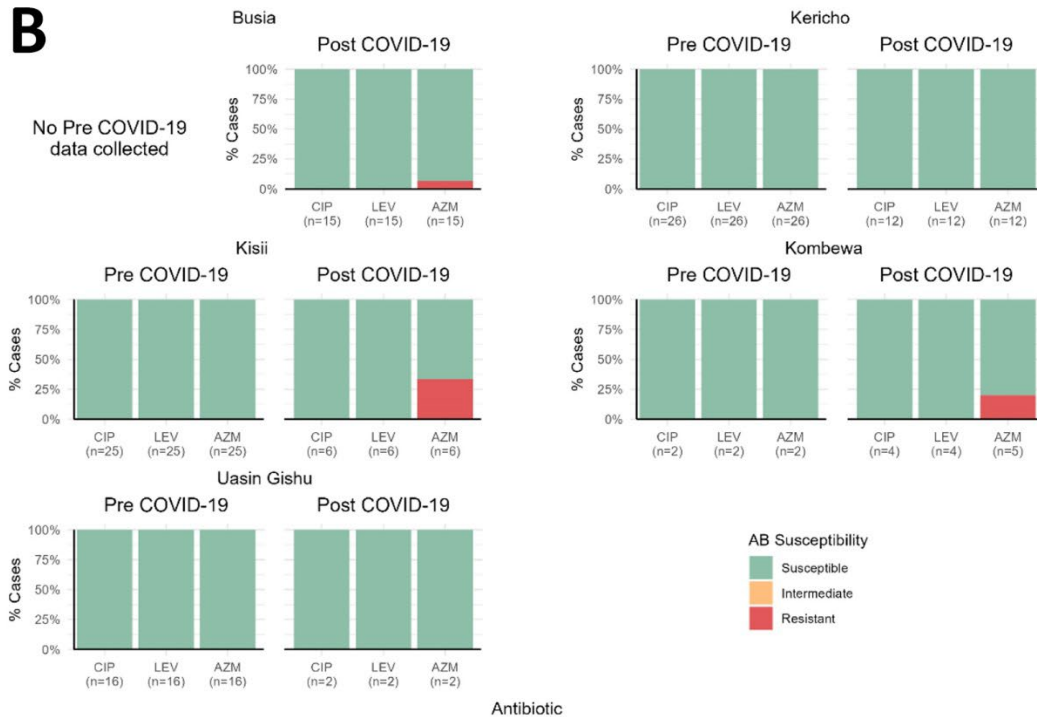


Appendix Figure 1. Map of Kenya showing the regional distribution of surveillance sites by residing county: 1) Busia, 2) Uasin Gishu, 3) Kisumu, 4) Kericho, 5) Kisii, 6) Lamu.

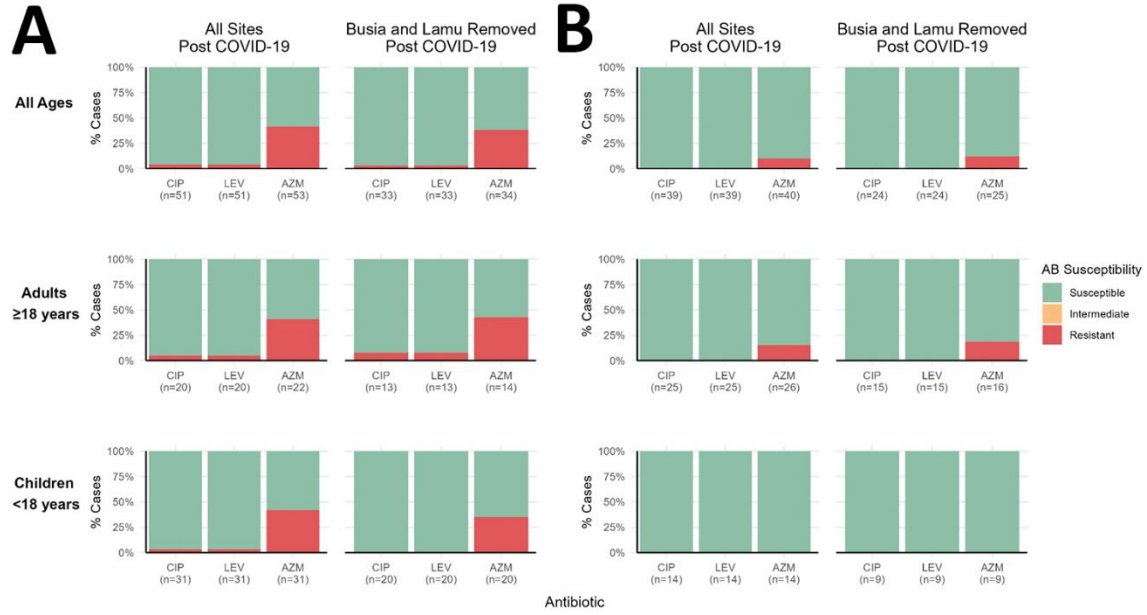
A



B



Appendix Figure 2. Antibiotic susceptibility profile of *E. coli* and *Shigella* spp. isolates by site. A) *E. coli* isolates, B) *Shigella* spp. isolates. CIP = Ciprofloxacin; LEV = Levofloxacin; AZM = Azithromycin.



Appendix Figure 3. Post-COVID-19 antibiotic susceptibility profile of *E. coli* and *Shigella* spp. isolates excluding post-COVID-only collection sites. A) *E. coli* isolates, B) *Shigella* spp. isolates. CIP = Ciprofloxacin; LEV = Levofloxacin; AZM = Azithromycin.