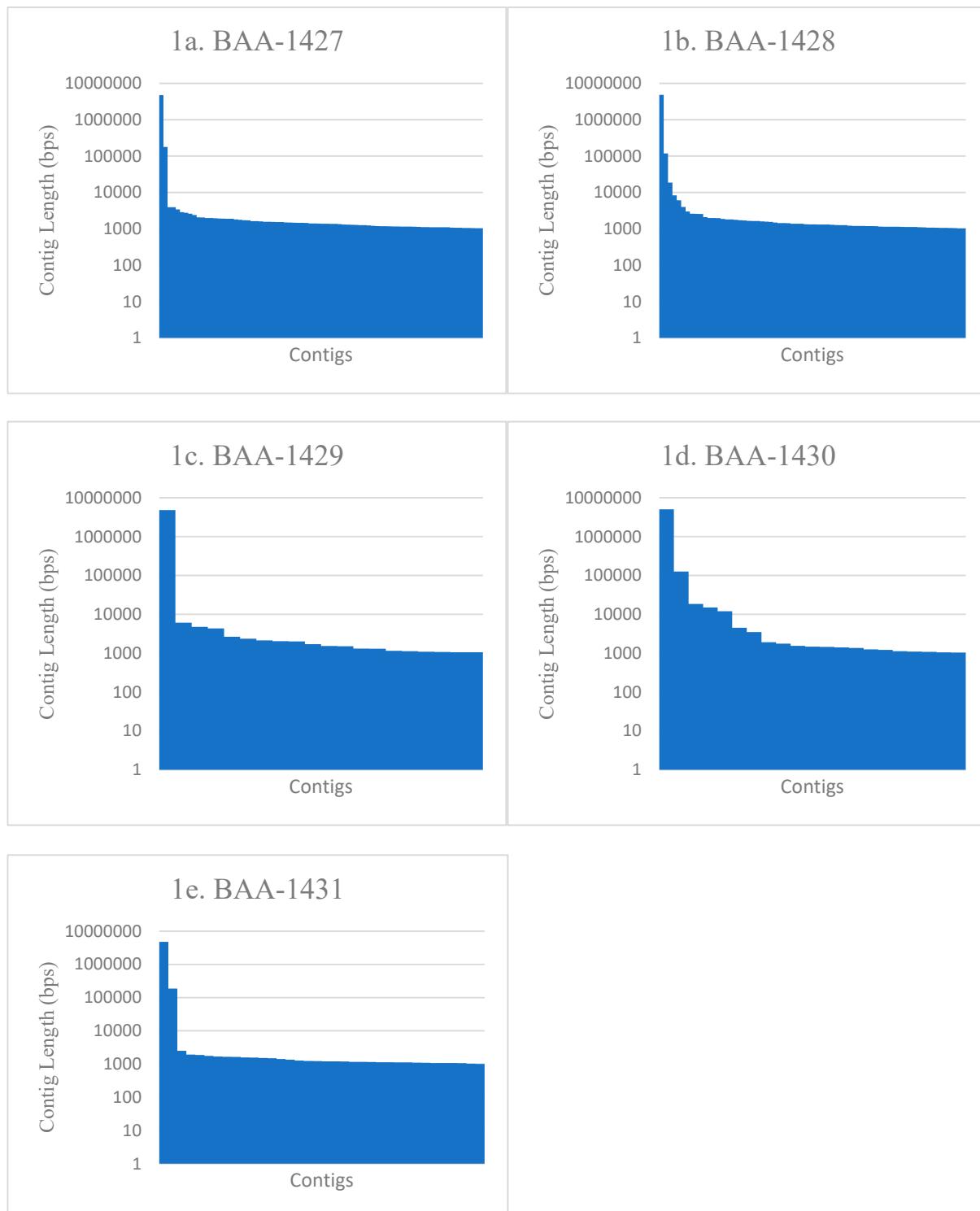
