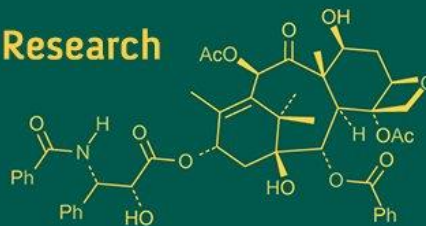
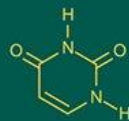


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CRISPR-based innovations in seed development and quality enhancement: A comprehensive review

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Abstract

CRISPR-based genome editing has emerged as a revolutionary tool in modern seed biotechnology, offering precise, rapid, and cost-effective approaches for crop improvement and seed quality enhancement. This review comprehensively examines recent advances in CRISPR-mediated innovations related to seed development, nutritional improvement, stress tolerance, and sustainable crop production. The study highlights the role of CRISPR/Cas systems, particularly Cas9, Cas12a, base editing, and prime editing, in modifying genes associated with seed size, germination, dormancy, vigor, storage longevity, and nutrient accumulation. Applications across major crops such as rice, wheat, maize, soybean, and horticultural species demonstrate the significant potential of genome editing for improving yield, disease resistance, oil quality, micronutrient enrichment, and climate resilience. The review also discusses emerging technologies including multiplex editing, nanotechnology-based delivery systems, artificial intelligence-assisted guide RNA design, and multi-omics integration for precision breeding. Furthermore, important regulatory, biosafety, ethical, and socioeconomic concerns related to CRISPR-edited crops are critically analyzed. Despite challenges such as off-target effects, transformation limitations, and regulatory uncertainty, CRISPR technology holds immense promise for developing high-quality, climate-smart seeds capable of supporting global food security, sustainable agriculture, and future resilient food systems.

Keywords: CRISPR-Cas Systems, genome editing, seed quality, seed development, crop improvement, abiotic stress tolerance, biofortification, precision breeding

1. Introduction

1.1 Background and Importance of Seed Quality

Seeds are the fundamental unit of agricultural production and play a decisive role in determining crop establishment, productivity, and food security. High-quality seeds contribute significantly to improved germination, uniform plant growth, enhanced stress tolerance, and higher yield potential in both field and horticultural crops. Seed quality is generally evaluated through several critical parameters, including viability, vigor, genetic and physical purity, dormancy behavior, and germination capacity, all of which directly influence crop performance under diverse environmental conditions (Ali *et al.*, 2023; Hasan *et al.*, 2023). In recent years, increasing global demand for nutritious and resilient crops has intensified the need for advanced seed improvement strategies capable of addressing both productivity and quality concerns. Conventional seed enhancement methods, although valuable, often require long breeding cycles and may lack precision in modifying complex seed-related traits (Shivani *et al.*, 2025). Furthermore, environmental stresses such as drought, salinity, heat, and emerging pests negatively affect seed development and storage longevity, thereby threatening sustainable agricultural systems (Farooq & Nadeem, 2022) [2, 11, 29, 49].

1.2 Need for Advanced Breeding Technologies

Traditional breeding approaches have contributed substantially to crop improvement; however, they are constrained by limited genetic variability, time-consuming selection processes, and linkage drag associated with undesirable traits. Similarly, transgenic technologies have faced regulatory, biosafety, and public acceptance challenges in many countries (Ricroch *et al.*, 2022) [27]. The growing impacts of climate change, rapid population expansion, shrinking arable land, and increasing food insecurity demand innovative breeding

technologies capable of generating climate-resilient and nutritionally superior seeds within shorter timeframes (Gao, 2021; Ahmed & Fatima, 2025). Precision breeding tools have therefore emerged as promising alternatives for targeted modification of genes associated with seed quality and crop adaptability [8, 44].

1.3 Emergence of CRISPR Technology

Among modern genome-editing technologies, the CRISPR-Cas system has revolutionized plant biotechnology because of its simplicity, efficiency, accuracy, and cost-effectiveness.

CRISPR enables precise editing of target genes involved in seed development, nutrient accumulation, dormancy regulation, and stress tolerance (Chen *et al.*, 2022; Tuncel *et al.*, 2025). Compared with earlier genome-editing platforms such as TALENs and zinc-finger nucleases (ZFNs), CRISPR systems are easier to design, highly multiplexable, and more adaptable across diverse crop species (Gupta *et al.*, 2022; Yin *et al.*, 2021). Consequently, CRISPR-based approaches are increasingly being integrated into seed biotechnology for developing high-yielding and quality-enhanced crop varieties [5, 33, 10, 40].

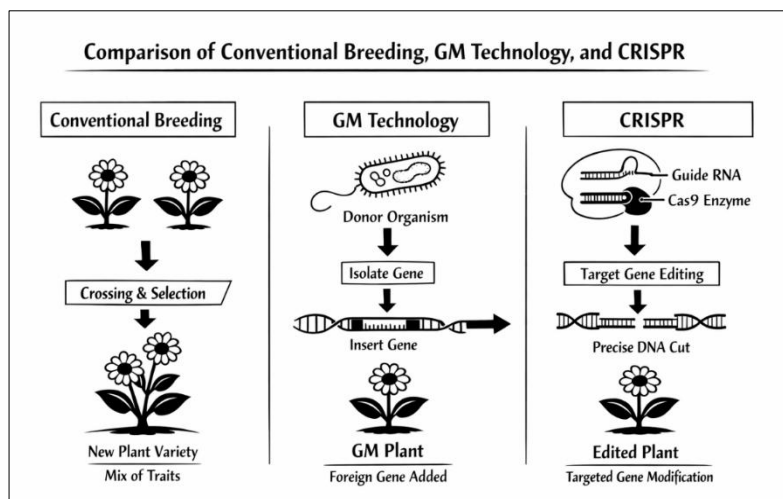


Fig 1.1: Schematic comparison of conventional breeding, genetic modification (GM) technology, and CRISPR-based genome editing in crop improvement.

