

Supplementary Materials: Application and Optimization of *relE* as a Negative Selection Marker for Making Definitive Genetic Constructs in Uropathogenic *Escherichia coli*

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Table S1. Strains and plasmids used in this study.

Strain	Genotype	Source/Reference
SLC-6	Wild type UTI89	Chen <i>et al.</i> , 2006 [1]
SLC2-33-1	UTI89 <i>fimH::fimH^{UTI89}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-46-1	UTI89 <i>fimH::fimH^{A62S}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-51-1-1	UTI89 <i>fimH::fimH^{A27V/V163A}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-35-1	UTI89 <i>fimH::fimH^{Q133K}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-68-4-1	UTI89 <i>fimH::fimH^{A27V/A62S/S70N/N78S}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-39-1	UTI89 <i>fimH::fimH^{A27V}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-45-4	UTI89 <i>fimH::fimH^{V163A}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC2-89-3	UTI89 <i>fimH::fimH^{A27V/A62S/V163A}</i> -kan	Chen <i>et al.</i> , 2009 [2]
SLC-502	UTI89 <i>fimH::kan-P_{rhaB}-relE</i>	Khetrpal <i>et al.</i> , 2015 [3]
SLC-463	UTI89 <i>fimH::fimH^{UTI89}</i>	Khetrpal <i>et al.</i> , 2015 [3]
SLC-464	UTI89 <i>fimH::fimH^{A62S}</i>	Khetrpal <i>et al.</i> , 2015 [3]
SLC-465	UTI89 <i>fimH::fimH^{A27V/V163A}</i>	Khetrpal <i>et al.</i> , 2015 [3]
SLC-478	UTI89 <i>fimH::fimH^{Q133K}</i>	Khetrpal <i>et al.</i> , 2015 [3]
SLC-479	UTI89 <i>fimH::fimH^{A27V/A62S/S70N/N78S}</i>	This study
SLC-482	UTI89 <i>fimH::fimH^{A27V}</i>	This study
SLC-483	UTI89 <i>fimH::fimH^{V163A}</i>	This study
SLC-484	UTI89 <i>fimH::fimH^{A27V/A62S/V163A}</i>	This study
SLC-516	UTI89 <i>ompC::kan-P_{rhaB}-relE</i>	Khetrpal <i>et al.</i> , 2015 [3]
Plasmid	Genotype	Source/Reference
pSLC-217	kan-P _{rhaB} - <i>relE</i>	Khetrpal <i>et al.</i> , 2015 [3]
pKM208		Murphy and Campellone, 2003 [4]

Table S2. FimH mutations and HA titres in UTI89.

Genotype	Hemagglutination Assay Titres	
	Using Positive Selection (Residual Kanamycin Marker)	Using Negative Selection (No Selection Marker)
Wild type UTI89	8	8
UTI89 <i>fimH::fimH^{UTI89}</i>	8	8
UTI89 <i>fimH::fimH^{A62S}</i>	5	5
UTI89 <i>fimH::fimH^{A27V/V163A}</i>	7	7
UTI89 <i>fimH::fimH^{Q133K}</i>	4	4
UTI89 <i>fimH::fimH^{A27V/A62S/S70N/N78S}</i>	8	8
UTI89 <i>fimH::fimH^{A27V}</i>	8	8
UTI89 <i>fimH::fimH^{V163A}</i>	8	8
UTI89 <i>fimH::fimH^{A27V/A62S/V163A}</i>	8	8

Table S3. Primers used in this study.

No	Sequence	Notes
1	AGTGATTAGCATCACCTATACCTACAGCTGAACCCAAAGAGATGAT TGTAGTGTAGGCTGGAGCTGCTTC	<i>fimH</i> knockout Forward
2	TAGCTTCAGGTAATATTGCGTACCTGCATTAGCAATGCCCTGTGATT TCTCATATGAATATCCTCCTTAG	<i>fimH</i> knockout Reverse
3	ATGATGTTGCGCTTGAGTTG	UTI89+4913234 <i>fimH</i>
4	ATCTGGCCTACAAAGGGCTAA	UTI89-4914539 <i>fimH</i>
5	ATACCCCTTAATGCCCCATC	UTI89+4912861 test <i>fimH</i> KO
6	ACCGCGCAAAACATCCAGTT	UTI89+4913338 test <i>fimH</i> KO
7	GGTATTCGGCATTGGCCTGA	UTI89-4914820 test <i>fimH</i> KO
8	GCTTGCAGTGGGCTTACATG	P1 forward for amplification of dual selection cassette with kanamycin without FRT sites (from template plasmid pSLC-217 and related plasmids (Khetrapal <i>et al.</i> , 2015 [3]))
9	TCAGAGCAGGATCGACGTCC	P2 reverse for amplification of dual selection cassette with kanamycin without FRT sites (from template plasmid pSLC-217 and related plasmids (Khetrapal <i>et al.</i> , 2015 [3]))
10	CTTCATTTAAATGGCGCGCC	P3 forward for amplification of dual selection cassette with chloramphenicol without FRT sites (from template plasmid pSLC-242 and related plasmids (Khetrapal <i>et al.</i> , 2015 [3]))
11	CATATGAATATCCTCCTTAG	Datsenko and Wanner original primer at priming site 2 (Datsenko and Wanner 2000 [5]). For amplification of dual selection cassette with chloramphenicol without FRT sites (from template plasmid pSLC-242 and related plasmids (Khetrapal <i>et al.</i> , 2015 [3]))

References

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