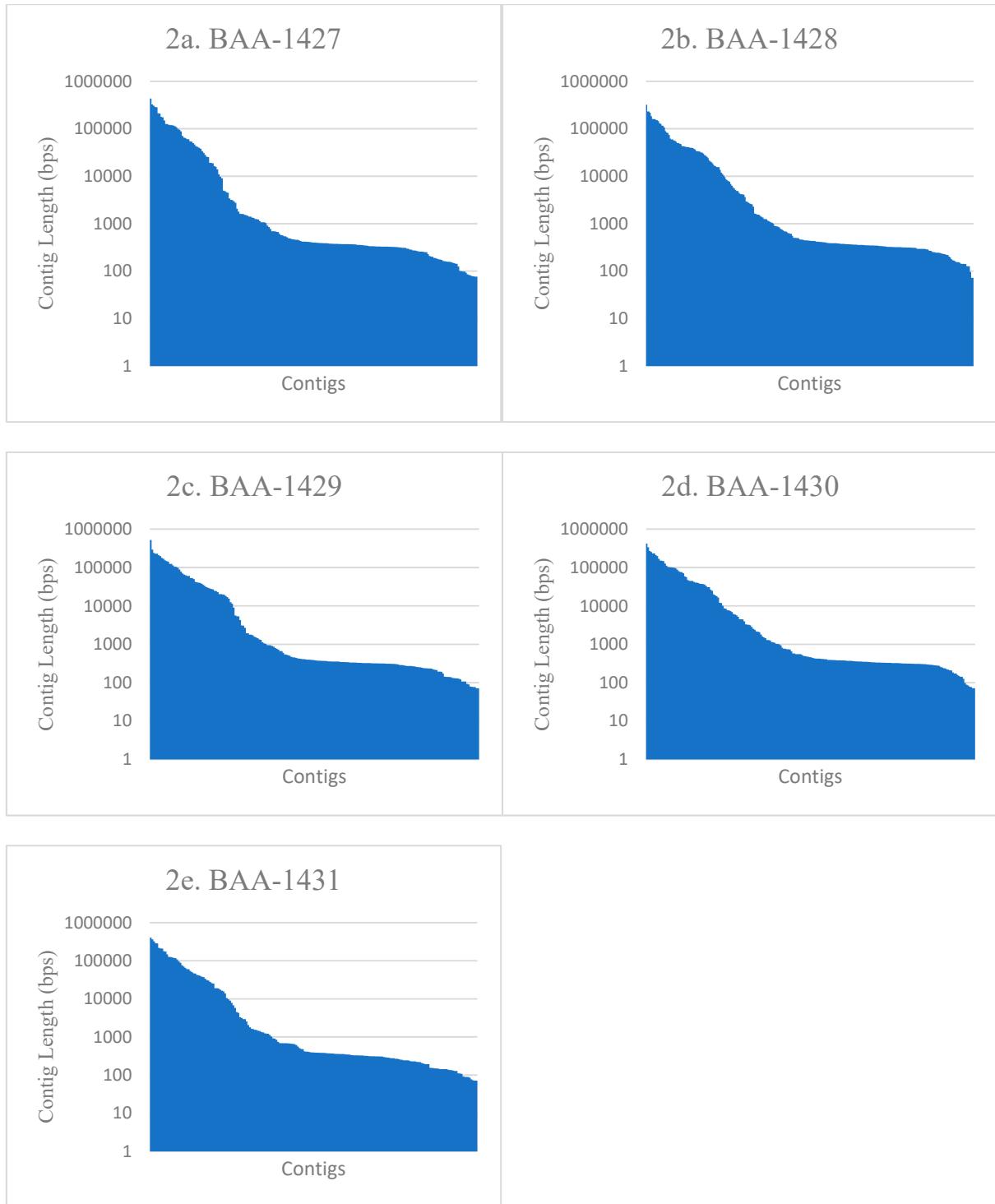