1.4 Objectives and Scope of the Review

This review comprehensively examines recent advances in CRISPR-mediated seed biotechnology with particular emphasis on seed development and quality enhancement. It highlights the applications of CRISPR technologies in improving seed vigor, germination, dormancy regulation, nutritional quality, storage properties, and resistance to environmental stresses across major crop species (Khan *et al.*, 2025; Islam *et al.*, 2023). The review also discusses emerging trends, challenges, biosafety considerations, and future prospects of CRISPR-driven innovations for sustainable agriculture and global food security [14, 51].

2. Fundamentals of CRISPR-Cas Systems

2.1 Historical Development of CRISPR

The CRISPR-Cas system has emerged as one of the most transformative genome-editing technologies in modern plant biotechnology. Initially discovered as unusual repetitive DNA sequences in bacterial genomes during the late 1980s, CRISPR was later recognized as a natural adaptive immune mechanism that protects bacteria and archaea from invading viruses and foreign genetic elements. Subsequent studies revealed that CRISPR-associated (Cas) proteins function alongside guide RNAs to identify and cleave target nucleic acids with remarkable precision (Jaganathan *et al.*, 2021) [12]. The adaptation of this microbial defense system for genome engineering revolutionized crop improvement research because it enabled precise, rapid, and cost-effective modification of plant genomes compared with conventional breeding and earlier gene-editing platforms such as zinc finger nucleases and TALENs (Gao, 2021; Yin *et al.*, 2021) [8, 40]. Since the development of CRISPR-Cas9 as a programmable editing tool, researchers have expanded the

technology into advanced platforms capable of targeted mutagenesis, gene insertion, transcriptional regulation, and epigenetic modification in crops (Zhang *et al.*, 2021) [41].

2.2 Components and Mechanism

The CRISPR-Cas system mainly consists of a guide RNA (gRNA) and a Cas nuclease. The gRNA directs the nuclease toward a complementary DNA sequence, enabling sequence-specific editing (Khatodia *et al.*, 2021). Cas9 remains the most widely used nuclease because of its high editing efficiency and versatility in plants. However, newer nucleases such as Cas12a (Cpf1) and Cas13 have broadened genome-editing applications due to their unique PAM requirements and RNA-targeting capabilities (Chen *et al.*, 2022). Once the target sequence is recognized, the Cas enzyme introduces double-stranded breaks that are repaired by cellular mechanisms including non-homologous end joining (NHEJ) or homology-directed repair (HDR), leading to gene knockout, insertion, or replacement (El-Mounadi *et al.*, 2021). Recent innovations such as base editing and prime editing allow nucleotide substitutions without inducing double-strand breaks, thereby improving editing accuracy and minimizing unintended mutations (Mishra *et al.*, 2022) [53, 5, 48, 54].

2.3 Types of CRISPR Systems

Among different CRISPR platforms, CRISPR-Cas9 is the most extensively utilized system in seed trait engineering due to its simplicity and efficiency in targeted genome modification (Gupta *et al.*, 2022). CRISPR-Cas12a/Cpf1 offers additional advantages such as staggered DNA cleavage and multiplex editing, making it valuable for complex trait improvement in crops (Wang *et al.*, 2022). Furthermore,

base-editing systems facilitate precise nucleotide conversion for improving seed nutritional quality, protein composition, and stress tolerance without introducing foreign DNA (Kaur & Saini, 2024). Prime editing represents a next-generation advancement that enables highly accurate insertions, deletions, and substitutions with reduced off-target effects, thereby expanding possibilities for precision breeding (Das *et al.*, 2025) [10, 36, 13, 6].

2.4 Delivery Methods in Plants

Efficient delivery of CRISPR reagents into plant cells is essential for successful genome editing. *Agrobacterium*-mediated transformation is widely used because of its stable gene integration and high transformation efficiency in many crops (Ricroch *et al.*, 2022). Particle bombardment, also known as biolistic transformation, is particularly useful for monocot species and recalcitrant plants where *Agrobacterium* methods show limitations (Kumar *et al.*, 2022). Protoplast transformation enables transient expression of CRISPR components and supports DNA-free editing approaches that reduce regulatory concerns (Pramanik *et al.*, 2021). More recently, nanotechnology-based delivery systems have gained attention as innovative tools for transporting CRISPR molecules into plant tissues with minimal cellular damage and enhanced editing precision (Tuncel *et al.*, 2025). These advancements collectively strengthen the application of CRISPR technology for sustainable seed development and crop quality enhancement [27, 15, 22, 33].

3. Seed Development: Biological and Genetic Basis

Seed development is a highly coordinated biological process that determines the productivity, nutritional value, viability, and adaptability of crop plants. It involves a sequence of physiological, biochemical, and genetic events beginning from fertilization and continuing through embryo formation, endosperm differentiation, and seed maturation. Recent advances in genome editing, particularly CRISPR/Cas systems, have provided unprecedented opportunities to understand and manipulate these developmental mechanisms for improving seed quality traits in major crops (Gao, 2021; Chen *et al.*, 2022). Modern seed biotechnology integrates molecular genetics, developmental biology, and precision breeding approaches to engineer seeds with enhanced vigor, nutritional composition, stress tolerance, and storage capacity (Gupta *et al.*, 2022; Tuncel *et al.*, 2025) [8, 5, 10, 33].

