Additional File 1

Table S1. List of genes, sequences of primers, cycling conditions for PCR assays, and amplicon sizes.

Assay	Gene	Primer sequence	PCR program[a]	Product size (bp)	Reference
E. coli confirmation	*uspA*	F: 5'-CCGATACGCTGCCAATCAGT-3'	94°C/ 120 s, 70°C/ 60 s, 72°C/ 60 s (30 cycles)	884	1
		R: 5'-ACGCAGACCGTAGGCCAGAT-3'			
Phylogenetic typing (Triplex method)	*chuA*	F: 5'-GACGAACCAACGGTCAGGAT-3'	94°C/ 30 s, 55°C/ 30 s, 72°C/ 30 s (30 cycles)	279	2
		R: 5'-TGCCGCCAGTACCAAAGACA-3'			
	yjaA	F: 5'-TGAAGTGTCAGGAGACGCTG-3'		211	
		R: 5'-ATGGAGAATGCGTTCCTCAAC-3'			
	TspE4.C2	F: 5'-GAGTAATGTCGGGGCATTCA-3'		152	
		R: 5'-CGCGCCAACAAAGTATTACG-3'			
Phylogenetic typing (Quadruplex method)	*chuA*	F: 5'-ATGGTACCGGACGAACCAAC-3'	94°C/ 5 s, 57°C/ 20 s (30 cycles)	288	3
		R: 5'-TGCCGCCAGTACCAAAGACA-3'			
	yjaA	F: 5'-CAAACGTGAAGTGTCAGGAG-3'		211	
		R: 5'-AATGCGTTCCTCAACCTGTG-3'			
	TspE4.C2	F: 5'-CACTATTCGTAAGGTCATCC-3'		152	

		R: 5′-AGTTTATCGCTGCGGGTCGC-3′			
	arpA (Group F)	F: 5′-AACGCTATTCGCCAGCTTGC-3′		400	
		R: 5′-TCTCCCCATACCGTACGCTA-3′			
	arpA (Group E)	F: 5′-GATTCCATCTTGTCAAAATATGCC-3′		301	
		R: 5′-GAAAAGAAAAAGAATTCCCAAGAG-3′			
	trpA (Group C)	F: 5′-AGTTTTATGCCCAGTGCGAG-3′	94°C/ 5 s, 59°C/ 20 s (30 cycles)	219	
		R: 5′-TCTGCGCCGGTCACGCCC-3′			
	trpA (Internal control)	F: 5′-CGGCGATAAAGACATCTTCAC-3′	94°C/ 5 s, 57°C/ 20 s (30 cycles)	489	
		R: 5′-GCAACGCGGCCTGGCGGAAG-3′			
DEC[b]	*eae*	F: 5′-GACCCGGCACAAGCATAAGC-3′	95°C/ 30 s, 54°C/ 90 s, 72°C/ 90 s (30 cycles)	384	4, 5
		R: 5′-CCACCTGCAGCAACAAGAGG-3′			
	stx1	F: 5′-ATAAATCGCCATTCGTTGACTAC-3′	95°C/ 30 s, 52°C/ 60 s, 72°C/ 60 s (35 cycles)	180	
		R: 5′-AGAACGCCCACTGAGATCATC-3′			
	stx2	F: 5′-GGCACTGTCTGAAACTGCTCC-3′		255	
		R: 5′-TCGCCAGTTATCTGACATTCTG-3′			

	ltA	F: 5’-GGCGACAGATTATACCGTGC-3’	94°C/ 60 s, 50°C/ 60 s, 72°C/ 120 s (35 cycles)	696	
		R: 5’-CCGAATTCTGTTATATATGTC-3’			
	st1A	F: 5’-TCTGTATTATCTTTCCCCTC-3’		186	
		R: 5’-ATAACATCCAGCACAGGC-3’			
	ipaH	F: 5’-GTTCCTTGACCGCCTTTCCGATACCGTC-3’	94°C/ 40 s, 60°C/ 60 s, 72°C/ 60 s (35 cycles)	620	
		R: 5’-GCCGGTCAGCCACCCTCTGAGAGTAC-3’			
	bfpA	F: 5’-ATTGAATCTGCAATGGTGC-3’	95°C/ 40 s, 55°C/ 40 s, 72°C/ 40 s (30 cycles)	461	
		R: 5’-ATAGCAGTCGATTTAGCAGCC-3’			
	aggA	F: 5’-ATGCATTACTTTGGGTTTAG-3’	94°C/ 60 s, 50°C/ 60 s, 72°C/ 120 s (35 cycles)	414	
		R: 5’-TCAACCTTGACACTTGCC-3’			
ExPEC[c]	*vat*	F: 5’-TCAGGACACGTTCAGGCATTCAGT-3’	94°C/ 30 s, 63°C/ 90 s, 72°C/ 90 s	1100	6
		R: 5’-GGCCAGAACATTTGCTCCCTTGTT-3’			
	fyuA	F: 5’-GTAAACAATCTTCCCGCTCGGCAT-3’		850	

		R: 5’-TGACGATTAACGAACCGGAAGGGA-3’	(30 cycles)		
	chuA	F: 5’-CTGAAACCATGACCGTTACG-3’		652	
		R: 5’-TTGTAGTAACGCACTAAACC-3’			
	yfcV	F: 5’-ACATGGAGACCACGTTCACC-3’		292	
		R: 5’-GTAATCTGGAATGTGGTCAGG-3’			

[a]Before starting the PCR cycle, DNA was first denatured at 95^{O}C/15 min. After completion of the cycle, there was a final primer extension at 72^{O}C/8 min.