**Figure S1.** MinION Assemblies: Distribution of Contig Lengths. Depicts the size distribution of contigs based on their overall assembled lengths (bps) for each bacterial genomic assembly that was generated via the MinION assembler.



**Figure S2.** Miseq Assemblies: Distribution of Contig Lengths. Depicts the size distribution of contigs based on their overall assembled lengths (bps) for each bacterial genomic assembly that was generated via the MiSeq assembler.



**Table S1.** List of virulence attributes examined.

Virulence Factor <sup>1</sup>	Related Gene	Pathogroup <sup>2</sup>	Pathogroup	NCBI Accession Number <sup>3</sup>
Afa/Dr family	<i>afaE-I, afaE-III, daaE, draE, draE2</i>	DAEC	Adherence	CAW30801, CAA54121, AAA23661, AAK16480, AAB65153
AAF's	<i>aafA, aafB, aafC, aafD, agg3A, agg3B, agg3C, agg3D, aggA, aggB, aggC, aggD, aggR</i>	EAEC	Adherence	AAB82330, AAD27809, AAD27810, AAD26595, AAM88298, AAM88297, AAM88296, AAM88295, AAA57454, AAA57453, AAA57452, AAA57451, CAA83535
Dispersin	<i>aap/aspU, aatA, aatB, aatC, aatD, aatP</i>	EAEC	Adherence	YP_006099176, YP_006099144, YP_006099145, YP_006099146, YP_006099147, YP_006099143
EAST-1	<i>astA</i>	EAEC	Toxin	BAA94855
Pet	<i>pet</i>	EAEC	Toxin	YP_006099165
Pic	<i>pic</i>	EAEC		YP_006098864
ShET1	<i>set1A, set1B</i>	EAEC	Toxin	YP_006098866, YP_006098865
ECP	<i>yagV/ecpE, yagW/ecpD, yagX/ecpC, yagY/ecpB, yagZ/ecpA, ykgK/ecpR</i>	EHEC	Adherence	NP_286006, NP_286007, NP_286008, NP_286009, NP_286010, NP_286011
Efa-1/LifA	<i>efa1</i>	EHEC	Adherence	AAD49229
Intimin	<i>eae</i>	EHEC	Adherence	NP_290259
Paa	<i>paa</i>	EHEC	Adherence	NP_287515
ToxB	<i>toxB</i>	EHEC	Adherence	YP_325655

**Table S1 Continued**

Chu	<i>chuA, chuS, chuT, chuU, chuW,</i> <i>chuX, chuY</i>	EHEC	Iron Uptake	NP_290080, NP_290079, NP_290082, NP_290086, NP_290083, NP_290084, NP_290085
EspP	<i>espP</i>	EHEC	Protease	YP_325580
StcE	<i>stcE</i>	EHEC	Protease	YP_325591
Ler	<i>ler</i>	EHEC	Regulation	NP_290288
Hemolysin	<i>hlyA, hlyB, hlyC, hlyD</i>	EHEC	Toxin	YP_325608, YP_325609, YP_325607, YP_325610
Stx (Shiga toxin)	<i>stx1A, stx1B, stx2A, stx2B</i>	EHEC	Toxin	NP_288673, NP_288672, NP_286976, NP_286977
Cif	<i>cif</i>	EHEC	Type III translocate protein	AAN07916
EspA, EspB, EspD,	<i>espA, espB, espD, espF, espG,</i>	EHEC	Type III translocate protein	NP_290256, NP_290254, NP_290255, NP_290250, NP_290289, NP_290264
EspF, EspG, EspH	<i>espH</i>			
Map	<i>map</i>	EHEC	Type III translocate protein	NP_290262
NleA/EspI, NleC, NleD	<i>nleA/espI, nleC, nleD</i>	EHEC	Type III translocate protein	NP_287961, NP_286533, NP_286535
Tir	<i>tir</i>	EHEC	Type III translocate protein	NP_290261
IcsA (VirG)	<i>icsA/virG</i>	EIEC	Actin-based Mobility	NP_858315
LPS	<i>gtrA, gtrB, gtrII</i>	EIEC		NP_706257, NP_706258, NP_706259
Aerobactin	<i>iucA, iucB, iucC, iucD, iutA</i>	EIEC	Iron Uptake	NP_709454, NP_709455, NP_709456, NP_709457, NP_709458
Shu	<i>shuA, shuS, shuT, shuU, shuV,</i> <i>shuX, shuY</i>	EIEC	Iron Uptake	YP_405019, YP_405020, YP_405017, YP_405014, YP_405013, YP_405016, YP_405015
IcsP (SopA)	<i>icsP/sopA</i>	EIEC	Protease	NP_858404
Pic	<i>pic</i>	EIEC	Protease	NP_708747

