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Review

CRISPR-Cas Genome Editing for Insect Pest Stress Management in Crop Plants

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Abstract: Global crop yield and food security are being threatened by phytophagous insects. Innovative methods are required to increase agricultural output while reducing reliance on hazardous synthetic insecticides. It appears to be quite effective at reducing production costs and boosting farm profitability to use the ground-breaking CRISPR-Cas technology to create plants that are insect resistant. In contrast, this new technique can modify an insect's genome to either produce gene drive or get beyond an insect's tolerance to various insecticides. This paper reviews and critically discusses the use of CRISPR-Cas genome editing technology in long-term insect pest management. The emphasis of this review is on the prospective uses of the CRISPR-Cas system for insect stress management in crop production by creating genome-edited crops and insects. The potential and difficulties of using CRISPR-Cas technology to reduce pest stress in crop plants are critically examined and discussed.

Keywords: CRISPR-Cas technology; pest management; plant stress resistance; insect resistance

1. Introduction

By directly feeding on crops and disseminating plant diseases, insects are the main biotic stressors that constitute a serious danger to crop losses globally [1]. Annual crop destruction by insects is thought to be about one-fourth of the crop [2]. New management strategies for phytophagous insect pests are needed for increasing crop productivity for ensuring the global food sucirity. Sap-sucking and crop-chewing pests are the main insects responsible for large drops of agricultural productivity [3]. Solutions to the issues are provided by recent developments in the molecular basis of interactions between insects and plants and biotechnological methods, such as genome editing [4]. Recently, it was discovered that designed nucleases have enormous potential for genome editing in both plants and insects [5, 6]. The application of genome editing methods has drammatically grown over time. In actuality, the CRISPR-Cas approach of genome editing is the one that is most frequently utilized right now [7, 8]. Genome editing using the CRISPR-Cas system has proven successful in creating a variety of agronomic traits, including long-lasting resistance to insect pests [5].

A method of genetic manipulation known as genome editing or gene editing involves inserting, deleting, labeling, changing, or substituting DNA into the genome of a living being in order to generate the desired attribute [9]. The four main categories of sequence-specific nucleases for gene editing to date are mega nucleases, zinc finger nucleases (ZFNs), transcription activator-like effector nucleases (TALENs), and clustered regularly

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interspaced short palindromic repeats (CRISPR)/CRISPR-associated protein (Cas) [10, 11]. CRISPR-Cas RNA-guided nucleases developed from bacterial innate immune mechanisms generated from Type II CRISPR-Cas mechanisms are the latest and most advanced genome editing technique [12].

In order to secure food supplies for the world's expanding population and fulfill Sustainable Development Goal 2 (zero hunger), contemporary agriculture practices using stress-resistant crops and genetically modified crops are given emphasis. The use of synthetic chemical insecticides to control insect pests in crop production is expensive, hazardous for humans and the environment. Additionally, it has a bad impact on biodiversity and unintended insects. We have already observed application of numerous insect resistance genes in genetically modified crops, such as *Bacillus thuringiensis* Bt-ICPs, which have had a significant influence on productivity and sustainability [13]. A powerful method for creating insect-resistant plants to further sustainable agriculture is CRISPR-Cas gene editing. By using this potential technique, it is possible to generate insect resistance by altering the effect of target interactions, eliminating host-susceptible genes, decoupling the antagonistic activity of defense hormones, and other methods [14]. CRISPR-Cas gene editing has been effectively used over the past ten years to create some insect-resistant plants or modify a number of insects. This approach has been demonstrated great promise for increasing crop output through the sustainable management of insect pests. Insect pest control in agriculture could be benefitted from the development of genome-edited agricultural plants and insects. This review aimed to summarize recent developments in the use of CRISPR-Cas in generation of insect-resistant plants as well as application of this revolutionary technology for modifying the genomes of phytophagous insects. We also discussed the difficulties and potential use of the CRISPR-Cas toolbox for sustainable management of insect pests.

2. CRISPR/Cas9 and its Mechanism

Clustered Regularly Interspaced Short Palindromic Repeats, or CRISPR, is an abbreviation, and the CRISPR linked protein is called Cas. It is a built-in defense mechanism that prokaryotic organisms like bacteria (45%) and archaea (84%) have in their genomes [15]. A specific DNA portion is targeted, a precise cut is made at the target site, and the gene is rendered inactive or a different version of the gene is substituted. In 2012, Jennifer Doudna and Emmanuelle Charpentier came up with an idea, these two women received the Nobel Prize in Chemistry in 2020 for developing a toolkit to utize the CRISPR-Cas system for editing the genome of any organisms [16]. The CRISPR-Cas9 editing method requires DNA to match a single RNA guide (sgRNA), and is one of several types that make up this technology [17]. The two primary components of the CRISPR/Cas system are the guide-RNA and Cas protein. The Cas9 protein, a nuclease enzyme commonly referred to as molecular-scissors, is responsible for cutting DNA. Guide-RNAs are molecules that direct Cas9 to our chosen spot in the genome where it will remove the existing sequence and replace it with the new one [18]. It has become a very effective, fast, and speedy genome editing tool recently [19, 20]. This CRISPR-Cas technology can now be used to edit multiple genes at a time and even a single base or epigenetic editing.

