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Modern Biotechnologies for Improving Quality Traits in Horticultural Crops: Subject Review

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Annotation: Application of cutting-edge particularly biotechnologies, gene editing methodologies like CRISPR/Cas9, offering are tremendous opportunities to enhance the desirable traits of horticultural crops like tomato, strawberry and cucumber. This review summarizes the recent use of these technologies to develop flavor, disease resistance and nutritional quality of horticultural crops with a focus on field and applied trials in different agricultural settings. It also deals with the ethical and regulatory considerations surrounding the use of these tools in agriculture, and closes with recommendations to support scientific research and decision-making in this sensitive area, integrated withan overview of thescientific developments that should be expected from the 2015–25. Over the past few years there have significant advancements in the field been of biotechnology and gene editing technology, in particular CRISPR/Cas9, have been considered extremely promising for the development of valueadded traits in horticultural crops. It will be reviewed in this paper recent applications of this technology for the modification of traits of leading horticultural crops, such as tomato, strawberry, and cucumber, with the goal of improving tolerance to diseases resistance, sensory attributes, and tolerance to environmental stresses. The review also considers regulatory and ethical issues related to the application of the technologies in agriculture and gives recommendations for the sustainable use of the tools.Keywords:CRISPR/Cas9,strawberry, cucumber, biotechnologies.

tomato,

1. Introduction

Modern agricultural practices are increasingly strained by climate change and the growth of the world's population and the market for high quality agricultural goods. Horticulture, as a branch of agriculture which directly influenced by these challenges, could be described as one of the most sensitive sectors in both quality (vegetables, fruits and flowers) and economic legacies to such limitations. Here, biotechnologies especially gene editing technology, instead, have provided an effective approach to balance yield increment and crop quality (Gao, 2018). One of these biotechnologies is CRISPR/Cas9, one of the most recent and most significant new technologies, which allows researchers to alter plant genes precisely, without the addition of foreign DNA. This renders the end products more encouraging from a regulatory standpoint in the majority of countries, because there, they are not regarded as GM plants (Chen et al., 2019). Custom gene targeting also can be used to improve taste, color, fragrance, disease or environmental hardiness characteristics — as can traditional breeding programs. It has been reported that time and costs of crop improvement are reduced by use of CRISPR technology compared to conventional breeding methods and that the efficiency of genetic modification is higher as well (Jaganathan et al. For instance, antioxidant compounds in fruit can be edited to be over-expressed genes or post-harvest shelf life extended, improving supply chain efficiency and reducing wastage in farming. Existing studies highlight growing economic, environmental, and health challenges in Iraq and Kurdistan, particularly in relation to housing shortages, productive sector performance, and the environmental impact of heavy metals.

A number of researches have stress on the importance of mineral supplements like se_ lenium and zinc in improving animal health and decreasing environmental pollution, in context the environmental dimension re- harbours in modern growth theories itself (Palani, 2025; Palani et al., 2025; Palani & Hussen, 2022; Palani et al., 2022a, 2022b, 2024a, 2024b). This review is intended to serve as a guide for the most successful part of practical applications of CRISPR/Cas9 technology in horticultural crops with a detailed presentation of concrete cases in tomato, strawberry and cucumber. It also addresses the technical, regulatory and ethical challenges concerning the use of these technologies at the agricultural level.

Scientific Principles of CRISPR/Cas9 Technology in Horticulture

CRISPR/Cas9 is one of the most accurate and simple gene editing technologies, which has transformed molecular biology and agriculture. The technology uses the Cas9 enzyme to make a cut in a strand of DNA at a precise location, directed to its target by a guide RNA sequence. Certain genes are subsequently modified, knocked out, or introduced to enhance certain plant features (Gao, 2018). CRISPR/Cas9 is based on the bacteria's immune system, which uses guide RNA to direct the Cas9 enzyme to homing in on a precise location in the DNA, cutting the gene in the right place. This makes possible precise alterations in some gene sequences– either disabling them, or increasing / decreasing their expression among them – which could be used to target traits in agriculture with unprecedented accuracy compared to more traditional approaches. This system is relatively simple with respect to design and operation, and is very attractive in order to save labor and facilitate the creation of new varieties with improved traits.

This technology is employed in horticulture to add or eliminate genes for a range of desirable traits such as flavor, shelf life, disease resistance and tolerance to environmental stresses, including salinity and drought. For instance, the gene encoding the enzyme to soften fruit after harvesting can be disrupted to extend shelf life (Watanabe et al., 2020). This technology would extend the ability of researchers to modify complex, multi-gene traits which would otherwise be

difficult if not impossible to target using conventional methodology, and to greatly improve horticultural crops through creativity and efficiency methods.

