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CRISPR/CAS-MEDIATED BIOFORTIFICATION OF CROP PLANTS: ADVANCING NUTRITIONAL ENHANCEMENT THROUGH PRECISION GENOME EDITING

Original Article

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ABSTRACT

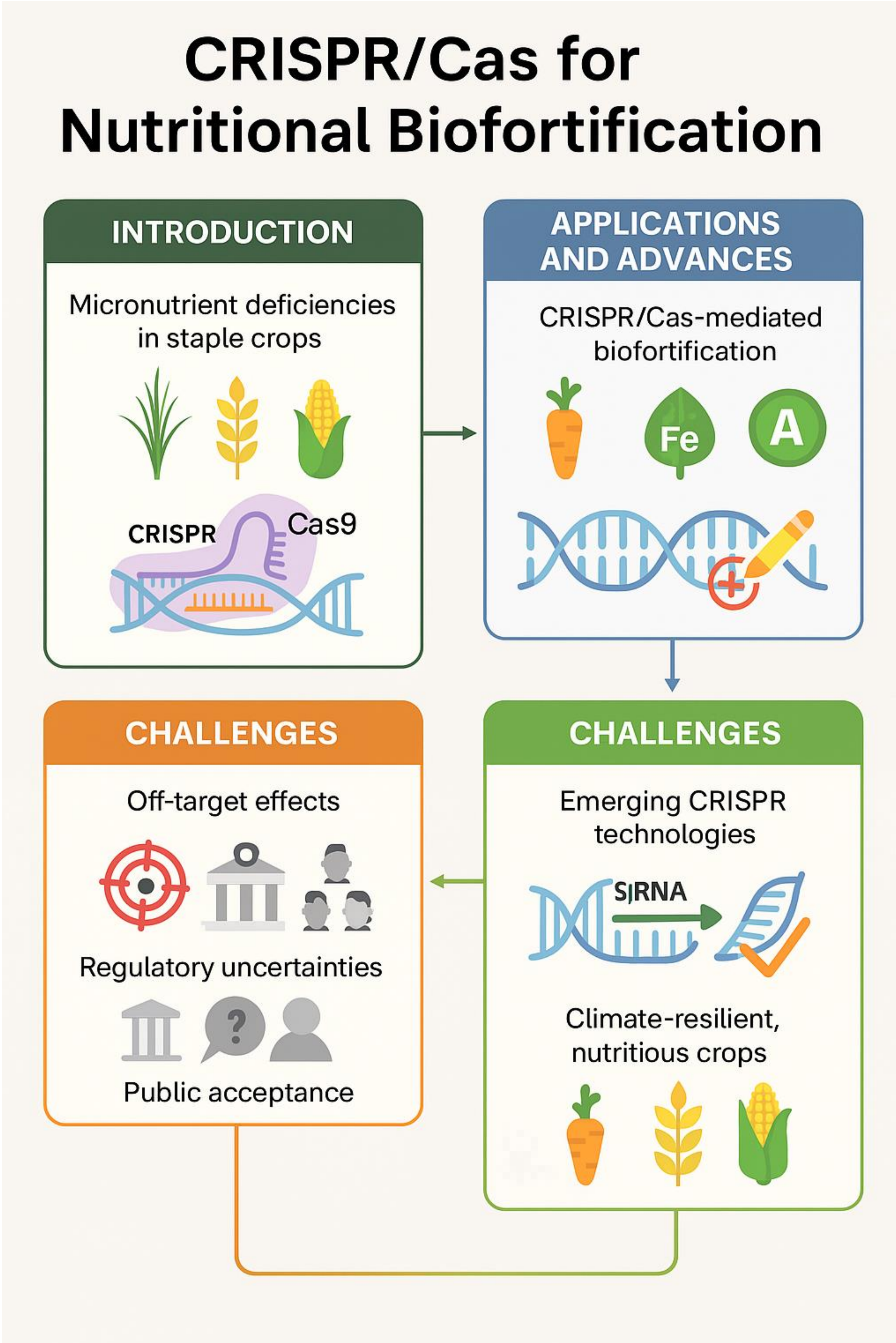
Background: Micronutrient malnutrition, also known as hidden hunger, continues to pose a major global public health burden, particularly in regions where populations rely heavily on staple crops such as rice, wheat, and maize. Traditional methods of biofortification, including selective breeding and agronomic practices, have had limited impact in overcoming the scale and complexity of these deficiencies.

