

## Supplementary Information

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### Figure legends

#### **Supplementary Figure 1 Mini-F plasmid vectors.**

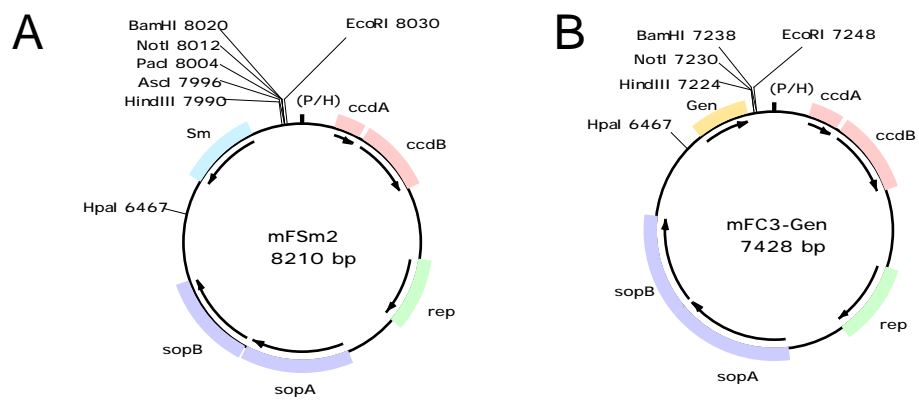
A schematic drawing of the mini-F plasmid vectors, mFSm2 and mFC3-Gen. *ccdA*, *ccdB*, *rep*, *sopA*, and *sopB* are necessary for the stable maintenance of the mini-F replicon. Sm and Gen indicate Streptomycin- and Gentamycin-resistance genes, respectively.

#### **Supplementary Figure 2 Construction of the complementing plasmids.**

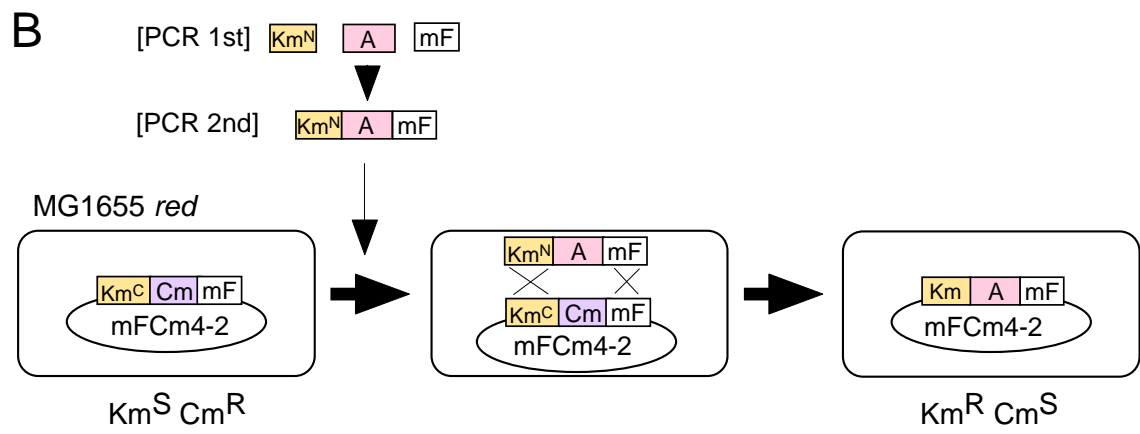
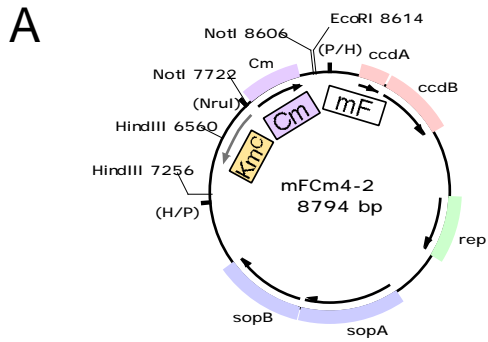
A schematic drawing of the mini-F plasmid vector, mFCm4-2 (A), and the *in vivo* cloning by *red* recombination used to construct the complementing plasmids (B). KmN and KmC represent the N- and C-termini of the Km<sup>R</sup> gene, respectively. The central part is contained in both KmN and KmC. The mFCm4-2 plasmid carries only the C-terminus of the Km<sup>R</sup> gene, resulting in Km<sup>S</sup>. The chromosome region to be cloned, A, KmN, and mF, which is a part of a mini-F plasmid, were amplified by a first and second PCR using three amplified DNA fragments as templates, and the KmN-A-mF fragment was prepared. This fragment was introduced into the MG1655 *red* strain, and Km<sup>R</sup> Cm<sup>S</sup> recombinants were selected, since recombination between KmN and KmC results in an intact Km<sup>R</sup> gene and recombination between the mF regions of the DNA fragment and plasmid removes the Cm<sup>R</sup> gene.

