

CRISPR-Cas9: A New Way to Improve Crops Through Gene Editing

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ABSTRACT

Biotic and abiotic stresses drive major global crop losses, a problem intensified by climate change. Classical breeding and genetic engineering, are useful but they are slow, limited in genetic diversity, and often face regulatory challenges. CRISPR-Cas9 genome editing offers a precise, affordable alternative, using a guide RNA and Cas9 enzyme to introduce targeted DNA changes without foreign gene insertion. India has emerged as a leader among developing nations, with public institutions developing transgene-free, genome-edited rice, wheat, and mustard varieties, while millets stand out as a promising next frontier for climate-resilient nutrition. Despite challenges—limited genomic resources, regulatory ambiguity, and smallholder access—supportive policies such as India's 2022 genome-editing guidelines are accelerating adoption. With continued investment and clear regulation, CRISPR-Cas9 can help build a more resilient, food-secure agricultural future.

INTRODUCTION:

Climate change, rapid population escalation, dwindling natural resources, and growing need for nutritious food are among the most pressing challenges of current world agriculture. These

macro-level challenges are increasingly expressed on the ground through two direct threats to crop productivity: biotic stress, caused by living agents like pests and disease,

and abiotic stress, caused by environmental conditions like drought, heat, and salinity.

Pests and diseases alone claim up to 40% of the world's crops each year, according to the FAO, with Indian government estimates putting the domestic toll at around 30%. Abiotic stress, meanwhile, is considered the single biggest constraint on yields, with studies showing losses exceeding 50% in major crops under such conditions — and climate change is intensifying both, by increasing the frequency of extreme weather and expanding the range of many pests and pathogens.

Plant breeding through selection and genetic manipulation has long served as a key strategy for improving food supply and biofortification, with classical breeding techniques with classical techniques integrating desirable traits into crops. However, it is limited by narrow genetic variation and requires multiple breeding cycles to achieve complex traits like stress resistance or better nutrition. Genetic engineering enables direct gene transfer but faces biosafety, regulatory, and public acceptance challenges. Genome editing technologies have therefore emerged as a promising alternative for precise crop improvement.

Among the available genome editing platforms, the Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-associated protein 9 (CRISPR-Cas9) system has gained prominence due to its simplicity, cost-effectiveness, and high specificity in targeting desired genomic sequences. It enables scientists to correct DNA sequence errors and control gene expression—activating or silencing specific genes—within cells and organisms, producing changes similar to those naturally occurring mutations, without introducing any foreign DNA (Chen *et al.*, 2019). CRISPR-Cas9 has found wide application in crop improvement, including the

development of higher-yielding varieties, enhanced nutritional content, and greater resistance to pests, diseases, and environmental stresses such as drought and salinity.

1. CRISPR-Cas9 Technique: Principle and Mechanism

CRISPR-Cas9 (Clustered Regularly Interspaced Short Palindromic Repeats–CRISPR associated protein 9) has emerged as a prominent genome -editing mechanism which is being developed from the immunological defence system of bacteria and archaea. In its original setting, the CRISPR system allows microorganisms to shield themselves against invading the plasmids and viruses by capturing fragments of foreign DNA segment and unifying them into their own genome. These captured fragments are likely to serve as the molecular memory, enabling the organism to recognize and cleave the same DNA segment upon any recurring infection (Barrangou *et al.*, 2007). This natural defence mechanism is essentially a set of **molecular scissors** — a system that can be programmed to cut DNA at a specific, chosen location.

The CRISPR-Cas9 system consists of two essential components: a guide RNA (gRNA) and the Cas9 enzyme. A short RNA molecule (the gRNA) acts like a **GPS**, guiding the Cas9 scissors to the exact spot in the DNA that needs editing, based on its complementary sequence to the target region. Since this discovery, CRISPR-Cas9 has become one of the most flexible and widely used tools for crop improvement and genomic research.

Guided entirely by the gRNA sequence, Cas9 first recognizes a short adjacent sequence called the protospacer adjacent motif (PAM), which confirms the correct target site before cutting. Cas9 then introduces a precise double-strand break in the DNA, which the cell's

repair machinery seals—and it is during this repair that desired genetic changes, such as improved crop traits, are introduced.

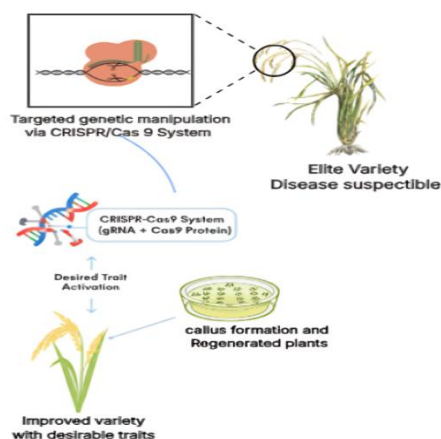


Fig 1. Application of CRISPR/Cas9 system for developing elite variety with desirable improved traits in rice crop.