Objective: This narrative review explores the role of CRISPR/Cas genome editing technology as a next-generation approach for the biofortification of staple crops to improve their nutritional quality and address widespread vitamin and mineral deficiencies.

Main Discussion Points: The review outlines the limitations of conventional and transgenic methods and highlights the advantages of CRISPR/Cas systems in enabling precise, efficient, and stable edits in biosynthetic and transport pathways related to iron, zinc, provitamin A, and other nutrients. It discusses key crop-specific applications, emerging CRISPR variants such as base and prime editing, and their potential in climate-resilient crop development. The review also identifies challenges such as off-target effects, regulatory uncertainty, and public perception, emphasizing the need for cross-sectoral solutions.

Conclusion: CRISPR/Cas technology presents a transformative opportunity for global nutritional enhancement. Its successful adoption depends on further interdisciplinary research, robust regulatory frameworks, and proactive public engagement to meet nutritional goals aligned with SDG-2 (Zero Hunger).

Keywords: CRISPR/Cas, Biofortification, Genome Editing, Micronutrient Deficiency, Nutritional Security, Sustainable Agriculture.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INTRODUCTION

Micronutrient malnutrition, often termed "hidden hunger," represents a significant public health challenge affecting billions of people worldwide. Despite advances in food production, many staple crops—especially those consumed widely in low- and middle-income countries—remain deficient in essential vitamins and minerals. These nutritional shortfalls have far-reaching consequences, particularly for vulnerable populations such as children and pregnant women. For example, the World Health Organization (WHO) reports that approximately 250 million children are deficient in vitamin A, placing them at increased risk of blindness and mortality. Similarly, iron deficiency anemia affects 1.62 billion people globally, contributing to fatigue, impaired cognitive development, and increased maternal mortality. Zinc deficiency, another widespread concern, affects more than 2 billion people, compromising immune function and elevating the risk of infectious diseases (El-Ramady et al., 2022). These deficiencies not only exacerbate global disease burdens but also hinder socioeconomic development by limiting educational attainment and workforce productivity. Traditional strategies aimed at reducing micronutrient deficiencies have included supplementation and food fortification programs. While such approaches have shown benefits, their implementation often faces logistical and financial constraints, particularly in rural and resource-limited settings. Moreover, these interventions tend to be reactive and rely heavily on sustained governmental or donor support. In contrast, biofortification—a proactive and sustainable strategy—offers a promising alternative. Biofortification seeks to improve the nutritional quality of crops at the source by increasing the concentration of essential micronutrients through agronomic practices, conventional breeding, or modern biotechnological tools (Avnee et al., 2023). Among these, genetic engineering and more recently CRISPR/Cas-based genome editing has emerged as revolutionary tools to enhance crop nutritional profiles with greater efficiency and precision.

