

**Figure S2.** T7 endonuclease I assays and sequencing results of different Cas9/gRNAs injected F<sub>0</sub> embryos. (A-D) T7EI assays of different Cas9/gRNAs injected F<sub>0</sub> embryos. 6 embryos were picked up randomly from each group. (E) Sequences of Cas9-induced mutations in the *EGFP-*, *urod-*, *mib-* targeted founder embryos. 16 samples were sequenced in each group. Blue color represents target sequence; red color indicates PAM region; "-" indicates deletion; green color indicates mutations. The number in brackets represents the times of the identified mutant allele.