**Table S1 Continued**

SigA	<i>sigA</i>	EIEC	Protease	NP_708742
T2SS ( <i>S. dysenteriae</i> )	<i>gspC, gspD, gspE, gspF, gspG,</i> <i>gspH, gspI, gspJ, gspK, gspL,</i> <i>gspM</i>	EIEC	Secretion System	YP_404599, YP_404600, YP_404601, YP_404602, YP_404603, YP_404604, YP_404605, YP_404606, YP_404607, YP_404608, YP_404609
shET1, shET2	<i>set1A, set1B, senB</i>	EIEC	Toxin	YP_177614, YP_177613, YP_406304
Stx ( <i>S. dysenteriae</i> (serotype 1))	<i>stxA, stxB</i>	EIEC	Toxin	YP_403025, YP_403026
BFP	<i>bfpA, bfpB, bfpC, bfpD, bfpE,</i> <i>bfpF, bfpG, bfpH, bfpI, bfpJ,</i> <i>bfpK, bfpL, bfpP, bfpU</i>	EPEC	Adherence	BAA84838, YP_002332159, YP_002332160, BAA84843, YP_002332163, BAA84845, YP_002332158, BAA84847, BAA84848, YP_002332168, BAA84850, YP_002332170, YP_002332165, BAA84842
Intimin	<i>eae</i>	EPEC	Adherence	AAC38392
Lymphostatin/ LifA	<i>lifA/efa1</i>	EPEC	Adherence	YP_002330705
Paa	<i>paa</i>	EPEC	Adherence	YP_006158657
EspC	<i>espC</i>	EPEC	Protease	AAG37043
Ler	<i>ler</i>	EPEC	Regulation	AAC38364
Per	<i>perA/bfpT, perB/bfpV,</i> <i>perC/bfpW</i>	EPEC	Regulation	BAA84859, BAA84860, BAA84861
CDT	<i>cdtA, cdtB, cdtC</i>	EPEC	Toxin	CAD48849, CAD48850, CAD48851
EAST1	<i>east1</i>	EPEC	Toxin	AHY03744
Cif	<i>cif</i>	EPEC	Type III translocate protein	PRJEA32571

**Table S1 Continued**

EspA, EspB, EspD,	<i>espA, espB, espD, espF, espG,</i>	EPEC	Type III translocate protein	AAC38394, AAC38396, AAC38395, AAC38400, AAC38363, AAC38387
EspF, EspG, EspH	<i>espH</i>			
Map	<i>orf19</i>	EPEC	Type III translocate protein	AAC38389
NleA/EspI, NleC, NleD	<i>nleA, nleC, nleD</i>	EPEC	Type III translocate protein	EIQ72111, YP_003498583, YP_003498587
Tir	<i>tir</i>	EPEC	Type III translocate protein	AAC38390
Adhesive fimbriae	<i>cfaB, cofA, cooA, cs3, csbA,</i> <i>cseA, csfA, csnA, cssA, csvA,</i> <i>cswA</i>	ETEC	Adherence	AAC41415, BAB62897, AAT07441, AAA23614, AAS89777, AAD30557, CAA11820, AAL31637, AAC45093, AAK09045, AAK09047
EtpA	<i>etpA, etpB</i>	ETEC	Adherence	YP_006203830, CBJ04459
Heat-labile toxin (LT)	<i>eltA, eltB</i>	ETEC	Toxin	AAA24685, AAA98064
Heat-stable toxin (ST)	<i>estIa</i>	ETEC	Toxin	YP_003294006
FdeC	<i>fdeC</i>	NMEC	Adherence	YP_002390132
S fimbriae	<i>sfaA, sfaB, sfaC, sfaD, sfaE,</i> <i>sfaF, sfaG, sfaH, sfaS</i>	NMEC	Adherence	YP_006100306, YP_006100305, YP_006100304, YP_006100307, YP_006100308, YP_006100309, YP_006100310, YP_006100312, YP_006100311
AslA	<i>aslA</i>	NMEC	Invasion	AAG10151
Ibes	<i>ibeA, ibeB, ibeC</i>	NMEC	Invasion	AAF98391, AAD30205, AAD28716
K1 capsule	<i>kpsD, kpsM, kpsT</i>	NMEC	Invasion	AAA21682, AAA24046, AAA24047
OmpA	<i>ompA</i>	NMEC	Invasion	AAF37887
TraJ	<i>traJ</i>	NMEC	Invasion	AAA92657

