

Table S1. Genes, primers, and annealing temperature used for amplification

Gene	Primer designation	Primer sequence (5' to 3')	Annealing temperature (°C)	Reference
16S rRNA ^a	9F (F24)	GAGTTTGATYMTGGCTAG	58	Harper CG,2003
	F16	TAGATAACCCYGGTAGTCC		Shen Z,2001
<i>clbA</i> ^a	IHAPJPN42 (clbAr)	CAGATACACAGATAACCATTCA	55	Johnson JR, 2008, Bubois D, 2010
	IHAPJPN46 (clbAf)	CTAGATTATCCGTGGCGATT		
<i>clbQ</i> ^a	IHAPJPN55 (clbQr)	TTATCCTGTTAGCTTCGTT	55	Johnson JR, 2008, Bubois D, 2010
	IHAPJPN56 (clbQf)	CTTGTATAGTTACACAACATTTC		
Multiplex <i>cnf</i>	CNF-s	TTATATAGACAAGATGGA	55	Toth I, 2003
	CNF-as	CATAAAGCTTACAATATTGA		
<i>cnf1</i>	CNF-1s	GGGGGAAGTACAGAAGAATTA	55	Toth I, 2003
	CNF-1as	TTGCCGTCCACTCTCACCACT		
<i>cnf2</i>	CNF-2s	TATCATACGGGCAGGAGGAAGCACC	55	Toth I, 2003
	CNF-2as	GTCACAATAGACAATAATTTCG		
multiplex <i>cdt</i>	cdt-s1	GAAAGTAAATGGAATATAATGTCCG	55	Toth I, 2003
	cdt-as1	AAATCACCAAGAACATCCAGTTA		
<i>yjaA</i>	YjaA.1	TGAAGTGTAGGAGACGCTG	55	Clermont D,2000
	YjaA.2	ATGGAGAATGCGTCTCAAC		
(DNA fragment)	TspE4C2.1	GAGTAATGTCGGGGCATTCA	55	Clermont D,2000
	TspE4C2.2	CGCGCCAACAAAGTATTAG		
<i>chuA</i>	ChuA.1	GACGAACCAACGGTCAGGAT	55	Clermont D,2000
	ChuA.2	TGCCGCCAGTACCAAGACA		
<i>svg</i>	svg.1	TCCGGCTGATTACAAACCAAC	55	Bidet P, 2007
	svg.2	CTGCACGAGGTTAGTCCTG		
<i>uidA</i>	P3	TATGAACTGTGCGTCACAGCC	55	Heininger A, 1999
	P4	CATCAGCACGTTATCGAATCC		

^a, Primers also used for sequencing;

