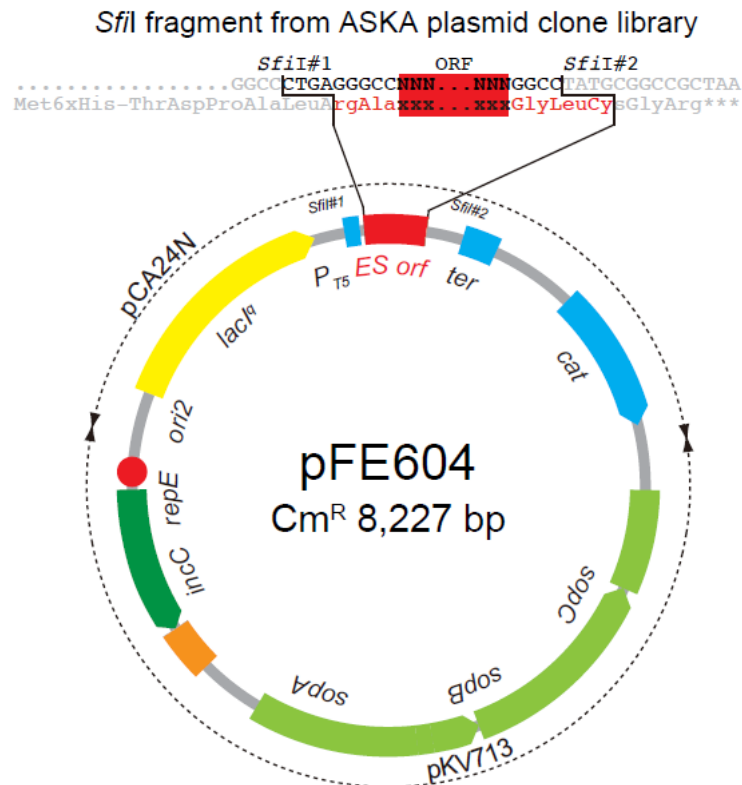


genes	terminus	Primer sequence
glnS	N-ter	5'-TAAGATCATA CGCCGTTATACGTTGTTTACGCTTTGAGGAATCCACGATG <b><i>ATTCCGGGGATCCGTC</i></b> <b><i>GACC</i></b>
	C-ter	5'-CGCCCCATCCGGCATAGCGAAACTTAAAATTACTCGCCTACTTTTCGCCCA <b><i>TGTAGGCTGGAGCTG</i></b> <b><i>CTTCG</i></b>
serS	N-ter	5'-AGGCTGTGGCCACATCTCCCATTTAATTCGATAAGCACAGGATAAGCATG <b><i>ATTCCGGGGATCCGTC</i></b> <b><i>GACC</i></b>
	C-ter	5'-GCGCTTTT TAGATT CAGAAAAATTGGGTATTAGCCAATATATTCCAGTCC <b><i>TGTAGGCTGGAGCTGC</i></b> <b><i>TTCG</i></b>

**Table S1. Primer sequences used for strain construction**

Sequences in (blue in bold and italic) are the sequences for pKD13. The rest of sequences are for the target genes' N and C-terminus regions for homologous recombination.



**Figure S1. Construction of essential gene complementing plasmid.**

pFE604 is a derivative of F plasmid almost the same as pFE604T as previously shown [1]. The essential genes' fragments were prepared by SfiI cut of the corresponding ASKA plasmid clones [2]. pCA24N is the vector of the ASKA plasmid clone library. pKV713 is the rreplication and partition responsible framgnet of miniF plasmid [3]

### References

1. Yong HT, Yamamoto N, Takeuchi R, Hsieh YJ, Conrad TM, Datsenko KA, Nakayashiki T, Wanner BL, Mori H: **Development of a system for discovery of genetic interactions for essential genes in Escherichia coli K-12.** *Genes Genet Syst* 2013, **88**(4):233-240. PMID: 24463526
2. Kitagawa M, Ara T, Arifuzzaman M, Ioka-Nakamichi T, Inamoto E, Toyonaga H, Mori H: Complete set of ORF clones of Escherichia coli ASKA library (a complete set of E. coli K-12 ORF archive): unique resources for biological research. *DNA Res* 2005, **12**(5):291-299, PMID: 16769691

3. Kawasaki Y, Wada C, Yura T: Roles of Escherichia coli heat shock proteins DnaK, DnaJ and GrpE in mini-F plasmid replication. *Mol Gen Genet* 1990, 220(2):277-282. PMID: 2183004)