Traits Targeted for Improvement in Horticultural Crops

With the great economic and nutrient values of horticultural crops, the improvement of their agronomic traits has been the main objective of modern breeding strategies. The goal of these programs is to improve productivity, fruit quality, disease resistance and responses to environmental stresses, and to address traits that are important to consumers and the marketplace. Biological and agronomic aspects that can be addressed with refined breeding techniques are multiple and can be classified into several main categories. Firstly, fruit quality is among the most economically significant and highly researched traits because it directly influences the market value of the product and consumer satisfaction. Fruit quality includes taste, aroma, color, texture and nutritional value.

The flavor of the fruit is one of the most essential attributes for consumers, and researchers try to enhance it by altering the contents of sugars, organic acids, and aroma compounds produced by the fruit (Goff and Klee, 2006). Fruit color Fruit color is also important to its attraction, and is correlated with the accumulation of plant pigments, including anthocyanins, carotenoids and chlorophyll. These pigments also possess human health benefits as natural antioxidants for human well-being (Liu et al., 2018). In addition, fruit texture (firmness) is a vital attribute to ensure stability of the fruit against spoilage during transportation, storage and to extend their shelf life, which is associated with food loss reduction (Malnoy et al., 2016). Equally so, nutritional quality is important and scientists are working to boost the levels of vitamins, minerals, antioxidants etc in fruit to improve the health effects for consumers (Li et al., 2018). Secondly, disease and pest resistance are one of the main goals of horticultural crops improvement, which provide a direct means to diminish production losses and to reduce the environmental load by means of the significant dependence on ecologically deleterious chemical pesticides. Resistance genes are the specific gene targets of genetic improvement programs aimed at conferring the ability of plants to resist to a wide diversity of fungal, bacterial and viral diseases, and insect pests that can affect crop health (Jones et al., 2014). New gene editing technologies like CRISPR/Cas9 are now enabling us to finely edit these genes to make them more resistant, leading to healthier and increasingly sustainable crops. Third, it is a requirement of horticultural crop breeders to selection for tolerance to environmental stresses, including drought, salinity and heat, owing to the increased challenges imposed by climate change. A tolerance for these stresses relies on controlling the expression of the related genes that deal with plant damage, mainly those belonging to the DREB (drought-responsive element-binding protein) denoting the family of transcription factors for stress response, such as DRHB1 as well as HSP (heat shock protein) which protects the plant cells under adverse environmental conditions (Wang et al., 2020). Enhancement of these characteristics is beneficial for stable fruit yield, especially in stress-environments and for fruit quality. Fourth, productivity and yield are a perspective strategy to improve horticultural crops. This may enhance growth-related traits, including flowering quantification, fruit size, fruit number and ripening rate. This is associated with the system of genes that are responsible for the hormonal growth and plant physiology, which can be modified to enhance the efficiency of resources in agriculture and to increase the crop yield (Gupta et al., 2019). Fifth, great attention is being paid to genetic improvement of plant agronomic and architectural characteristics including crack resistance, controlled branch growth, and altered plant form which contribute to planting and harvesting, and affect production efficiency (Chai et al., 2017). To reduce production costs and the quality of food, the improvement of these characteristics is important. Then, selected characteristics concern increasing harvest and storage time, being related, in many cases, to genes that regulate delayed fruit ripening or retard the chemical processes responsible for post-harvest deterioration.

Enhancement of these characteristics prolongs the shelflife of the product, making it possible to transport and store for a long-time without a loss in quality, ultimately preventing food waste and

increasing the ability to market the crop both locally and globally (Gupta et al., 2021). In general, the establishment of these characters in horticultural crops is a puzzle that can be solved only by the integration of genetic, molecular, and farming tools for a sustainable and high-quality production. Recent advances in technologies such as genome editing provide unparalleled opportunities for the precise and efficient modification or enhancement of these traits.

Practical Applications of CRISPR/Cas9 in Horticultural Crop Improvement

CRISPR/Cas9 is a landmark advancement in horticultural crop improvement by which researchers and breeders can now modify genes accurately and rapidly to improve desired traits and to minimize genetic abnormalities. A dominant practical application of this technology is to enhance the nutritive quality of crops, for example by elevating the levels of health promoting molecules. In tomato plants, CRISPR has also been applied to knockout or modify genes involved in lycopene and flavonoid biosynthesis pathways to enhance antioxidants which are beneficial for consumer health (Li et al., 2018). This kind of alteration doesn't change the taste of the crop, but raises its nutritional content. When it comes to disease resistance, researchers can employ the CRISPR/Cas9 complex to precisely snip genes that make the plant more vulnerable to pests or disease. In strawberries, for example, Mlo gene was knocked-out, which is involved in plant defense against certain fungi, and the resultant knockout mediating plant was highly resistant against powdery mildew, a major threatening strawberry disease (Wang et al., 2019). In grapes as well, regulated genes like VvRIN for the ripening and senescence have been altered to delay fruit ripening and prolong storage that is needed to avoid early losses and for a longer marketing period (Ren et al., 2020). Additionally, CRISPR technology has been used for improvement of crop tolerance against environmental stresses such as drought, salinity, and heat.