3.1 Seed Formation and Development

Seed formation begins immediately after double fertilization, where one sperm nucleus fuses with the egg cell to form the embryo while the second fuses with polar nuclei to generate the endosperm. Embryogenesis is a critical developmental phase that establishes the basic body plan of the future plant, including shoot and root meristems. This process is regulated by complex gene networks involving transcription factors, signaling molecules, and epigenetic regulators (Pramanik *et al.*, 2021). Studies have shown that CRISPR-mediated editing of developmental genes can precisely alter embryo architecture, thereby improving seed viability and crop establishment (Wang *et al.*, 2023) [22, 37].

Endosperm development plays a central role in nutrient accumulation and seed filling. The endosperm serves as the primary storage tissue for starch, proteins, and lipids, which directly influence grain quality and nutritional value. Alterations in genes associated with carbohydrate

metabolism and storage protein biosynthesis have significantly enhanced seed composition in cereals and legumes (Tyagi *et al.*, 2024; Sharma & Dubey, 2024). Genome editing technologies have enabled targeted modification of pathways controlling starch synthesis, oil accumulation, and amino acid content, thereby improving both yield and nutritional quality (Li *et al.*, 2023; Singh *et al.*, 2025).

Seed maturation represents the final stage of seed development and is characterized by desiccation tolerance, reserve accumulation, and dormancy induction. During this stage, protective proteins and antioxidants accumulate to maintain seed longevity and viability during storage (Raza *et al.*, 2023). CRISPR-based approaches have facilitated the manipulation of maturation-related genes to improve seed storability and resistance to environmental stress (Das *et al.*, 2025). Enhanced understanding of these developmental phases has accelerated the development of climate-resilient and nutritionally enriched crop varieties (Ahmed & Fatima, 2025) [34, 28, 16, 31, 26, 6, 44].

3.2 Genetic Regulation of Seed Traits

Seed traits such as size, weight, dormancy, and germination are controlled by multiple quantitative trait loci and regulatory genes. Seed size and weight are major determinants of crop yield and market value. Genes involved in cell proliferation, hormonal signaling, and nutrient transport significantly influence these characteristics (Liu *et al.*, 2021). CRISPR/Cas-mediated editing of yield-associated genes has demonstrated substantial improvements in grain weight and seed biomass in rice, wheat, and maize (Ahmad *et al.*, 2024; Chandra & Patel, 2023) [1].

Seed dormancy is an adaptive trait that prevents premature germination under unfavorable conditions. However, excessive dormancy may reduce crop uniformity and germination efficiency. Hormonal balance, particularly between abscisic acid (ABA) and gibberellins (GA), plays a key role in dormancy regulation (Devi & Choudhary, 2024). Genome editing of dormancy-related genes has enabled the development of varieties with optimized germination behavior and improved field emergence (Ali *et al.*, 2023). Germination control genes regulate water uptake, enzymatic activation, and metabolic reprogramming during seed sprouting. Manipulation of these genes through CRISPR technology has improved seed vigor, accelerated germination, and enhanced tolerance to abiotic stress conditions such as salinity and drought (Farooq & Nadeem, 2022; Hasan *et al.*, 2023) [17, 1, 46, 7, 2, 11].

3.3 Molecular Pathways Influencing Seed Quality

Seed quality is strongly influenced by hormonal signaling pathways and stress-responsive molecular mechanisms. Among plant hormones, ABA regulates seed maturation, dormancy, and desiccation tolerance, whereas GA promotes germination and embryo growth. Auxins contribute to cell division, seed enlargement, and nutrient allocation during early development (Yang *et al.*, 2022). Genome editing technologies have enabled precise modulation of hormone biosynthesis and signaling pathways, resulting in enhanced seed performance and improved stress adaptability (Khatodia *et al.*, 2021; Banerjee *et al.*, 2024).

Stress-responsive pathways are equally important in maintaining seed quality under changing climatic conditions. Environmental stresses such as heat, drought, and salinity can

impair seed filling, reduce viability, and alter nutritional composition. CRISPR-based editing of stress-responsive transcription factors and defense-related genes has improved tolerance to oxidative stress and enhanced seed resilience in several crops (Das *et al.*, 2025; Ghosh *et al.*, 2026). In addition, molecular interventions targeting antioxidant systems, osmoprotectants, and stress-signaling cascades have contributed to better seed longevity and germination efficiency under adverse environments (Shivani *et al.*, 2025). Consequently, integrating molecular biology with genome editing technologies is transforming seed science and paving the way for sustainable crop improvement and global food security [39, 45, 53, 50, 29].

4. CRISPR Applications in Seed Development

CRISPR/Cas-based genome editing has emerged as a transformative technology in modern seed science due to its precision, efficiency, and ability to modify targeted genes associated with agronomically important traits. The technology has significantly accelerated seed development programs by enabling direct manipulation of genes controlling seed size, yield, germination, nutritional composition, and stress tolerance (Gao, 2021; Yin *et al.*, 2021). In recent years, CRISPR applications have expanded beyond basic crop improvement to include the development of climate-resilient and high-quality seeds capable of meeting global food security demands under changing environmental conditions (Tuncel *et al.*, 2025; Tyagi & Sharma, 2026). The ability of CRISPR systems to generate precise edits without introducing foreign DNA has further strengthened their acceptance in sustainable breeding programs and next-generation seed biotechnology (Ricroch *et al.*, 2022; Banerjee *et al.*, 2024) [8, 40, 33 59, 27, 45].