Genome editing technologies, particularly CRISPR/Cas systems, have rapidly transformed the field of agricultural biotechnology. The CRISPR/Cas9 system, derived from a naturally occurring defense mechanism in bacteria, has become a powerful genome editing tool due to its simplicity, cost-effectiveness, and remarkable precision. This system utilizes a guide RNA (gRNA) to direct the Cas9 nuclease to a specific DNA sequence, where it introduces a double-stranded break. The host's natural DNA repair mechanisms then fix the break, either through non-homologous end joining or homology-directed repair, allowing for precise genetic modifications (Liu et al., 2021). Compared to older genome editing technologies such as zinc finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs), CRISPR/Cas9 offers increased targeting accuracy, scalability, and ease of use (Kumar et al., 2022). These advantages make it particularly suitable for applications in crop biofortification, where fine-tuned modifications in specific genes can significantly enhance micronutrient accumulation. Current research has demonstrated the potential of CRISPR/Cas-mediated biofortification across various staple crops. For instance, CRISPR-based editing has been employed to increase provitamin A content in rice by modifying genes in the carotenoid biosynthetic pathway. Similarly, researchers have used CRISPR to enhance iron and zinc levels in wheat and maize by targeting genes involved in metal transport and storage (Tavakoli et al., 2021). The ability to make such precise genetic edits without introducing foreign DNA also makes CRISPR-edited crops more acceptable to regulatory agencies and the public, potentially reducing the societal resistance associated with genetically modified organisms (GMOs). Despite these advancements, certain challenges persist. The long-term ecological impacts, off-target effects, and ethical considerations surrounding gene editing remain active areas of investigation. Additionally, regulatory frameworks for genome-edited crops vary significantly across countries, posing hurdles for global deployment.

While the foundational science and initial applications of CRISPR/Cas in crop biofortification have been promising, gaps remain in our understanding of the broader implications and optimization of this technology. For example, relatively few studies have explored the pleiotropic effects of gene edits on plant physiology, yield, or stress tolerance. Moreover, there is limited data on the stability of enhanced nutrient traits across multiple generations and environmental conditions. Another underexplored area is the synergistic enhancement of multiple micronutrients within a single crop variety—a goal that may be attainable with multiplexed CRISPR approaches but remains technically and biologically complex (Dhaliwal et al., 2022). Additionally, integrating CRISPR/Cas-based strategies with traditional agronomic and breeding techniques could offer more robust and resilient biofortification outcomes, yet interdisciplinary studies in this domain are scarce. The primary objective of this review is to explore the application of CRISPR/Cas genome editing technology in the biofortification of major staple crops, including rice, wheat, maize, and cassava. It aims to elucidate how precise genetic modifications facilitated by CRISPR/Cas can enhance the nutritional value of these crops by targeting pathways responsible for the biosynthesis and accumulation of essential micronutrients such as iron, zinc, and provitamin A. Through an examination of recent case studies and scientific advancements, the review will assess the current landscape of CRISPR-mediated biofortification, highlight successful interventions, and evaluate the implications for food security and public health.

This review will specifically focus on peer-reviewed studies and developments published within the past five years, emphasizing translational research and field-applicable outcomes. It will include both laboratory-based findings and those validated through greenhouse or field trials. Furthermore, the scope encompasses both direct nutritional enhancements (e.g., increased vitamin or mineral content) and indirect contributions such as improved bioavailability and metabolic stability. By integrating data from molecular biology, plant physiology, nutrition, and agricultural sciences, this review seeks to offer a holistic understanding of how CRISPR/Cas technology can be leveraged to address global micronutrient deficiencies. The significance of this review lies in its timely synthesis of emerging research in a rapidly evolving field. As global food systems grapple with the dual challenges of increasing population and declining nutritional quality, there is a pressing need for innovative solutions that can sustainably enhance the nutrient profile of staple foods. CRISPR/Cas-mediated biofortification stands at the forefront of such innovation, offering a scalable, precise, and cost-effective approach to improving global health outcomes. By summarizing current knowledge, identifying research gaps, and outlining future directions, this review aims to provide scientists, policymakers, and agricultural stakeholders with a comprehensive reference point to guide further research and implementation strategies. In doing so, it contributes to the broader mission of ensuring nutritional security and promoting public health through science-driven agricultural development.

