

Figure S3. T7EI assays and sequencing results at the *susd4* target. (A) *Bst*CI digestion revealed mutagenesis rates (loss of *Bst*CI site) represented by the upper uncut DNA bands of 72% and 19%, respectively, at two target sites of *susd4* gene. (B) Sequence analysis of the *susd4* gRNA-induced indel mutations in the target site. Blue color represents target sequence of *susd4-*1 or *susd4-*2; red color indicates PAM region; "-" indicates deletion; green color indicates mutation. The number in brackets represents the times of the identified mutant allele. In knock-in experiment, 16 F0 embryos were randomly selected and 20 clones were sequenced at each F0 embryos. 2 or 3 clones have the correct point mutation at the 3 positive F0 embryos.