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CRISPR/Cas: A New Horizon in Plant Breeding

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Abstract

Over the past few decades, the human population has increased significantly, raising serious concerns about the challenge of feeding a constantly expanding population. In such circumstances, increasing agricultural output is essential for providing humankind with both food and nourishment. Furthermore, because to their high time and labour requirements, standard breeding techniques are insufficient to supply this need. CRISPR-Cas enters the field of molecular approaches as a result to offer an alternative to the traditional techniques in order to overcome these conditions. Base editing, knock out or knock in of desirable genes, fine tuning of several genes, promotion of antiviral defence, and alteration of various demanding biochemical pathways through this specific gene editing system has proven its ability to spread new wings for accelerating crop advancement in last few years. Since the last few years, CRISPR has been significant in the advancement of studies involving genome editing.

Keywords: Breeding, CRISPR, Genetic modification, Genome editing

Introduction

In plant breeding, the traditional or conventional technique has been overwhelmingly successful. The majority of today's high yielding varieties (HYVs) and hybrids are the outcome of the same. It was crucial to the advancement of crops over time. But in order to support today's rapidly expanding human population in the challenging environment of a changing climate together with a lack of available water and arable land resources, certain novel breeding strategies were needed for enhancing genetic gain, productivity, and agricultural sustainability. Because of its lengthy processes and time-consuming nature, conventional breeding cannot satisfy the demands for increased agricultural output of today, highlighting genome editing as the most effective approaches.

CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) is a technology that allows for precise editing of genetic material. It is based on a natural defence mechanism used by bacteria and archaea to protect them from invading viruses. The CRISPR system can be programmed to target specific sequences of DNA and can be used to make precise changes to the genetic code. It has been widely used in laboratory research and has potential applications in the treatment of genetic diseases, crop improvement, and much more.

Story of CRISPR Finding

The story of the discovery of CRISPR begins with the observation of a repeating pattern of genetic sequences in the DNA of bacteria and archaea. This pattern, which is now known as CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats), was first reported in the scientific literature in the early 1980s, but its function was not initially understood. In the late 1990s and early 2000s, several research groups independently discovered that the CRISPR sequences were associated with a set of genes that code for proteins known as Cas (CRISPR-associated) proteins. They found that these Cas proteins were able to cut and degrade foreign DNA, such as that from viruses, which led to the hypothesis that CRISPR and Cas proteins together form a bacterial immune system. Further research revealed that the CRISPR sequences act as a kind of "memory" for the bacterial immune system, allowing it to recognize and target specific viral invaders. The bacteria use an enzyme called Cas9 to chop up the viral DNA at specific locations, which effectively disables the virus.

Doudna and Charpentier (2014) demonstrated that the

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CRISPR-Cas9 system could be reprogrammed to target specific sequences of DNA in a simple, efficient and precise way. This marked the beginning of the CRISPR revolution, as the discovery of the CRISPR-Cas9 system made it possible to create a new method for editing genomes that is faster, cheaper and more accurate than previous techniques. Since then, CRISPR-based techniques have been used to make precise changes to the genomes of many organisms, including plants, animals and humans.

Why CRISPR is Important?

Traditional crop breeding methods, such as crossbreeding and selection, can be effective in producing new crop varieties with improved traits. However, these methods can be time-consuming and may not always result in the desired outcomes.

In some cases, traditional breeding methods may not be able to introduce certain desirable traits because the genes responsible for those traits are not present in the crop's wild relatives or are not able to be crossed into the crop. Genetic engineering can help to overcome these limitations by allowing for the introduction of specific genes from other organisms, including wild relatives or even unrelated species, into a crop plant.

Additionally, the world population is increasing and climate change is affecting the crop growing conditions, which means that we need to increase food production in shorter time. Genetic engineering can help to achieve these goals by introducing new traits into crop plants more quickly and efficiently than traditional breeding methods. This is why genetic engineering is considered as a valuable tool in crop improvement and food production.

Overall, while traditional breeding methods have played a vital role in improving crop yields, genetic engineering can offer additional benefits and can be an important tool in meeting the challenge of feeding a growing global population. The types, applications and future possibilities of CRISPR/Cas9 are also explained in Figure 1.