Thematic Discussion

The integration of CRISPR/Cas genome editing into crop biofortification has transformed how micronutrient deficiencies are addressed at the source—through precise genetic modifications in staple food crops. This thematic discussion evaluates the major areas of advancement and challenges, focusing on three broad themes: the transition from conventional biofortification to genome editing, molecular mechanisms and tools for CRISPR-based enhancements, and applications across key staple crops. The synthesis of recent studies emphasizes the comparative effectiveness of genome editing strategies and identifies critical research gaps in the field.

From Traditional Breeding to Precision Editing: A Paradigm Shift in Biofortification

Historically, conventional breeding and agronomic interventions served as primary strategies for crop biofortification. These methods, though valuable, often required multiple growing seasons, extensive phenotyping, and were limited by the available natural genetic variation within crop species. For instance, selective breeding increased iron and zinc content in pearl millet and beans, but gains were incremental and environment-dependent (Avnee et al., 2023). Agronomic practices such as fertilizer application enriched micronutrients temporarily, yet lacked sustainability and long-term effectiveness (Koç and Karayiğit, 2022). In contrast, CRISPR/Cas-mediated genome editing allows for targeted, heritable modifications without the need for crossbreeding. This approach dramatically reduces breeding timelines and enables precision targeting of biosynthetic or transport pathways directly involved in nutrient accumulation. In one comparison, Liu et al. (2021) highlighted that, conventional methods for vitamin A biofortification in rice required a decade of development, while CRISPR strategies achieved similar outcomes in less than three years. Thus, genome editing represents a transformative shift in the biofortification paradigm, addressing earlier bottlenecks related to time, cost, and precision.

CRISPR/Cas Systems: Tools for Precision in Nutritional Enhancement

The CRISPR/Cas9 system has become the foundational tool for genome editing due to its simplicity, efficiency, and adaptability across species. Recent work has expanded into newer systems, such as CRISPR/Cas12a and base/prime editors, which offer advantages in specificity and off-target control. For example, Meliawati et al. (2021) demonstrated that Cas12a edits yielded fewer off-target effects when enhancing vitamin E biosynthesis pathways in oilseed crops. Prime editing, a newer approach allowing for precise insertions and deletions without double-strand breaks, was used by Movahedi et al. (2023) to increase folate content in maize with minimal unintended mutations. Saini et al. (2023) reported on the use of multiplexed CRISPR systems to simultaneously edit multiple loci in wheat, resulting in increased zinc, reduced phytic acid, and improved protein content—outcomes previously difficult to achieve through single-gene approaches. This level of genomic precision allows for both the knockout of anti-nutritional factors, such as phytate-producing genes, and the upregulation of biosynthetic enzymes such as PSY1 and CRT1 involved in carotenoid production. The breadth of editing tools and the ability to fine-tune gene expression offer unmatched control over plant nutritional composition.

CRISPR Applications Across Crops: Case-Based Synthesis

A growing body of evidence supports the successful application of CRISPR/Cas systems in enhancing micronutrient content across major staple crops. In rice, multiple studies including those by Dong et al. (2020) and Patel-Tupper et al. (2024) have demonstrated that editing the carotenoid biosynthesis pathway resulted in significant increases in beta-carotene levels. Additionally, CRISPR-induced mutations in iron transport genes such as OsIRT1 and ferritin-related genes have yielded up to 50% increases in grain iron content (Koç and Karayiğit, 2022). In wheat, Sánchez-León et al. (2024) employed CRISPR/Cas9 to enhance protein quality by editing glutenin subunits and puroindoline genes associated with grain hardness. Furthermore, the targeting of TaVIT2 has successfully improved iron bioavailability in wheat grains (Zhang et al., 2021). Such modifications offer dual benefits of nutritional enhancement and improved processing quality, addressing both consumer health and industry needs. Maize has also emerged as a key target for biofortification. Movahedi et al. (2023) showed that folate and beta-carotene biosynthesis genes could be co-edited using prime editors, improving the levels of both nutrients simultaneously. Similarly, Wang et al. (2022) reviewed CRISPR patents and found that a substantial portion focused on yield and quality traits, such as increased kernel number and starch biosynthesis, alongside nutritional improvements like enhanced vitamin E through editing of ZmCYP450 genes.