2. Applications of CRISPR-Cas9 in Global Agriculture

CRISPR-Cas9 technology has been rapidly integrated into crop development and agricultural research programs across both developed and developing nations. Compared to traditional breeding techniques, its ability to make precise, targeted genetic modifications has allowed researchers to address major crop production constraints more effectively. As a result, CRISPR-Cas9 is increasingly applied to improve stress tolerance, disease resistance, yield potential, and quality traits across a wide range of economically important crops.

2.1 Enhanced Abiotic Stress Resistance

In rice, researchers have precisely altered genes that control drought and salinity response, improving stress tolerance without reducing yield. Similarly, in soybean and maize, CRISPR-based modifications have enhanced tolerance to water scarcity and heat stress (Zhang *et al.*, 2018). These outcomes illustrate the technology's potential for building climate resilience into staple crops — by directly editing the genes that govern how a

plant responds to drought, salinity, and high temperature, three of the leading constraints on global crop productivity.

2.2 Resistance to Pests and Diseases

In wheat and tomato, editing of the *MLO* gene has produced effective defence against powdery mildew, a major fungal disease. In citrus, CRISPR-mediated modification of the susceptibility gene *CsLOB1* has increased disease resistance. These examples highlight one of CRISPR's most effective agricultural uses: engineering disease resistance by altering the host's own susceptibility genes rather than introducing foreign DNA — an approach that reduces dependence on chemical pesticides and supports both environmental sustainability and economic viability in farming systems (Wang *et al.*, 2014).

2.3 Increasing Yield and Improving Quality

In rice, editing of the *Gn1a* and *GS3* genes — which govern grain size and yield potential — has improved productivity. In tomato, CRISPR-based edits have enhanced fruit size, shelf life, and ripening consistency (Li *et al.*, 2016). These gains have been achieved by focusing on genes responsible for grain development, plant architecture, and flowering time — demonstrating how targeted edits to a small number of genes can meaningfully shift yield and quality outcomes.

CRISPR in Action

In India, CRISPR-Cas9 has driven notable crop improvements: in rice, ICAR has edited genes like *Gn1a* and *DEP1* to boost yield, enhanced drought and salinity tolerance, and achieved earlier maturity for climate resilience—all without foreign DNA, keeping these lines non-transgenic; in wheat, researchers have improved resistance to rust and powdery mildew, reduced gluten aggregation by targeting gliadin genes, and increased grain size and yield; and in mustard,



CRISPR-edited varieties show improved fatty acid composition and oil quality along with greater resistance to fungal infections and aphids, offering a less contentious alternative to conventional GM mustard hybrids.

Millets: the next frontier. Millets — including finger millet, pearl millet, foxtail millet, and sorghum — are gaining growing national and international attention for their exceptional nutritional value, climate resilience, and suitability for marginal, dryland environments. Yet despite this importance, millets remain comparatively underexplored in genetic research compared to major cereals. CRISPR-Cas9 offers a powerful opportunity to close that gap, enabling precise modification of native genes linked to stress tolerance, yield, nutrition, and disease resistance:

- **Abiotic stress tolerance:** *DREB* (Dehydration Responsive Element Binding) genes can be modified to improve drought and heat resistance, while targeted edits to ABA signalling genes — including *NCED*, *HKT*, and *SOS* — hold potential for improving water-use efficiency and salinity tolerance.
- **Yield improvement:** Editing genes such as *TB1* (Teosinte Branched 1) and *DEP1*, which regulate tillering, branching, and panicle structure, can help optimize plant growth and boost grain quantity. Genes like *GS3* and *GW2*, which govern grain size and weight, offer further routes to increased yield.
- **Nutritional biofortification:** Since millets are naturally rich in micronutrients, edits to genes like *NAS* (Nicotianamine Synthase) and *ZIP* transporter genes could further increase iron and zinc content and improve micronutrient transport efficiency; edits to storage protein genes could improve protein content and digestibility.
- **Disease resistance:** Editing genes such as *SWEET* (linked to bacterial infection spread) and *MLO* (linked to powdery mildew susceptibility) could reduce reliance on chemical pesticides while strengthening the crop's natural disease resistance.

Challenges and Opportunities

Despite its promise, CRISPR-Cas9 in agriculture faces key limits: many crops lack reference genomes and functional genomics data, and high heterozygosity or polyploidy complicates precise, stable editing. Poor tissue regeneration protocols further restrict adoption in several species. Regulatory confusion with GMOs, inconsistent global policies, and unequal access for smallholder farmers add further challenges. Still, progress continues—India's 2022 guidelines eased regulations for certain edits, and initiatives like the International Year of Millets (2023) are boosting investment in resilient crops.

CONCLUSION

CRISPR-Cas9 has emerged as a transformative tool in modern agriculture, enabling precise genetic improvement at a pace conventional breeding cannot match. India's progress in genome-edited rice, wheat, and mustard demonstrates this potential in practice — and millets stand to benefit most, given their natural resilience and nutritional value, at a time when climate change and food insecurity make such traits increasingly valuable. Realizing this potential will require sustained investment in research, clear regulatory support, and integration with existing breeding programs. With responsible implementation, CRISPR-Cas9 could play a defining role in building a more resilient and food-secure agricultural future.

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