

Genome editing: a paradigm shift for crop biotechnology?

Editorial

Availability of genome sequences for commercially important crop plants revolutionized the basic understanding of their genetic architecture. Several genes were predicted and most of them were further confirmed by transcriptome sequencing. Transcriptome sequences obtained from different stages of crop development and stress conditions widen our understanding of basic molecular mechanisms to produce crop plants that can tolerate the altering environmental conditions.

Several reports are available in literature, where genes for different traits were tested by either overexpressing or RNAi mediated silencing. Nevertheless, discovery of sequence specific nucleases (SSN) for precisely modifying the gene of interest emerged as powerful genetic tools for editing any known region of the genome. Zinc finger nuclease (ZFN), TALEN (Transcription Activator like Effector Nucleases) and CRISPR-Cas9 (Clustered Regularly Interspaced Short Palindromic Repeats-CRISPR associated protein 9) are SSN used for targeted genome editing. ZFN and TALEN have DNA binding properties and fused with unspecific nuclease 'FokI'. Both ZFN and TALEN bind to the specific region of DNA, forms dimer and produce DNA double strand break (DSB) by catalytic activity of FokI nuclease. Whereas, CRISPR-Cas9 system utilizes gRNA (guide RNA) mediated cleavage of its complementary DNA by Cas9 nuclease. Beauty of CRISPR-Cas9 system lies in the fact that, a gRNA (~20 nucleotides) recognizes the DNA sequence of interest, whereas in case of ZFN and TALEN, protein recognizes the DNA region. Therefore, CRISPR-Cas9 system is an interesting choice for editing multiple genetic target sequences using different gRNAs simultaneously. SSN creates DSB within the target DNA sequences. DNA ends at DSB are repaired by cellular NHEJ (Non-Homologous End Joining) pathway, and mostly produce deletion of nucleotides near the cleavage site, thus causes mutation.

However, in presence of foreign DNA sequences containing

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homology arm corresponding to target DNA sequence, HDR (Homology Dependent Repair) pathway initiates and results into insertion of foreign DNA at the DSB cleavage site. Consequently, SSN have advantage not only in mutating the desired genomic sequence but also for inserting any gene of choice or correcting any undesired mutation in the genome - a great innovation in the post genome sequencing era. Scientists have also tested the efficacy for delivery of RNP (ribonucleoprotein) complex composed of gRNA-Cas9, directly into protoplast, and observed Cas9 RNP-induced mutations 24hours after transfection without any mosaicism. Crops produced through genome editing/Cas9-RNP technology will certainly enhance the precision breeding approach for useful traits and minimize the hurdle of deregulation for a sustainable agriculture.

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Conflict of interest

The author declares no conflict of interest.