In cassava, Mehta et al. (2019) demonstrated that CRISPR edits targeting CRT-B and DXS genes boosted beta-carotene accumulation, while knockout of the MeCYP79D1 gene reduced toxic cyanogenic glycosides, thereby improving both nutrition and food safety (Gomez et al., 2023). These dual improvements represent a unique advantage of CRISPR systems—simultaneously improving multiple traits without introducing foreign DNA. Potato and other underutilized crops have also benefitted from CRISPR interventions. Tiwari et al. (2022) reported that editing GBSS gene in potatoes created amylose-free starch, and further studies enhanced carotenoid levels and reduced glycoalkaloid toxicity. In millet, barley, and pulses, early-stage studies indicate improved drought resistance and increased bioactive compounds, such as beta-glucans and iron (Li et al., 2022; Garcia-Gimenez et al., 2020).

Contrasts and Convergences in Biofortification Strategies

A comparative analysis reveals important distinctions in how different crops respond to CRISPR-based biofortification. Cereals like rice and wheat show more predictable outcomes due to their well-characterized genomes and abundant molecular tools. In contrast, root and tuber crops like cassava and potato present more complex editing challenges due to high heterozygosity and polyploidy. However, the advantages of editing non-reproductive tissue traits in vegetatively propagated crops make them attractive targets for nutritional interventions (Kumar et al., 2022). Furthermore, regulatory and societal acceptance varies significantly. Genome-edited crops that do not introduce foreign DNA (non-transgenic) tend to face fewer regulatory hurdles, particularly in countries such as the USA, Japan, and Brazil. In contrast, Europe maintains strict GMO regulations even for CRISPR-edited crops. These disparities affect the scalability and adoption of biofortified crops across different regions, limiting their impact in some high-need areas.

Table 1 Comparison of different variants of CRISPR/Cas systems.

CRISPR Variant	Guide RNA Structure	DNA Edit Type	Precision	Advantages	Applications
CRISPR/Cas9	Standard gRNA	Double-strand break	Moderate	Widely used, easy to design and implement	Gene knockout, targeted mutations
CRISPR/Cas12a (Cpf1)	Simpler gRNA structure	Staggered cut	Higher	Fewer off-target effects, easier gRNA design	Gene editing, multiplexing
CRISPR/Cas13	Specific to RNA	Single-strand break	High	RNA targeting, used for RNA knockdown	Gene regulation, antiviral applications
Base Editing	Requires two gRNAs	No double-strand break	Very high	Precise single-nucleotide changes without DSB	Point mutations in nutrient biosynthesis genes
Prime Editing	Requires two gRNAs and a template	Targeted insertions/deletions	Very high	Highly versatile, precise edits with minimal errors	Complex edits in metabolic pathways

Table 2 Summary of key CRISPR/Cas applications in biofortifying specific crops

Crop	Targeted Nutrient(s)	Edited Gene(s)	Resulting Phenotypic Improvement	Reference(s)
Rice	Vitamin A (Beta-Carotene)	*PSY1*, *CRT1*	Increased beta-carotene content, development of Golden Rice	Kumar et al., 2022
Wheat	Iron	*TaVIT2*	Enhanced iron bioavailability in wheat grains	Connorton et al., 2017
Maize	Zinc, Vitamin E	*ZmIRT1*, *ZmCYP450*	Increased zinc content, enhanced vitamin E levels	Ludwig et al., 2024