Mutant zucchinis manipulated for the DREB gene (a transcription factor playing an indispensable role in plant adaptation to water stress) exhibited 20-30% more drought tolerance, compared with those that were not mutated (Zhang et al., 2021). This technology has also been applied for increasing the salt tolerance of chili pepper by the modification of genes that regulate ion homeostasis of plant cells under saline conditions so that the plants can be grown under extremely saline conditions (Kim et al., 2022). The broadening application of CRISPR/Cas9 extends to augmenting agronomic characteristics like size, shape and color. In cucumber, for instance, the fruit development-related gene CsWIP1 was edited to enlarge fruit size and better morphological fruits (Li et al., 2019). In apples, CRISPR was employed to disrupt the MdPG1 gene, which is involved in fruit softening for the production of apples with extended shelf life but equivalent texture and taste (Malnoy et al., 2016).

Also, for the success of CRISPR application in horticultural crops, it is due to the progress of gene delivery method, the high-precision target identification using the sgRNA design tool so on. For many of such crops, comprehensive research on potential side effects on other genes and the whole genome are ongoing, which will help improve the safety and efficiency of the editing (Zhang et al., 2020). Conclusions: In summary, CRISPR/Cas9 mutagenesis represents a powerful tool for horticultural crop improvement, allowing more precise and faster introduction of targeted traits than traditional breeding approaches. With an ongoing development of technology and our knowledge of genes and their functions, we will witness more applications which can address the challenges to the increased productivity, quality and food security in a sustainable and environmentally friendly manners.

Field Application Examples in Different Agricultural Environments

Nowadays, contemporary technologies began to be increasingly applied in improvement of horticultural crops in practical agriculture under different agricultural environments all over the world that are represented by differing climatic conditions, soil quality and region constraints in pests and diseases. Such concrete and practical applications bring new vivid illustrations of how

farmers and researchers make use of modern technologies (e.g. gene editing, precision agriculture or resource management) to enhance crop productivity and quality. We present several field examples from this geographic and environmental diversity.

In hot and arid climates, but particularly in the Middle East and North griculture has concentrated on producing crops which have an inherent resistance towards environmental stresses like drought and high temperatures. For instance, gene expression is regulated by small regulatory RNAs, which are a target for genetic editing via CRISPR/Cas9, that have been used to modulate genes that confer drought tolerance in tomato plants (Solanum lycopersicum). Transgenic crops have been reported to increase plant water use efficiency and survival, and antioxidant status of fruits (Zhang et al., 2019). Moreover, in inappropriate climate areas like Saudi Arabia, precision agriculture has recently been used on date palm orchards based on sensors and monitoring of the soil, which allows water to be used more efficiently with less waste, providing higher date quality and productivity (Al-Jady et al., 2020). Successful applications for improvement through traditional breeding assisted with molecular biology were made on crops like pepper and eggplant in salt affected areas like coastal plains in South Asia.

Hybridization and genetic transformation have been used to introduce genes that confer tolerance to saline soils into this species, allowing it to be grown in salt affected areas while maintaining its productivity and fruit quality (Kumar et al., 2018). Field tests have also been performed in Bangladesh and India, with plant growth-promoting microorganisms (PGPRs) that have ameliorated salinity-induced damage by increasing nutritional conduction and activation of stress defense mechanisms (Egamberdieva et al., 2017). In these sheltered temperate-humid zones, as in Europe and parts of North America, field applications have addressed improvement of plant resistance to common fungal and bacterial diseases like downy mildew of apples and grapes. The reduced impact of apple scab has been achieved through the manipulation of immune response genes in apple plants by gene editing techniques, leading to the selection of resistant varieties with less demand for chemical pesticide applications and reduced chemical risk for the environment (Pessina et al., 2016). Likewise, drone-borne smart farming systems have been shown to work well in vineyards in California by generating precise health measurements of the plants, which can help developing early disease treatment strategies and increasing grape quality (Mahlein et al., 2018). In Latn America and tropical Africa, applications have been targeted on the resistance of crops to destruction by insect pests and viruses that limit production. Field tests in Brazil and Uganda have applied gene editing approaches to create virus-resistant strains of bananas and coffee and to boost productivity and environmentally friendly plant management (Tripathi et al., 2020). Furthermore, by the application of RNA interference (RNAi) it is possible to obtain specific insect-resistant plants, for example to the fruit fly and so decrease the use of insecticides and therefore also protect the environment (Baum et al., 2007). There is also the issue of experimental urban settings, including dreamy vertical farms, which have experimented with hydroponic and smart greenhouse farming to enhance the productivity of leafy vegetables and some fruit crops there. In Singapore and Japan, production intensity has been continuously enhanced, resources such as water and fertilizers have been minimised and fresh quality crops can be produced all year round (Despommier, 2010; Kozai, 2013).