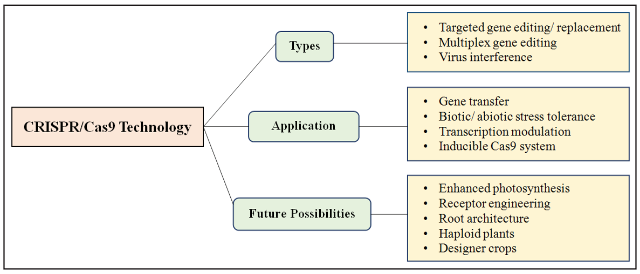

Figure 1: The types, applications and future possibilities of CRISPR/Cas9 [Adopted and modified from Khatodia *et al.* (2016)]

CRISPR Use in Agriculture

CRISPR technology has been used in agriculture to improve crop plants in several ways. By introducing genes that provide resistance to specific pathogens, researchers can create crops that are more resilient to disease. For example, researchers have used CRISPR to create a strain of rice that is resistant to the bacterial blight pathogen. Introducing genes that help plants cope with environmental stressors such as drought, high temperatures, or salinity, researchers can create crops that can grow in challenging conditions. For example, researchers have used CRISPR to create a strain of tomato that is tolerant to salt. Bu introducing genes that increase the levels of specific nutrients in crops, researchers can create crops that are more nutritious. For example, researchers have used CRISPR to create a strain of rice that is high in Vitamin A. By introducing genes that increase the number of fruits or grains produced by a plant, researchers can create crops that have higher yields. For example, researchers have used CRISPR to create a strain of tomato that has more fruit plant^{-1}. Overall, CRISPR technology has enormous potential to help improve crop plants and increase food production. However, it's worth noting that before these crops are commercialized, they will have to go through strict regulation processes and safety assessments before they can be distributed and consumed.

Benefits of CRISPR in Plant Breeding

Any gene in any species of plants can be edited by genome editing using CRISPR-Cas9. It enables faster genetic alteration than other approaches due to its simplicity, effectiveness, low cost, and ability to target many genes. Moreover, plants that were previously neglected can be genetically modified using this method. It is impossible to overstate the possibilities that this has for crop breeding and the advancement of sustainable agriculture (Zhang *et al.*, 2019). CRISPR is a relatively inexpensive and efficient method for creating new crop varieties, compared to traditional breeding methods. By introducing genes that improve the crop's yield, disease resistance, or nutritional content, CRISPR can help to increase the overall production of food, which is vital to feed the growing population. By introducing disease-resistant genes, the use of pesticides can be reduced which can help to protect the environment and human health. CRISPR allows precise changes to the genome without introducing any foreign DNA, which can reduce concerns about the safety and regulation of genetically modified crops. It's worth noting that, as with any new technology, there are also some concerns and risks associated with the use of CRISPR in plant breeding that need to be carefully considered, including ethical, social and environmental issues.

CRISPR and GMOs

Crops that have been genetically modified using CRISPR technology are considered to be genetically modified organisms (GMOs). GMOs are organisms whose genetic material has been altered in a way that does not occur naturally through mating or natural recombination. The genetic modification can be done through traditional breeding methods or through genetic engineering techniques such as CRISPR. The term GMO is often used to refer specifically to organisms that have been modified using recombinant DNA technology, which is a form of genetic engineering. However, the term can also be used more broadly to include organisms that have been modified using any form of genetic modification, including those modified using CRISPR.

Future Perspective

Future crops with improved nutritional value, improved pest resistance, and the ability to live in changing climates are ones that could be used in genome editing for sustainable, profitable agriculture. The future of crop development will involve climate resilient agriculture to battle abiotic and biotic stress employing genome editing for both the targeted mutagenesis mediated manipulation and exploration of transcriptional regulation by deconstructing physiological and molecular cross talk under combined stress. CRISPR-Cas9 technology makes it simple and effective to apply the strategy to staple crops. If photorespiration efficiency in plants can be successfully improved, more food might be produced for the world's expanding population while still using the same farmland and avoiding the need to clear more forest for agricultural use. Food security can also be achieved by using genome editing to produce crops that produce better food (Narayanan *et al.*, 2019).

Conclusion

CRISPR-Cas9 in particular, as well as genome editing in general, is a ground-breaking tool that has the possibility of impacting society, food production, and science. By enhancing plants' nutritional content and yield while making them more resilient to biotic and abiotic stressors, CRISPR-Cas9 holds immense promise for revolutionising agriculture. These qualities are required to satisfy the demands of a growing global population. The scientific community must address the different biosafety and societal concerns associated with this technology in order to employ it successfully and durably in crop development. Also, there is a need to review the laws regulating genome-edited plants and to educate the general public about their capabilities.

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