

OsGTL1 promoter editing using CRISPR/Cas9 in rice Oryza sativa L.

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Abstract: *GT2-LIKE1* (*GTL1*) gene is a negative regulator of stomatal development. It regulates stomatal number in plants. The CRISPR/Cas9 system has been used to modify the *OsGTL1* promoter. This research aims to screen *Cas9*-free rice with *OsGTL1* promoter modification. *Cas9* specific primers were designed for *Cas9* screening in all tillers of 8 T₃ rice lines. Only a single T₃ line is the Cas9-free in every tiller, while 3 out of 8 lines had Cas9 in all tillers. Seeds from Cas9-free tillers could be obtained from 5 independent lines. No significant difference in leaf greenness, tiller number per plant and leaf number per plant among the modified plants and wild type (WT). However, 7 out of 8 modified lines were the significantly smaller than WT. The nucleotide sequences of *OsGTL1* promoter in some Cas9-free plants revealed the modification in *OsGTL1* promoter, which included small deletion, insertion, and big deletion in the target region.

Graphical abstract:



Keywords: CRISPR/Cas9; OsGTL1; Oryza sativa L.

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