

Revolutionizing plant breeding with CRISPR-Cas genome editing tools

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Clustered regularly interspaced short palindromic repeats (CRISPR)-Cas9 and Cas12a (CRISPR from *Prevotella* and *Francisella* 1, Cpf1) are two leading genome editing nuclease systems for plant genome editing. In recent years, we have developed efficient and easy-to-use Cas9 and Cas12a toolkits for plant genome editing. We will report successful examples of using these reagents to edit the genomes of Arabidopsis, rice, maize, tomato and carrot. For example, we have targeted multiple QTL genes for improving rice yield in elite cultivars. To understand the scope of off-target mutations in Cas9 or Cas12a-edited crops, we recently conducted a whole-genome sequencing (WGS) analysis of 34 plants edited by 12 guide RNAs for Cas9 and 15 plants edited by three guide RNAs for Cas12a in T0 and T1 generations along with 20 diverse control plants in rice, a major food crop with a genome size of ~380 Mb. The sequencing depth ranged from 45X to 105X with reads mapping rate above 96%. Our comprehensive and rigorous analysis of WGS across multiple sample types suggests both Cas9 and Cpf1 nucleases are very specific in generating targeted DNA modifications in plants.