4.1 Enhancement of Seed Size and Yield

One of the most significant applications of CRISPR technology is the enhancement of seed size and grain yield through targeted editing of yield-related genes. Researchers have successfully manipulated genes regulating grain length, seed weight, and panicle architecture in major crops such as rice, wheat, maize, and soybean. Editing genes such as *GS3*, *GW2*, and *TGW6* in rice has resulted in increased grain size and improved productivity (Liu *et al.*, 2021; Ahmad *et al.*, 2024). Similarly, CRISPR-mediated modifications in wheat and maize have improved kernel weight and seed uniformity, thereby enhancing overall yield potential (Chen *et al.*, 2022; Zhang *et al.*, 2021). In soybean and oilseed crops, targeted genome editing has also contributed to better seed composition and enhanced oil biosynthesis (Rahman & Karim, 2023; Singh *et al.*, 2025). These advancements demonstrate the effectiveness of CRISPR in accelerating precision breeding for high-yielding seed varieties [17, 1, 5, 42, 57, 31].

4.2 Improvement of Seed Germination and Vigor

Seed germination and vigor are critical determinants of crop establishment and productivity. CRISPR technology has enabled precise manipulation of dormancy-related genes and hormonal pathways involved in seed germination. Editing genes associated with abscisic acid and gibberellin signaling has improved germination efficiency and reduced undesirable seed dormancy in several crops (Devi & Choudhary, 2024). Genome editing approaches have also enhanced seed vigor under adverse environmental conditions such as drought,

salinity, and oxidative stress (Ali *et al.*, 2023). Recent studies indicate that CRISPR-edited seeds exhibit better metabolic activity, rapid emergence, and improved stress adaptability during early seedling development (Hasan *et al.*, 2023; Biswal *et al.*, 2026). Such improvements are particularly important for ensuring stable crop establishment in regions facing climatic variability and soil degradation [7, 2, 11, 4].

4.3 Hybrid Seed Production

Hybrid seed production has benefited substantially from CRISPR-assisted breeding strategies. The technology has been widely used to develop male sterility and fertility restoration systems, which are essential for efficient hybrid seed production. By editing genes controlling pollen development and reproductive fertility, breeders can rapidly generate stable male-sterile lines without lengthy conventional breeding procedures (Kumar *et al.*, 2022; Chandra & Patel, 2023). CRISPR-mediated hybrid breeding has been successfully demonstrated in rice, maize, and other cereals, resulting in improved hybrid vigor, yield stability, and uniformity (Jaganathan *et al.*, 2021). Furthermore, CRISPR-based approaches facilitate faster introgression of elite traits into parental lines, thereby shortening breeding cycles and enhancing the efficiency of commercial hybrid seed systems (Ahmed & Fatima, 2025) [15, 46, 12, 44].

4.4 Seed Longevity and Storage Potential

Seed longevity is a major factor influencing seed quality during storage and distribution. CRISPR technology has enabled targeted editing of genes associated with oxidative stress responses, cellular repair mechanisms, and seed aging processes. Modifications in antioxidant-related pathways have shown potential in reducing seed deterioration and improving storage life (Raza *et al.*, 2023). Enhanced regulation of reactive oxygen species and protective metabolites through genome editing can maintain seed viability and vigor over extended storage periods (Farooq & Nadeem, 2022). Such innovations are particularly valuable for seed banks, germplasm conservation, and sustainable agricultural systems where long-term seed preservation is essential [26, 49].

4.5 Development of Climate-Resilient Seeds

The development of climate-resilient seeds is one of the most promising applications of CRISPR-based seed biotechnology. Genome editing has been extensively used to enhance tolerance against drought, salinity, heat, and cold stress by targeting stress-responsive genes and regulatory networks (Das *et al.*, 2025). In rice, wheat, and maize, CRISPR-mediated editing of transcription factors and signaling pathways has improved water-use efficiency and osmotic adjustment under drought conditions (Khatodia *et al.*, 2021). Similarly, salinity tolerance has been enhanced through modifications in ion transport and stress-responsive genes, leading to better seed germination and plant establishment in saline soils (Farooq & Nadeem, 2022). Heat- and cold-tolerant seed varieties have also been developed through precise editing of temperature-responsive genes, thereby ensuring stable productivity under extreme climatic fluctuations (Prakash & Reddy, 2025; Ghosh *et al.*, 2026). Collectively, these innovations highlight the immense potential of CRISPR technology in developing resilient seed systems for future sustainable agriculture [6, 53, 49, 50].

5. CRISPR-Based Seed Quality Enhancement

CRISPR/Cas-mediated genome editing has emerged as a transformative approach for improving seed quality traits in major food and oilseed crops. Unlike conventional breeding, which is often time-consuming and limited by genetic variability, CRISPR technology enables precise modification of target genes associated with nutritional composition, disease resistance, processing quality, and seed storage characteristics (Gao, 2021; Gupta *et al.*, 2022). Recent advances in genome engineering have accelerated the development of nutritionally superior and climate-resilient crop varieties with enhanced consumer and industrial value (Tuncel *et al.*, 2025; Tyagi & Sharma, 2026) [8, 10, 33, 59].