#### **Supplementary Figure 3 DNA fragments used to construct deletions.**

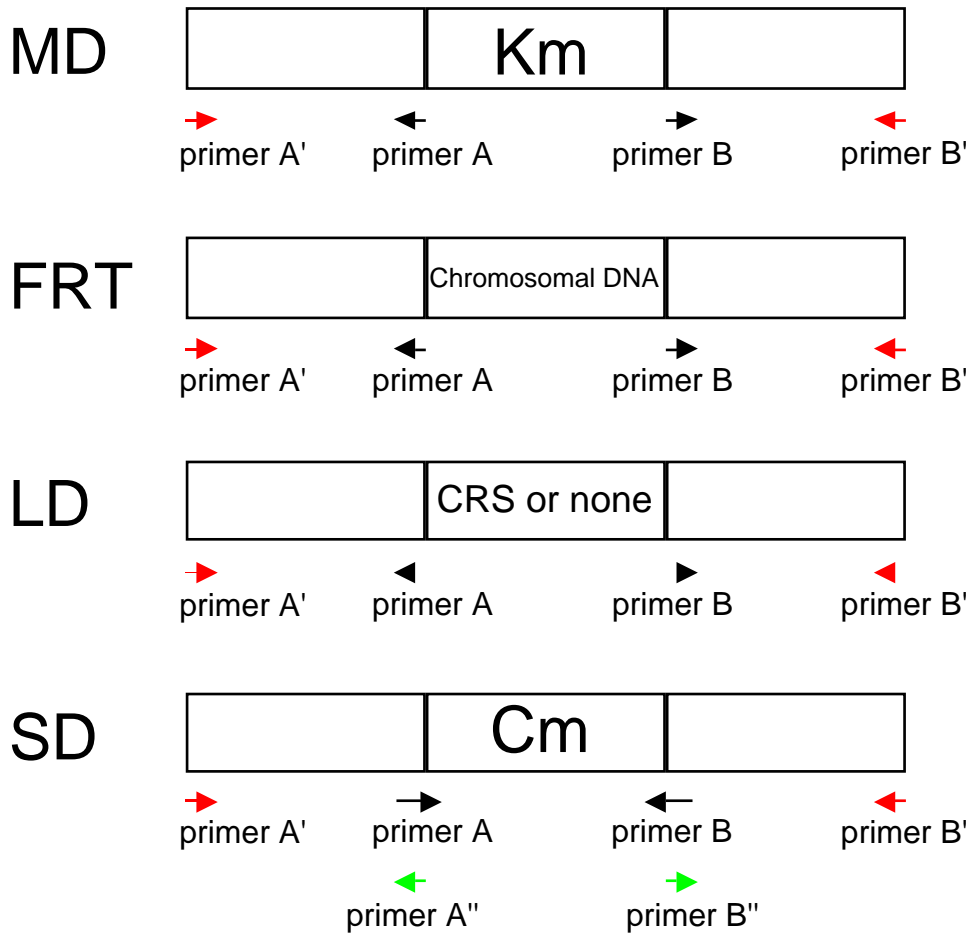
The DNA fragments and primers used to construct the deletion mutations are shown. White boxes represent DNA fragments containing chromosomal sequences flanking the region to be deleted. Primers A, A', A'', B, B', and B'' are listed in Supplementary Table I.



Supplementary Figure 1 Kato and Hashimoto



Supplementary Figure 2 Kato and Hashimoto



Supplementary Figure 3 Kato and Hashimoto