**Table S1 Continued**

CNF-1	<i>cnf1</i>	NMEC	Toxin	CAA50007
Dr adhesins	<i>draA, drab, draC, draD, draE, draP</i>	UPEC	Adherence	AAK16475, AAK16476, AAK16477, AAK16478, AAK16480, AAK16479
F1C fimbriae	<i>focA, focC, focD, focF, focG, focH focI</i>	UPEC	Adherence	NP_753153, NP_753155, NP_753156, NP_753157, NP_753158, NP_753159, NP_753154
P fimbriae	<i>papA, papA, papB, papB, papC, papC, papD, papD, papE, papE, papF, papF, papG, papG, papH, papH, papI, papJ, papJ, papK, papK</i>	UPEC	Adherence	NP_755467, NP_757036, -, -, NP_755465, NP_757034, NP_755464, NP_757033, NP_755460, NP_757029, NP_755459, NP_757028, NP_755458, NP_757027, NP_755466, NP_757035, NP_755468, NP_757037, NP_755463, NP_757032, NP_755461, NP_757030
S fimbriae	<i>sfaA, sfaB, sfaC, sfaD, sfaE, sfaF, sfaG, sfaH, sfaS, sfaX, sfaY</i>	UPEC	Adherence	YP_540124, YP_540123, YP_540122, YP_540125, YP_540126, YP_540127, YP_540128, YP_540130, YP_540129, YP_540132, YP_540131
Type 1 fimbriae	<i>fimA, fimB, fimC, fimD, fimE, fimF, fimG, fimH, fimI</i>	UPEC	Adherence	NP_757241, NP_757239, NP_757243, NP_757244, NP_757240, NP_757245, NP_757247, NP_757248, NP_757242
TcpC	<i>tcpC</i>	UPEC	Immune Evasion	NP_754290
Aerobactin	<i>iucA, iucB, iucC, iucD, iutA</i>	UPEC	Iron Uptake	NP_755502, NP_755501, NP_755500, NP_755499, NP_755498
Chu	<i>chuA, chuS, chuT, chuU, chuV, chuW, chuX, chuY</i>	UPEC	Iron Uptake	NP_756170, NP_756169, NP_756175, NP_756179, NP_756180, NP_756176, NP_756177, NP_756178

**Table S1 Continued**

Enterobactin	<i>entA, entB, entC, entD, entE, entF, fepA, fepB, fepC, fepD, fepE, fepG</i>	UPEC	Iron Uptake	NP_752614, NP_752613, NP_752611, NP_752599, NP_752612, NP_752604, NP_752600, NP_752610, NP_752606, NP_752608, NP_752605, NP_752607
IroN	<i>iroN</i>	UPEC	Iron Uptake	NP_753164
Pic	<i>pic</i>	UPEC	Protease	NP_752289
Sat	<i>sat</i>	UPEC	Protease	NP_755494
Tsh	<i>vat</i>	UPEC	Protease	NP_752330
CNF-1	<i>cnf1</i>	UPEC	Toxin	YP_543855
Hemolysin	<i>hlyA, hlyB, hlyC hlyD</i>	UPEC	Toxin	NP_755445, NP_755448, NP_755444, NP_755449

<sup>1</sup> Table was constructed from pre-existing *E. coli* data located on the VFDB [52-55]

<sup>2</sup> Diffusely adherent *E. coli* (DAEC), Enteropathogenic *E. coli* (EAEC), Enterohemorrhagic *E. coli* (EHEC), Enteroinvasive *E. coli* (EIEC), Enteropathogenic *E. coli* (EPEC), Enterotoxigenic *E. coli* (ETEC), Neonatal meningitis-associate *E. coli* (NMEC), Uropathogenic *E. coli* (UPEC).

<sup>3</sup> NCBI accession numbers are listed in the same order as the related gene they are associated with. [52-55]

**Table S2.** Comprehensive list of alterations within the *rpoB* genes of each sample and assembly type for each virulence factor screened.