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Table S2. Virulence Genes

Isolates	Gene	Product	Virulence Function/Comments	References
S1, S2	<i>astA</i>	Enterotoaggregative E. coli (EaggEC) Heat-Stable Enterotoxin 1 (EAST1)	Activates guanylyl cyclase resulting in ion secretion Possibly contributes to watery diarrheal disease	Croxen, M.A., 2010; Kaper, J.B., 2004; Turner, S.M., 2006
S1, S2, S4, S5	<i>celB</i>	Endonuclease Colicin E2	Bacteriocin – DNA nuclease that translocates into susceptible E. coli strains and causes cell death via DNA damage Hypothesized to provide competitive survival advantage colicin-expressing, pathogenic E. coli strains. Mostly identified in B2 or D phylogroup	Alonso, G., 2000; Duche, D., 2007; Gordon, D.M., 2006; Lechner, M., et al., 2016; Micenková, L., 2016
S1, S2, S3, S6, S9, S10	<i>cnf1</i>	Cytotoxic Necrotizing Factor 1	Glutamine deaminase that targets Rho family GTPases (RhoA, Cdc42 and Rac) leading to constitutive “on” activity that causes aberrant actin cytoskeletal alterations, G2/M cell cycle arrest with multi-nucleation and cellular enlargement, and anti-apoptotic/pro-survival signaling pathways Promotes persistence of epithelial colonization niches and invasion/dissemination of infection. May contribute toward carcinogenesis	Buc, E., 2013; Croxen, M.A., 2010; Flores-Mireles, A.L., 2015; Kaper, J.B., 2004
S1-S10	<i>gad</i>	Glutamate Decarboxylase	Promotes survival in acid environments by neutralizing cytoplasmic pH via decarboxylation of glutamate and export of γ-aminobutyric acid May allow survival through stomach for intestinal colonization during transmission of infection	Bergholz, T.M., 2007; Grant, M.A., 2001; Shin, S., 2001
S3, S6, S9, S10	<i>iha</i>	Iron-Regulated Gene Homologue Adhesin	Adhesion factor – adherence to epithelial cells to promote colonization of host Expression increased in iron depleted environments	Johnson, J.R., 2005; Rashid, R.A., 2006; Tarr, P.I., 2000
S1, S2, S4, S5	<i>iroN</i>	Enterobactin Siderophore Receptor Protein	Iron acquisition – transportation of iron-chelated siderophore into bacteria	Caza, M., 2013; Flores-Mireles, A.L., 2015; Kaper, J.B., 2004
S1-S10	<i>iss</i>	Increased Serum Survival/Bor Protein Precursor	Outer membrane protein – promotes resistance against killing by host complement proteins Possibly promote survival against innate immune system during systemic dissemination/infection	Johnson, T.J., 2008; Lynne, A.M., 2007; Mijajlovic, H., 2014
S7, S9	<i>lpfA</i>	Long Polar Fimbriae	Adhesion factor – adherence to intestinal epithelial cells to promote colonization of host	Jordan, D.M., 2004; Ross, B.N., et al., 2015; Torres, A.G., 2009
S1, S2, S4, S5	<i>mchB, mchC, mchF, mcmA</i>	Microcin H47 Synthesis and Transport	Bacteriocin - ribosomally-synthesized peptide antibiotic against susceptible bacterial species Hypothesized to provide competitive survival advantage microcin-expressing, pathogenic E. coli strains. Mostly identified in B2 or D phylogroup. Association between microcin H47 and higher number of virulence genes in genome	Abraham, S., 2012; Budic, M., 2011; Micenková, L., 2014.; Micenková, L., 2016; Lavina, M., 1990; Rodriguez, E., 1990
S4, S5	<i>pic</i>	Protease Involved in Intestinal Colonization	Serine Protease autotransporters of Enterobacteriaceae (SPATE) Mucinase activity may mediate degradation of mucus or mucin as a nutrient source and facilitate penetration/invasion through mucous barrier layer Protease activity may cause serum resistance possibly by degradation of complement proteins	Croxen, M.A., 2010; Dautin, N., 2010; Kaper, J.B., 2004; Welch, R.A., 2016
S1, S2, S4, S5	pks Gene Island (<i>clbA-clbR</i>)	Colibactin	Genotoxin that causes G2/M cell cycle arrest, megalocytosis, and DNA damage to cell lines in vitro Associated with colorectal cancer in human patients and potentiates carcinogenesis/tumor formation in mouse model	Arthur, J.C., 2012; Buc, E., 2013; García, A., 2016; Nougayrede, J.P., 2006
S1, S2, S4, S5	<i>vat</i>	Vacuolating Autotransporter Toxin	Serine Protease Autotransporters of Enterobacteriaceae (SPATE) Vacuolating activity causes intracellular vacuoles to cell lines in vitro	Guignot, J., A., 2015; Welch, R.A., 2016
S3, S6, S9, S10	<i>sat</i>	Secreted Autotransporter Toxin	Serine Protease Autotransporters of Enterobacteriaceae (SPATE) Cytotoxicity characterized by vacuolating activity, elongated morphology, plate detachment, and death to cell lines in vitro May degrade tight junctions between intestinal epithelial cells to promote invasion	Dautin, N., 2010; Welch, R.A., 2016

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