5.1 Nutritional Biofortification

One of the most significant applications of CRISPR technology in seed improvement is nutritional biofortification. Genome editing has been widely used to enhance seed protein quality by modifying genes involved in amino acid biosynthesis and seed storage proteins. In legumes and cereals, CRISPR-mediated editing has improved lysine content, digestibility, and overall protein balance, thereby addressing protein malnutrition in developing regions (Sharma & Dubey, 2024; Tyagi *et al.*, 2024). Similarly, targeted manipulation of genes regulating micronutrient accumulation has enabled the enrichment of iron, zinc, provitamin A, and folate in staple crops such as rice, wheat, and maize (Barman *et al.*, 2025; Kaur & Saini, 2024). Researchers have also reported significant progress in enhancing nutraceutical compounds and antioxidant metabolites through CRISPR-based metabolite engineering (Mehta *et al.*, 2025; Nidhi & Verma, 2024). In oilseed crops, editing genes associated with fatty acid biosynthesis has improved oil quality by increasing oleic acid content and reducing undesirable saturated fats, resulting in healthier edible oils with greater oxidative stability (Mahapatra & Roy, 2024; Singh *et al.*, 2025). These developments demonstrate the potential of CRISPR technology to support global nutritional security and functional food production [28, 34, 3, 13, 20, 55, 19, 31].

5.2 Disease and Pest Resistance: CRISPR-based editing has also contributed substantially to enhancing resistance against seed-borne and field pathogens. By targeting susceptibility genes or activating defense-related pathways, scientists have developed crop varieties with improved resistance to fungal, bacterial, and viral diseases (Jaganathan *et al.*, 2021; Hasan *et al.*, 2023). For example, CRISPR-mediated disruption of pathogen susceptibility genes in rice and wheat has improved tolerance against blast disease and bacterial blight (Liu *et al.*, 2021; Zhang *et al.*, 2021). In addition, genome editing has enabled the development of insect-resistant crops through modification of genes involved in plant defense signaling and secondary metabolite production (Rajput *et al.*, 2021). Such targeted interventions reduce dependence on chemical pesticides and promote environmentally sustainable agriculture. Emerging CRISPR platforms, including base editing and prime editing, are further improving the accuracy and efficiency of disease-resistance breeding programs (Mishra *et al.*, 2022; Khan *et al.*, 2025) [12, 11, 18, 42, 24, 54, 14].

5.3 Reduction of Anti-Nutritional Factors: Another important area of seed quality enhancement involves reducing anti-nutritional compounds and allergenic proteins.

CRISPR technology has been successfully used to eliminate toxic metabolites and allergenic seed proteins that negatively affect human health and nutrient absorption (Liu *et al.*, 2021; Yang *et al.*, 2022). Low-phytate crop varieties developed through targeted gene editing exhibit improved mineral bioavailability because phytate normally binds essential micronutrients such as iron and zinc (Jha *et al.*, 2021). Likewise, editing gluten-related genes in wheat has shown promise for developing low-gluten products suitable for individuals with gluten intolerance and celiac disease (Tyagi *et al.*, 2024). Such innovations not only improve nutritional quality but also expand the market potential of edited crops in health-conscious consumer sectors [18, 39, 52, 34].

5.4 Improvement in Seed Processing and Industrial Traits

CRISPR applications are increasingly being extended to seed processing and industrial quality traits. Editing genes associated with seed senescence and oxidative degradation has improved seed longevity and shelf life during storage (Raza *et al.*, 2023). In cereals and pulses, genome editing has enhanced milling efficiency, cooking quality, starch composition, and seed texture, thereby improving industrial processing performance (Li *et al.*, 2023; Yadav *et al.*, 2024). Furthermore, manipulation of genes controlling seed coat thickness, hardness, and pigmentation has contributed to better seed handling, improved appearance, and increased resistance to mechanical damage (Wang *et al.*, 2023; Chandra & Patel, 2023). Collectively, these advancements highlight the broad potential of CRISPR technology in developing high-quality seeds that meet the evolving demands of agriculture, food industries, and consumers [26, 16, 38, 37, 46].

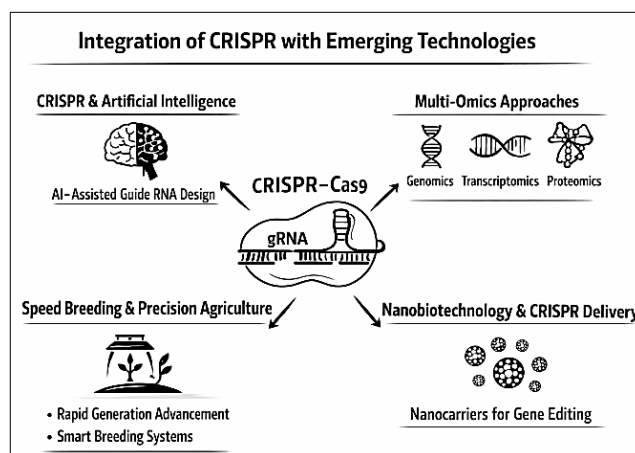


Fig 5.1: Integration of CRISPR Technology with Artificial Intelligence, Multi-Omics,

Precision Agriculture, and Nanobiotechnology for Advanced Seed Improvement and Smart Breeding Systems

The schematic diagram illustrates the integration of CRISPR technology with emerging scientific and digital innovations to accelerate modern crop improvement and seed biotechnology. At the center, the CRISPR-Cas9 system functions as the core genome-editing platform connected with four major technological domains. Artificial Intelligence (AI) supports efficient guide RNA (gRNA) design, improving editing precision and reducing off-target effects. Multi-omics approaches, including genomics, transcriptomics, and proteomics, provide comprehensive molecular insights for identifying target genes associated

with seed quality and stress tolerance. The diagram also highlights the role of speed breeding and precision agriculture in enabling rapid generation advancement and smart breeding systems for faster crop development. In addition, nanobiotechnology-based delivery systems use nanocarriers for efficient transport of CRISPR components into plant cells. Collectively, these integrated technologies enhance the efficiency, accuracy, and sustainability of next-generation crop breeding and seed improvement programs.