[b]Diarrheagenic *E. coli* are defined by the following genes: *eae*+*bfpA*/*eae* (EPEC), *ltA*+*st1A*/*ltA*/*st1A* (ETEC), *ipaH* (EIEC), *eae*+*stx1*+*stx2*/*stx1*/*stx2* (STEC), and *aggA* (EAEC).

[c]Extra-intestinal pathogenic *E. coli*

References in Table 1

1. Chen J, Griffiths MW. PCR differentiation of *Escherichia coli* from other Gram-negative bacteria using primers derived from the nucleotide sequences flanking the gene encoding the universal stress protein. Lett Appl Microbiol. 1998; 27: 369–71.

2. Clermont O, Bonacorsi S, Bingen E. Rapid and simple determination of the *Escherichia coli* phylogenetic group. Appl Environ Microbiol. 2000; 66: 4555–8.

3. Clermont O, Christenson JK, Denamur E, Gordon DM. The Clermont *Escherichia coli* phylo-typing method revisited: Improvement of specificity and detection of new phylo-groups. Environ Microbiol Rep. 2013; 5: 58–65.

4. Robins-Browne RM, Bordun A-M, Tauschek M, Bennett-Wood VR, Rusell J, Oppedisano F, et al. *Escherichia coli* and community-acquired gastroenteritis, Melbourne, Australia. Emerg Infect Dis. 2004; 10: 1797-1805.

5. Sethabutr O, Venkatesan M, Yam S, Pang LW, Smoak BL, Sang WK, et al. Detection of PCR product of the *ipaH* gene from *Shigella* and enteroinvasive *Escherichia coli* by enzyme-linked immunosorbent assay. Diagn Microbiol Infect Dis. 2000; 37: 11-16.

6. Spurbeck RR, Dinh PC Jr, Walk ST, Stapleton AE, Hooton TM, Nolan LK, et al. *Escherichia coli* isolates that carry *vat*, *fyuA*, *chuA*, and *yfcV* efficiently colonize the urinary tract. Infect Immun. 2012; 80: 4115–22.

Table S2. Sample collection dates and locations, and phylogenetic groupings of *E. coli* by triplex and quadruplex PCR assays.

Date of sample collection	Location (no. of confirmed *E. coli*)	Phylogenetic grouping by triplex PCR (no. of isolates)	Phylogenetic grouping by quadruplex PCR (no. of isolates)
14 May 2018	J[a] (1) Z[b] (2)	B1 (2) D (1)	B1 (2) D (1)
11 June 2018	J (7) Z (2) H[c] (5)	A (6) B1 (3) D (5)	A (6) B1 (3) D (4) E (1)
9 July 2018	J (1) Z (3) H (2)	A (3) B1 (1) B2 (1) D (1)	A (3) B1 (1) B2 (1) D (1)
13 August 2018	J (6) Z (6) H (7)	A (9) B1 (6) B2 (2) D (2)	A (9) B1 (6) B2 (2) D (1) E (1)
10 September 2018	J (2) Z (1) H (9)	A (8) B1 (3) B2 (1)	A (7) B1 (3) B2 (1) C (1)

8 October 2018	J (4) Z (1) H (4)	A (4) B1 (3) D (2)	A (4) B1 (3) D (1) F (1)
12 November 2018	J (4) Z (1) H (4)	A (7) B1 (2)	A (7) B1 (2)
10 December 2018	J (3) Z (4) H (3)	A (3) B1 (3) D (4)	A (3) B1 (3) D (2) F (2)
7 January 2019	J (4) Z (5) H (5)	A (3) B1 (3) B2 (2) D (6)	A (3) B1 (3) B2 (2) D (3) F (1) E (2)
4 February 2019	J (4) Z (2) H (2)	A (3) B1 (3) D (2)	A (3) B1 (3) F (2)
11 March 2019	J (3) Z (6) H (3)	A (6) B1 (1) B2 (4) D (1)	A (6) B1 (1) B2 (4) D (1)
8 April 2019	J (7) Z (7) H (10)	A (16) B1 (5) D (3)	A (14) B1 (5) D (1) F (2) C (2)

[a]Jabriya.

[b]Zahraa.

[c]Hateen.

Table S3. Distribution of ExPEC according to location and date of collection of samples.

Month/Year	No. of ExPEC isolates	Location		
		Jabriya	Zahraa	Hateen
14 May/2018	0	0	0	0
11 June/2018	0	0	0	0
9 July/2018	1	0	1	0
13 August/2018	2	0	0	2
10 September/2018	0	0	0	0
8 October/2018	1	1	0	0
12 November/2018	0	0	0	0
10 December/2018	0	0	0	0
7 January/2019	2	1	1	0
4 February/2019	3	0	1	2
11 March/2019	4	2	1	1
8 April/2019	1	1	0	0