Crop	Targeted Nutrient(s)	Edited Gene(s)	Resulting Phenotypic Improvement	Reference(s)
Cassava	Vitamin A (Beta-Carotene)	*CRT-B*, *DXS*	Higher beta-carotene accumulation in cassava roots	Mehta et al., 2019
Potato	Iron, Vitamin C	*FER1*, *GMPase*	Increased iron content, improved vitamin C concentration	Tiwari et al., 2022
Tomato	Folate	*FPGS1*, *DHFR*	Enhanced folate levels in tomato fruit	Khan et al., 2023
Banana	Provitamin A	*LCYB*, *PSY*	Improved provitamin A content	Zhang et al., 2022
Barley	Zinc	*HvZIP*	Improved zinc uptake and accumulation in barley grains	Křenek et al., 2021
Soybean	Omega-3 Fatty Acids	*FAD2*, *FAD3*	Enhanced production of omega-3 fatty acids	Ma et al., 2021
Millet	Iron, Zinc	*FRO2*, *NAS3*	Increased iron and zinc concentrations in millet grains	Kumar et al., 2022

Challenges, Limitations, and Emerging Directions

Despite the promising outcomes, challenges remain. Off-target effects, although reduced in newer CRISPR variants, still raise safety concerns. Additionally, limited access to genome editing tools and regulatory clarity in low-income countries may prevent equitable deployment. Many nutrient pathways involve complex polygenic traits and epigenetic factors, which are not fully understood. Moreover, the integration of CRISPR strategies with conventional breeding and field trials remains underexplored, especially in developing regions. Future research must focus on multiplex gene editing to enable simultaneous enhancement of multiple traits. More robust field data are needed to assess the long-term stability and efficacy of biofortified traits across environmental conditions. In addition, expanding public-private partnerships and investing in capacity-building in genomics and biotechnology will be essential to translate laboratory successes into real-world impact.

Critical Analysis and Limitations

The existing body of literature on CRISPR/Cas-mediated biofortification presents a wealth of promising data, but upon critical evaluation, several limitations and concerns emerge that challenge the robustness and translational potential of the findings. While many studies have reported successful enhancement of micronutrient content in staple crops, such outcomes are often derived from experiments conducted under highly controlled laboratory or greenhouse conditions, limiting their ecological validity. A recurrent limitation across studies is the small scale of experimental design. Most investigations, including those by Liu et al. (2021) and Saini et al. (2023), focus on a handful of edited lines, often lacking comprehensive replication across multiple genotypes or environments. This constrains the interpretation of data and reduces the reliability of conclusions regarding field performance or long-term stability of the edited traits. Another issue arises from the lack of standardized methodologies for assessing the impact of genetic edits. Nutrient content measurements are frequently based on a single developmental stage or tissue type, with minimal attention to how environmental variables—such as soil quality, climate stress, or pest pressure—may modulate nutrient expression. This variability in experimental protocols hinders meaningful cross-comparisons between studies. For instance, beta-carotene enhancement in rice and maize has been quantified using different carotenoid extraction methods, making it difficult to synthesize findings across experiments or establish consistent benchmarks for nutritional gain (Dong et al., 2020; Movahedi et al., 2023).

Moreover, the studies often fall short in terms of long-term follow-up. Most CRISPR-based trials analyze the T0 or T1 generations, providing limited insight into the heritability, stability, and potential pleiotropic effects of the edits across multiple generations. This is especially relevant for traits like micronutrient enhancement, which may be influenced by complex epistatic interactions and may not consistently manifest in diverse agro-climatic zones. Without longitudinal data, it remains uncertain whether the biofortified traits would retain efficacy and consistency under real-world farming conditions. Methodological bias also permeates the literature. Many studies do not incorporate proper controls or blinded assessments, increasing the risk of performance bias. In some cases, the choice of genes for editing is heavily skewed toward well-characterized biosynthetic enzymes, neglecting broader systems-level interactions that may also impact nutrient bioavailability or plant fitness. This reductionist approach could lead to overestimation of benefits, as other interconnected pathways may counteract or mask the intended outcomes. For example, editing a zinc transporter gene may increase zinc concentration in grains, but could also inadvertently alter plant growth or resistance profiles—variables not often reported in CRISPR biofortification trials (Koç and Karayiğit, 2022; Kumar et al., 2022).