6. Major Crop-Specific Case Studies

6.1 Rice

Yield Enhancement and Blast Resistance

Rice has emerged as one of the most extensively studied crops for CRISPR/Cas-mediated seed improvement and trait enhancement. Researchers have successfully edited genes associated with grain yield, panicle architecture, and disease

resistance to develop superior rice cultivars with improved productivity and resilience. Editing of yield-related genes such as *Gn1a*, *DEP1*, and *GS3* has contributed to enhanced grain number, seed size, and overall yield performance. In addition, CRISPR/Cas9-mediated modification of susceptibility genes has significantly improved resistance against rice blast disease caused by *Magnaporthe oryzae*, thereby reducing yield losses and dependence on chemical fungicides. Recent studies also demonstrated successful development of herbicide-resistant and stress-tolerant rice lines through homology-directed repair and multiplex genome editing approaches. These innovations indicate that CRISPR technology can accelerate precision breeding in rice while maintaining desirable seed quality traits, nutritional value, and environmental adaptability (Gao, 2021; Sun *et al.*, 2021; Wang *et al.*, 2022; Ahmad *et al.*, 2024; Tuncel *et al.*, 2025) [8, 32, 36, 1, 33].

Table 1: CRISPR-Based Case Studies for Yield Enhancement and Blast Resistance in Rice

Trait Targeted	Gene/Approach	Major Outcome	References
Grain yield enhancement	<i>Gn1a</i> , <i>GS3</i> , <i>DEP1</i> editing	Increased grain size and panicle productivity	Liu <i>et al.</i> (2021); Ahmad <i>et al.</i> (2024) [17, 1]
Blast resistance	Susceptibility gene editing	Enhanced resistance to <i>Magnaporthe oryzae</i>	Gao (2021); Jaganathan <i>et al.</i> (2021) [8, 12]
Herbicide resistance	HDR-based CRISPR/Cas9 editing	Development of herbicide-tolerant rice lines	Sun <i>et al.</i> (2021) [32]
Stress tolerance	Multiplex CRISPR editing	Improved climate resilience and seed vigor	Das <i>et al.</i> (2025); Hasan <i>et al.</i> (2023) [6, 11]

6.2 Wheat

Grain Quality and Disease Resistance

CRISPR-based genome editing has significantly improved wheat breeding by enabling precise modification of genes controlling grain quality and disease resistance. Wheat quality enhancement primarily focuses on editing genes related to gluten composition, seed storage proteins, and starch biosynthesis to improve processing and nutritional properties. Researchers have targeted gluten-associated genes to develop low-gluten wheat varieties suitable for individuals

with gluten sensitivity while preserving baking quality. Simultaneously, CRISPR/Cas systems have been used to improve resistance against fungal diseases such as powdery mildew and rust by editing susceptibility genes like *MLO*. These developments contribute to higher yield stability and reduced fungicide dependency. Advanced genome-editing approaches, including base editing and multiplex editing, are also facilitating rapid development of climate-resilient wheat cultivars with enhanced grain characteristics and improved seed longevity

Table 2: Applications of CRISPR/Cas Systems for Grain Quality Improvement and Disease Resistance in Wheat

Trait Targeted	Gene/Approach	Major Outcome	References
Grain quality improvement	Seed storage protein editing	Improved nutritional and processing quality	Li <i>et al.</i> (2023); Tyagi <i>et al.</i> (2024) [16, 34]
Low-gluten wheat	Gluten gene modification	Reduced allergenic potential	Liu <i>et al.</i> (2021); Yang <i>et al.</i> (2022) [18, 39]
Powdery mildew resistance	<i>MLO</i> gene editing	Enhanced fungal disease resistance	Jaganathan <i>et al.</i> (2021); Khatodia <i>et al.</i> (2021) [12, 53]
Seed longevity	Precision genome editing	Improved storage and seed viability	Raza <i>et al.</i> (2023) [26]

6.3 Maize

Stress Tolerance and Nutritional Traits

Maize has benefited considerably from CRISPR-mediated interventions aimed at improving abiotic stress tolerance, nutrient content, and seed quality traits. Genome editing strategies have targeted genes associated with drought tolerance, heat stress adaptation, and nitrogen-use efficiency to enhance maize productivity under changing climatic conditions. In addition, biofortification efforts have focused on increasing provitamin A, essential amino acids, and micronutrient accumulation in maize kernels. CRISPR/Cas technology has also enabled modification of starch biosynthesis pathways, improving industrial and nutritional quality. These approaches are increasingly important for ensuring food security and sustainable crop production in

stress-prone environments. Recent advances in base editing and multiplex gene editing further support rapid improvement of complex seed traits in maize breeding programs.

