



Figure S4. First round of target genomic editing by CRISPR-Cas9 system. (A) Schematic of homologous recombination (HR) of *cre-1*, *res-1* and *gh1-1* mediated by Cas9, sgRNAs and donor DNA. (B) PCR analysis of triple-gene deletion of *cre-1*, *res-1* and *gh1-1* in selected transformants using one primer (*cre1/res1/gh1-1-out-F*) located upstream of the 5' flanking region of genomic DNA and the other primer (*cre1/res1/gh1-1-in-R*) located in the 3' flanking region of genomic DNA. The expected lengths of disrupted transformants of *cre-1*, *res-1* and *gh1-1* were 0.8, 0.7 and 1.9 kb, respectively, while those of WT strain (rightmost lane) was 1.2, 0.9 and 1.0 kb, respectively. Heterokaryotic transformants showed two PCR bands (both of wild-type and knockout). The symbol of star indicated deletion mutant. HDR, homology-directed repair; WT, wild type.