Targeted Viral-Mediated Plant Genome Editing Using CRISPR/Cas9

Novel plant genome engineering system for prevention or attenuation of plant viral infections

Targeted genome editing has emerged as an alternative to classic plant breeding and transgenic methods to improve crop plants. Until recently, available tools for introducing site-specific double strand DNA breaks were restricted to zinc finger nucleases and TAL effector nucleases. However, these technologies have not been widely adopted by the plant research community due to complicated design and laborious assembly of specific DNA binding proteins for each target gene. Within the last few years, a simpler and easier method has emerged based on the bacterial type II CRISPR (clustered regularly interspaced short palindromic repeats)/Cas (CRISPR-associated) immune system. Researchers at KAUST have adapted the CRISPR/Cas9 system to function as molecular immunity machinery against DNA viruses. The system has been demonstrated against multiple virus infections.

Benefits

- Allows targeted cleavage of genomic DNA guided by a customizable small noncoding RNA
- Results in high genomic modification frequencies
- Facilitates multiplexing and could lead to germinal transmission of the genomic modifications in the progeny
Applications

- Agricultural biotechnology
- Plant genome engineering
- CRISPR/Cas9

Opportunity

This technology is part of KAUST’s technology commercialization program that seeks to stimulate development and commercial use of KAUST-developed technologies.

Opportunities exist for joint development, patent licensing, or other mutually beneficial relationships.

For More Information

ip@kaust.edu.sa

Technology Details

The CRISPR/Cas system is programmable to target any RNA, unlike any other current genome editing systems. The CRISPR-associated protein Cas9 acts as an RNA-guided endonuclease when complexed with two short RNA molecules, which act as guide RNAs, complementary to a nucleic acid target. The system developed by the researchers could enable precise manipulation of specific genomic elements across a number of plant species and could help in determining target gene function in plant biology and diseases.

How It Works

The system is a highly efficient viral-mediated genome-editing platform that facilitates multiplexing, obviates stable transformation, and is applicable across plant species. The researchers have demonstrated the system in the RNA2 genome of the tobacco rattle virus, used frequently in virus-induced gene splicing applications. The RNA2 genome was engineered to carry and systemically deliver the guide RNA molecules into Nicotiana benthamiana plants overexpressing Cas9 endonuclease. High genomic modification frequencies were observed in inoculated as well as systemic leaves, including the plant growing points. The researchers have also demonstrated the system against the tomato leaf curly virus infections. The editing platform could be useful in plant genome engineering and be applicable across plant species amenable to viral infections for agricultural biotechnology applications.

Why It Is Better

The CRISPR/Cas system allows targeted cleavage of genomic DNA guided by a customizable small noncoding RNA, resulting in gene modifications by both non-homologous end joining and homology-directed repair mechanisms. This system facilitates multiplexing and could lead to germinal transmission of the genomic modifications in the progeny, thereby obviating the requirements of repeated transformations and tissue culture. The system enables the introduction of plant genome modifications, which are indistinguishable from those introduced by conventional breeding and mutagenesis. Furthermore, the system can deliver gRNA molecules to all parts of plants, including meristems. This may provide a general method for easily recovering seeds with the desired modification, obviating the need for transformation and/or tissue culture.

IP Protection

KAUST has a patent pending for this technology.