6.4 Soybean

Oil Quality and Protein Enhancement

Soybean is a major oilseed and protein crop where CRISPR technology has shown remarkable potential in improving seed composition and nutritional quality. Genome editing has been widely used to modify fatty acid biosynthesis pathways, leading to increased oleic acid content and reduced undesirable fatty acids, thereby improving oil stability and nutritional value. Researchers have also targeted genes

Table 3: CRISPR-Mediated Innovations for Stress Tolerance and Nutritional Enhancement in Maize

Trait Targeted	Gene/Approach	Major Outcome	References
Drought tolerance	Stress-responsive gene editing	Improved adaptation under water deficit	Farooq & Nadeem (2022); Das <i>et al.</i> (2025) [49, 6]
Nutritional enhancement	Biofortification pathways	Increased vitamin and micronutrient content	Barman <i>et al.</i> (2025); Dutta & Bose (2025) [3, 47]
Nitrogen-use efficiency	Precision genome editing	Better nutrient utilization	Gupta <i>et al.</i> (2022) [10]
Seed quality improvement	Starch pathway modification	Enhanced industrial and food quality	Chen <i>et al.</i> (2022); Hasan <i>et al.</i> (2023) [5, 11]

associated with seed protein accumulation to enhance amino acid composition and overall protein quality. Such modifications are particularly important for food, feed, and industrial applications. Furthermore, CRISPR-based editing has contributed to improved stress tolerance and seed storage

characteristics in soybean. These advancements demonstrate the potential of precision breeding to develop high-value soybean cultivars with superior seed traits and better market acceptance

Table 4: Genome Editing Approaches for Oil Quality and Protein Enhancement in Soybean

Trait Targeted	Gene/Approach	Major Outcome	References
Oil quality enhancement	Fatty acid biosynthesis editing	Increased oleic acid content	Mahapatra & Roy (2024); Singh <i>et al.</i> (2025) [19, 31]
Protein enhancement	Seed protein gene modification	Improved amino acid composition	Sharma & Dubey (2024); Jha <i>et al.</i> (2021) [28, 52]
Stress resilience	CRISPR/Cas-mediated editing	Improved adaptability and seed quality	Rahman & Karim (2023) [57]
Nutraceutical improvement	Metabolite engineering	Enhanced health-promoting compounds	Mehta <i>et al.</i> (2025); Nidhi & Verma (2024) [20, 55]

6.5 Horticultural and Vegetable Crops

Tomato, Canola, Potato, etc

CRISPR technology has accelerated the improvement of horticultural and vegetable crops by enabling targeted modification of genes associated with seed quality, nutritional composition, shelf life, and stress resistance. In tomato, genome editing has been employed to improve fruit ripening, seed vigor, disease resistance, and nutritional quality. Potato breeding programs have utilized CRISPR/Cas systems to reduce enzymatic browning and improve starch quality, while canola improvement efforts have focused on

enhancing oil composition and stress tolerance. Researchers have also explored de novo domestication approaches in wild horticultural species to rapidly introduce desirable agronomic and seed traits. Recent innovations integrating genome editing with seed coating and microbiome-based technologies are further strengthening sustainable horticultural production systems. These developments highlight the growing role of CRISPR in producing high-quality vegetable seeds and resilient horticultural crops with improved consumer and industrial value

Table 5: CRISPR Applications in Horticultural and Vegetable Crops for Seed and Quality Improvement

Crop	Trait Targeted	Major Outcome	References
Tomato	Fruit ripening and nutrition	Improved shelf life and nutritional quality	Wang <i>et al.</i> (2023); Zsögön <i>et al.</i> (2021) [37, 60]
Potato	Starch quality and browning reduction	Better processing quality	Wang <i>et al.</i> (2023) [37]
Canola	Oil composition	Enhanced edible oil quality	Mahapatra & Roy (2024); Pandey <i>et al.</i> (2026) [19, 21]
Horticultural crops	Seed vigor and resilience	Improved germination and sustainability	Biswal <i>et al.</i> (2026); Singh <i>et al.</i> (2026) [4, 30]

8. Regulatory, Ethical, and Biosafety Considerations

The rapid advancement of CRISPR-based genome editing in seed science has generated significant interest among researchers, policymakers, and agricultural industries worldwide. However, alongside its scientific potential, the technology has raised important regulatory, ethical, and biosafety concerns that influence its global adoption and commercialization. Different countries have adopted contrasting regulatory frameworks for gene-edited crops. In the United States, CRISPR-edited crops that do not contain foreign DNA are often regulated less stringently and may bypass extensive GMO regulations, thereby accelerating innovation and commercialization (Gao, 2021; Ricroch *et al.*, 2022). In contrast, the European Union continues to classify

most genome-edited crops under strict genetically modified organism (GMO) legislation, resulting in complex approval procedures and limited field deployment (Ricroch *et al.*, 2022). China has recently introduced supportive regulatory guidelines for gene-edited crops to strengthen food security and agricultural competitiveness, whereas India is gradually developing a differentiated framework that exempts certain SDN-1 and SDN-2 edited crops from strict transgenic regulations (Gupta *et al.*, 2022; Tuncel *et al.*, 2025). These regulatory disparities create uncertainty for international trade and research collaborations. Biosafety remains another major concern associated with CRISPR applications in seed development. Although CRISPR/Cas systems are considered highly precise,

unintended off-target mutations may still occur, potentially affecting non-target genes and altering plant metabolism or agronomic performance (El-Mounadi *et al.*, 2021; Yin *et al.*, 2021). Researchers have therefore focused on improving guide RNA specificity, high-fidelity Cas variants, and advanced base-editing systems to minimize unintended genomic alterations (Mishra *et al.*, 2022). Gene flow from edited crops to wild relatives or conventional varieties also raises ecological concerns, particularly regarding biodiversity conservation and the emergence of herbicide-resistant weeds (Rajput *et al.*, 2021). Long-term environmental monitoring and biosafety assessments are therefore essential before widespread commercialization of gene-edited seeds.