An additional challenge is the potential for publication bias within the field. The enthusiasm surrounding CRISPR and its revolutionary potential may inadvertently contribute to the underreporting of inconclusive or negative findings. Studies that fail to achieve significant enhancement or encounter off-target complications may be less likely to be published, skewing the literature towards positive outcomes. This selective reporting restricts a balanced understanding of the technology's true efficacy and safety profile. A meta-analysis of the literature would be difficult to perform accurately due to this asymmetry in data availability. The variability in measurement outcomes further complicates synthesis of the existing evidence. The methods used to quantify iron, zinc, or carotenoid levels vary not only in analytical technique but also in how outcomes are expressed—whether as dry weight concentration, bioavailable fraction, or relative increase from control. These inconsistencies challenge efforts to directly compare the impact of genome edits across different studies and crops. Additionally, most studies do not evaluate actual nutrient bioavailability in human or animal models, which is critical for assessing real-world nutritional outcomes.

Perhaps one of the most significant limitations lies in the generalizability of the current findings. The majority of published research originates from high-resource laboratories, often using elite or model crop varieties with well-sequenced genomes and established transformation protocols. This limits applicability in genetically diverse landraces or crops of regional importance in nutritionally vulnerable regions. For example, while substantial progress has been made in rice and wheat, limited studies address millets, sorghum, or other climate-resilient crops crucial to food security in sub-Saharan Africa and South Asia (Li et al., 2022). Additionally, socio-political factors such as regulatory uncertainty and public perception of gene-edited foods may further restrict the translation of laboratory success into large-scale agricultural adoption. To move the field forward, future studies must embrace larger, more diverse study designs with robust field trials, standardized nutrient quantification protocols, and multi-generational assessments. Interdisciplinary approaches that integrate genomics, agronomy, nutrition science, and socioeconomics are necessary to develop CRISPR-based biofortified crops that are not only nutritionally effective but also agronomically viable and publicly acceptable.

Implications and Future Directions

The integration of CRISPR/Cas technology into crop biofortification marks a paradigm shift in addressing micronutrient malnutrition through agricultural innovation. This review underscores the transformative potential of genome editing in enhancing the nutritional value of staple crops, offering significant implications for public health, agricultural policy, and global food security. The findings emphasize that while CRISPR/Cas systems offer unmatched precision and efficiency in nutrient trait enhancement, translating these laboratory-based advancements into practical, field-ready solutions demands comprehensive policy frameworks and continued interdisciplinary research. From a policy standpoint, there is an urgent need for the development of globally harmonized regulatory guidelines that distinguish between traditional GMOs and genome-edited crops produced through CRISPR, particularly those that do not incorporate foreign DNA. Current regulatory ambiguity, especially in regions like the European Union and parts of Africa, creates barriers to the dissemination of CRISPR-biofortified crops in areas where they are most needed. National and international policymakers must recognize the critical role of gene editing in meeting the nutritional targets of Sustainable Development Goal 2 (Zero Hunger) and should establish evidence-based frameworks to accelerate responsible innovation, promote equitable access, and ensure food safety (Zaidi et al., 2020; Sami et al., 2021). Simultaneously, public engagement initiatives should be fostered to build trust in CRISPR-derived products, ensuring transparency and inclusivity in decision-making processes.