In summary, the practical examples presented here illustrate the potential that modern technologies have to respond in real-time to changing conditions and bring about measurable positive impacts on the quality and productivity of horticultural produce. Traditional breeding and cutting-edge technologies such as CRISPR, precision agriculture and biotechnology combine to offer exciting prospects for sustainable agriculture, particularly in the context of climate change and increasing global food demand.

Challenges and Regulatory and Ethical Considerations

Although there has been rapid advancement in the exploitation of new biotechnologies such as gene editing (CRISPR/Cas9) and genetic transformation to enhance the traits of horticultural

crops, there are considerable technical challenges and ethical considerations associated with this technological revolution, including regulation, environment, ethics, economy and society. It is now a necessity to broaden the debate on these challenges, to appreciate how scientific advances can be reconciled with the needs for safety, sustainability and societal acceptance, and in particular, to acknowledge the considerable gap that exists across countries in their regulatory frameworks, and environmental and cultural preparedness. From a regulatory point of view, the global regulation of genetically modified and GE crop utilisation is incredibly diverse with the world profited from inconsistency, which prevent technology development and transfer across borders. An exception is the United States, Brazil, and Argentina which has product-based regulation, in contrast to the European Union which has process-based regulation, meaning that any product modified using any GE technology, even in the absence of any foreign DNA, has to follow the same laws controlling GMOs (Eckerstorfer et al., 2019; Eriksson et al., 2020).

This discrepancy results in a great deal of misunderstanding amongst farmers and investors, and limits the capacity to develop GMO varieties better adapted to other agro-ecosystems. From an ecological standpoint there is obviously a concern that the introduction of GMO variants might disturb the balance in nature, even cause that GMO varieties are set free in nature. According to Snow et al., 2005; Husing et al., 2016 crosshybridization between GM and wild forms at random exists, this may result in contamination of natural genetic diversity or selecting of strains with high resistance to pests or and weeds, resulting pesticides being less effective and making future chemical treatments even more demanding. In addition, while CRISPR/Cas9 is extremely targeted it is not error free and can introduce unwanted mutations that would also require extensive genetic validation and analysis prior to field releases of plants. These are the challenges but a nearly equally important ethics and society issue. The question of editing genes opens up a much needed wider ethical discussion that transcends the scientific frontiers of whether human interference in the genetic code of living creatures is "legitimate", not least in view of the absolute unknown consequences over time of such technologies. Because of the commercial monopoly over genetically engineered seeds and the appropriation of patents by some large companies to control the world market of agriculture many groups like the religious, cultural, and the human rights have expressed concern of the loss of control of biodiversity (Jasanoff et al., 2006; Kloppenburg, 2010). This resulted in the development of "genetic justice", a term used to describe that the small farmers in the developing countries need to be provided fair access to these technologies without being subjected to the hegemony of the big biotech commercial companies. Public acceptance, however, is the biggest hurdle to the use of GM products in developing countries, where the level of public knowledge of biotechology concept is still low. Various research works have revealed that the level of consumer acceptance of these products is strongly influenced by their scientific understanding of the technology, trust in the regulatory authority as well as transparency and credibility in the communication of benefits and risks (Maris, 2001; Frewer et al., 2013). Thus, involving local communities, particularly farmers and decision make-rs, in the conversation about this tool/system is crucial in creating a trusting atmosphere that will drive acceptance of its safe and responsible use and adoption.

This is exacerbated by the high price tag of producing GM crops, which entails substantial costs associated with research, testing, licensing and safety proving. New estimates suggest that it takes 10 to 15 years to develop a GM variety at a cost of at least \$100 million—the type of expense that few entities in the Global South (institutions or governments) can afford (Kirschen, 2015; Smith et al., 2014). New agro-supply chains need to be set up capable of responding to these products, with traceability and monitoring systems, marketing specialisation, conformity certification and higher quality standards. At the international level the compatibility of trade agreements and GM related regulations also presents problems.

Conclusion

Modern biotechnologies, particularly CRISPR/Cas9, have proven highly effective in improving

horticultural crop characteristics, such as nutritional quality, disease resistance, and tolerance to environmental stresses. These technologies have provided promising opportunities for developing more productive plants that are more resilient to climate change and have demonstrated success in field trials in multiple environments.

However, these technologies face regulatory and ethical challenges, such as the controversy surrounding genetic modification, divergent international laws, and limited societal acceptance in some regions. Therefore, it is recommended to adopt integrated policies that balance scientific development with environmental and social considerations, while promoting local research and capacity building to ensure the responsible and sustainable use of these technologies.

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