Assembly	Sample	Codon <sup>1</sup>	Mutation	Alteration	Amino Acid Shift
MiSEQ	1427	206*	Transition	GCG → GCA	A to A
	1428	486*	Transition	ACC → ACT	T to T
	1430	489*	Transversion	CCA → CCT	P to P
	1430	623*	Transition	TTG → CTG	L to L
	1430	846*	Transversion	GGT → GGG	G to G
MINION	1427	9	Deletion	AAA → AA-	-
	1427	115	Deletion	AAA → AA-	-
	1427	206*	Transition	GCG → GCA	A to A
	1427	265	Deletion	AAA → AA-	-
	1427	319	Deletion	CTG → C-G	-
	1427	431	Deletion	AAG → A-G	-
	1427	468	Transition	CTG → CCG	L to P
	1427	618	Deletion	CAG → C-G	-
	1427	643	Deletion	CTG → C-G	-
	1427	671	Transition	CTG → CCG	L to P
	1427	941	Deletion	AAA → AA-	-
	1427	1117	Deletion	CCT → C-	-
	1427	1238	Deletion	CTG → --G	-
	1428	8	Deletion	AAA → AA-	-

**Table S2 Continued**

1428	83	Deletion	CAG → C-G	-
1428	114	Deletion	GTA → GT-	-
1428	226	Deletion	GAA → G-A	-
1428	264	Deletion	GAA → G-A	-
1428	278	Deletion	GAA → G-A	-
1428	319	Deletion	CTG → C-G	-
1428	372	Deletion	CCT → CC-	-
1428	468	Deletion	CTG → C-G	-
1428	486*	Transition	ACC → ACT	A to A
1428	563	Deletion	ACC → A-C	-
1428	618	Transition	CAG → CGG	Q to R
1428	643	Deletion	CTG → C-G	-
1428	649	Transition	CAG → CGG	Q to R
1428	671	Deletion	CTG → C-G	-
1428	940	Deletion	GAA → G-A	-
1428	987	Deletion	GAG → GA-	-
1428	1038	Transition	CAG → CGG	Q to R
1428	1062	Transition	CCT → CCC	R to P
1428	1117	Deletion	CTG → C-G	-
1428	1238	Deletion	CTG → --G	-

**Table S2 Continued**

1430	4	Transition	TCC → CTC	S to L
1430	8	Deletion	AAA → -AA	-
1430	83	Deletion	CAG → C--	-
1430	112	Transition	GGC → AGC	G to S
1430	113	Deletion	AAC → AA-	-
1430	114	Deletion	GGT → --T	-
1430	115	Deletion	ACC → -CC	-
1430	183	Transition	TGG → CGG	W to R
1430	227	Deletion	AAA → AA-	-
1430	265	Deletion	AAA → AA-	-
1430	319	Deletion	CTG → C-G	-
1430	372	Deletion	CCT → C-G	-
1430	431	Deletion	AAG → A-G	-
1430	468	Transition	CTG → CCG	L to P
1430	489*	Transversion	CCA → CCT	P to P
1430	535	Deletion	CCA → CC-	-
1430	618	Transition	CAG → CGG	Q to R
1430	623*	Transition	TTG → CCG	L to P
1430	633	Deletion	CTG → C-G	-
1430	846*	Transversion	GGT → GGG	G to G

**Table S2 Continued**

	1430	1038	Deletion	CAG → C-G	-
	1430	1062	Transition	CCT → CCC	P to P
	1430	1117	Deletion	CTG → C--	-
	1430	1238	Deletion	CTG → C-G	-
	1430	1253	Transition	CTG → CCG	L to P
Hybrid	1427	206*	Transition	GCG → GCA	A to A
	1428	486*	Transition	ACC → ACT	T to T
	1430	489*	Transversion	CCA → CCT	P to P
	1430	623*	Transition	TTG → CTG	L to L
	1430	846*	Transversion	GGT → GGG	G to G
Rif-Mutant	1427	206*	Transition	GCG → GCA	A to A
	1427	533 <sup>R</sup>	Transition	CTC → CCC	L to P
	1428	486*	Transition	ACC → ACT	T to T
	1428	512 <sup>R</sup>	Transition	TCT → CCT	S to P
	1430	489*	Transversion	CCA → CCT	P to P
	1430	526 <sup>R</sup>	Transition	CAC → TAC	H to Y
	1430	623*	Transition	TTG → CTG	L to L
	1430	846*	Transversion	GGT → GGG	G to G

<sup>1</sup>Codon number within the rpoB gene of the *E. coli* str. K-12 substr. MG1655 (NC\_000913.3) reference strain.

\* Indicates that this mutation was found within all four assemblies.

<sup>R</sup>Documented to confer Rif-resistance.