Despite the substantial progress in CRISPR-based biofortification, several unanswered questions remain that warrant further investigation. One major gap lies in understanding the long-term stability and ecological impacts of edited traits under field conditions across multiple seasons and environments. Most current studies are limited to early-generation lines and do not provide data on trait persistence or potential unintended consequences over time. Moreover, the interaction between edited genes and native metabolic pathways remains inadequately explored, particularly for traits involving polygenic inheritance such as protein quality or multi-nutrient enrichment. A deeper mechanistic understanding of pleiotropic effects, gene-environment interactions, and potential metabolic trade-offs is essential to ensure the durability and biosafety of nutritional gains (Guo et al., 2023; Rai et al., 2023). Another critical gap is the limited focus on orphan and underutilized crops that serve as dietary staples in many nutritionally vulnerable regions. Most CRISPR research has concentrated on globally traded crops such as rice, wheat, and maize. Expanding the application of genome editing to crops like sorghum, millet, teff, and pulses would help diversify dietary sources of micronutrients and reduce reliance on a few major crops. In parallel, there is a need for bioavailability studies that validate whether the enhanced nutrient content in edited crops translates into measurable health improvements in humans and livestock, particularly in populations affected by malnutrition. Such translational research would support health-economic evaluations and inform nutrition-sensitive agriculture policies.

To strengthen the evidence base, future studies should adopt robust experimental designs, including multi-location field trials, multi-generational analyses, and large-scale omics-based assessments of unintended effects. Randomized controlled field trials comparing CRISPR-edited and conventionally bred lines under real agronomic stress conditions would provide high-quality data to inform regulatory approvals and public health recommendations. Additionally, interdisciplinary studies involving agronomists, molecular biologists, nutritionists, and social scientists are essential to evaluate the socio-cultural acceptability, economic feasibility, and dietary impact of these crops in diverse contexts (Shaheen et al., 2023). As next-generation editing tools—such as base editors, prime editors, and epigenome modifiers—continue to evolve, their integration into biofortification pipelines opens new possibilities for precision enhancement without genomic disruption. Temperature-tolerant Cas variants and inducible systems further extend the utility of CRISPR tools to diverse agro-climatic settings, enabling scalable interventions even under challenging environmental conditions (Zaidi et al., 2020; Rai et al., 2023). These innovations not only advance technical capabilities but also call for updated regulatory perspectives that account for the nuanced differences between editing techniques. Ultimately, the path forward in CRISPR-mediated biofortification must be guided by a balance of innovation, equity, and responsibility. As genome editing becomes more accessible and democratized, global collaborations will be pivotal in sharing technology, resources, and regulatory expertise to ensure that the benefits of this transformative science reach the most food-insecure regions. The potential to simultaneously enhance crop nutrition, yield, resilience, and safety offers a holistic solution to pressing global challenges such as malnutrition and climate change. Through well-coordinated policy action and sustained research investment, CRISPR/Cas biofortification can evolve from a scientific breakthrough into a practical tool for global health and development.

CONCLUSION

CRISPR/Cas-mediated biofortification represents a transformative advancement in agricultural biotechnology, offering precise, efficient, and sustainable solutions to address global micronutrient deficiencies through targeted genome editing of staple crops such as rice, wheat, maize, and cassava. The reviewed literature provides compelling evidence that CRISPR can significantly enhance the nutritional profiles of crops by increasing essential vitamins and minerals like iron, zinc, and provitamin A, while also offering resilience to environmental stresses. However, while the strength of current findings is promising, much of the evidence remains limited to controlled environments and early-generation studies, highlighting the need for broader, long-term, and field-based validations. Regulatory inconsistencies, public skepticism, and technical limitations—such as off-target effects and the complexity of editing polygenic traits—remain key barriers. Researchers are therefore encouraged to prioritize interdisciplinary collaborations, develop robust and transparent methodologies, and explore underrepresented crops and regions. As CRISPR continues to evolve with next-generation tools and climate-adaptive applications, its alignment with global health objectives, particularly the United Nations Sustainable Development Goal 2, positions it as a pivotal strategy in combating hidden hunger. Continued investment in translational research, public engagement, and harmonized policy frameworks will be essential to unlock its full potential in achieving nutritional security worldwide.

Author Contributions

Author	Contribution
Maria Latif*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Muqaddas Mustafa	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published

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