Ethical and socioeconomic dimensions are equally critical in determining the future of CRISPR-based seed technologies. Public acceptance of gene-edited crops varies across regions and is strongly influenced by awareness, transparency, and trust in regulatory systems (Gill *et al.*, 2021). Many consumers perceive genome editing differently from traditional GMOs because CRISPR can introduce targeted mutations without inserting foreign genes; nevertheless, misinformation and ethical debates continue to shape public opinion (Hasan *et al.*, 2023). Intellectual property rights surrounding CRISPR patents also create challenges, as licensing restrictions may limit access for public-sector researchers and developing countries (Banerjee *et al.*, 2024). Furthermore, there is concern that expensive genome-editing technologies may primarily benefit large agribusiness corporations while marginalizing smallholder farmers who lack financial and technical resources. Ensuring equitable access, affordable seed systems, and inclusive innovation policies will therefore be essential for sustainable adoption of CRISPR technologies in agriculture (Ahmed & Fatima, 2025).

9. Challenges and Limitations

Despite remarkable progress in CRISPR-mediated seed engineering, several scientific and practical limitations continue to restrict its broader application. One of the major technical challenges is inconsistent editing efficiency across crop species and genotypes, particularly in polyploid and recalcitrant crops where precise editing remains difficult (Chen *et al.*, 2022). Off-target mutations, although reduced through improved editing platforms, still represent a concern for genomic stability and biosafety (Yin *et al.*, 2021). In many crops, low transformation and regeneration efficiency further limit the successful production of edited plants, especially in legumes and horticultural species (Wang *et al.*, 2023). Regulatory uncertainties across countries also slow commercialization and international acceptance of gene-edited seeds. Additionally, the high costs associated with laboratory infrastructure, sequencing technologies, regulatory approvals, and product commercialization create barriers for research institutions in developing nations (Khan *et al.*, 2025). These limitations highlight the need for more efficient, affordable, and universally accepted genome-editing strategies.

10. Future Perspectives and Research Directions

Future developments in CRISPR technology are expected to revolutionize seed biotechnology through the emergence of next-generation genome-editing tools such as base editing, prime editing, multiplex editing, and CRISPR-associated transposases, which offer greater precision and flexibility in

crop improvement (Mishra *et al.*, 2022; Tuncel *et al.*, 2025). The concept of *de novo* domestication using CRISPR has also opened new opportunities for converting wild plant species into climate-resilient crops with superior nutritional and agronomic traits (Zsögön *et al.*, 2021). Researchers are increasingly focusing on climate-smart seed systems capable of tolerating drought, salinity, heat, and emerging pests under changing climatic conditions (Das *et al.*, 2025). Another promising direction involves the genome editing of orphan crops and underutilized species that are important for regional food security but have historically received limited research attention (Tyagi & Sharma, 2026). Integration of CRISPR technology with microbiome engineering, seed coating innovations, and digital breeding platforms may further enhance seed quality and agricultural sustainability (Biswal *et al.*, 2026; Singh *et al.*, 2026). Overall, CRISPR-based seed innovations hold immense potential to support sustainable agriculture, improve nutritional security, and strengthen global food systems in the coming decades. [36, 42, 44, 45, 46, 47, 50, 51, 59]

Conclusion

CRISPR-based genome editing has transformed the field of seed biotechnology by providing highly precise, efficient, and flexible tools for improving seed development and quality traits in crop plants. The technology has significantly accelerated modern breeding programs by enabling targeted manipulation of genes associated with seed size, germination, dormancy, nutrient composition, stress tolerance, disease resistance, and storage longevity. Compared with conventional breeding methods, CRISPR systems offer faster and more accurate genetic improvement without extensive linkage drag or prolonged selection cycles. Applications in major crops such as rice, wheat, maize, soybean, and several horticultural species clearly demonstrate the enormous potential of CRISPR-mediated breeding for enhancing agricultural productivity, nutritional quality, and climate resilience.

The review further highlights the growing integration of CRISPR with advanced scientific approaches including artificial intelligence, multi-omics technologies, nanobiotechnology, and precision agriculture. These innovations are improving editing efficiency, target specificity, and large-scale breeding applications, thereby opening new opportunities for sustainable crop improvement. Emerging genome-editing platforms such as base editing, prime editing, and multiplex CRISPR systems are expected to further expand the possibilities for precise and complex trait engineering in seeds.

Despite these promising advancements, several challenges remain, including regulatory inconsistencies, biosafety concerns, off-target mutations, limited transformation efficiency in certain crops, and unequal access to advanced technologies. Addressing these issues through transparent regulatory frameworks, improved biosafety assessment, public awareness, and inclusive innovation policies will be essential for responsible and equitable adoption of CRISPR-based seed technologies.

Overall, CRISPR-driven innovations represent a major breakthrough in next-generation agriculture and seed science. With continued research, technological refinement, and global collaboration, CRISPR technology has the potential to

support sustainable food production, strengthen nutritional security, conserve agricultural resources, and develop climate-resilient cropping systems capable of meeting future global food demands.

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