Several CRISPR-Cas applications have been described to change the DNA sequences of the insect or plant genome [21, 22, 23]. *Streptococcus pyogenes* (Sp) is the source of the Cas9 protein that is currently most frequently employed [24]. In this procedure, a Cas9 protein-associated single-guide RNA (sgRNA) cleaves a particular target DNA region next to a protospacer adjacent motif (PAM), triggering the cellular DNA's repair system to create a double-strand break (DSB). Without the homologous repair template, error-prone non-homologous end-joining (NHEJ) pathways are activated, resulting in spontaneous insertion/deletion or even replacements at the DSB site, which typically disrupts gene function. On the other hand, error-free homology-directed repair (HDR) mechanisms are activated leading to mutations that undertake precise gene alteration,

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including knock-in, knock-out, or mutation, if the donor DNA templates are available that are similar to sequence surrounding DSB site [25]. The NHEJ and HDR have currently been successfully co-opted for genome editing in a variety of insects and plants [14, 22, 26]. Following a successful genome modification, the CRISPR-Cas9 construct is introduced into plant cells using particle bombardment or *Agrobacterium*-mediated transformation techniques, and into insect embryos using microinjection, transfusion, or electroporation-mediated transformation techniques, with the goal of regenerating transgenic species with desired traits [14, 22, 27]. The work flow of CRISPR-Cas genome editing in plants and insects are briefly illustrated in Figure 1.

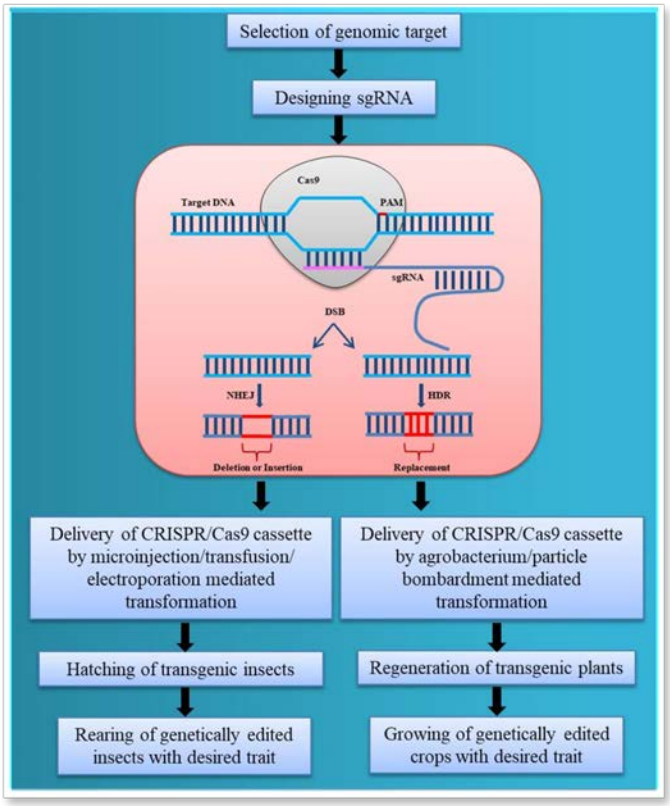

Figure 1: Workflow for CRISPR-Cas9-based genome editing in insects and plants for insect resistance.

3. CRISPR-Cas Genome Editing in Agriculture for Managing Insect Pests

Biotechnology plays a critical role in the management of insect pests to safeguard crops and increase yields, from breeding for pest resistance to the introduction of new genes through genetic modification [28]. The use of genome editing techniques to create insect-resistant plants is still in its early stages. By modifying the genes of both plants and insects, genome editing can be used to manage insect populations. Crop pests can be controlled by inducing sterility in insect pests, interrupting pesticide resistance, or creating de novo resistance if adequate R-genes are lacking. Using CRISPR-Cas9 genome editing technology, novel research is being done to modify insects to prevent them from feeding and injuring plants, and to modify plants to increase their efficacy in repelling insects [22, 29]. In this way, the Genome Editing Platform has opened up new possibilities for creating designer plants, particularly when a targeted deletion is likely to result in elite and superior traits or to start a gene drive that spreads mutations that are responsible for the lethality of female insect populations.

The agricultural biotechnology sector has been threatened by the problem of insect resistance to the Bt trait, thus biotech companies are searching for novel, economically

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viable, and environmentally responsible solutions to the problem. In the biotech sector, CRISPR-Cas9 gene editing has emerged as the leading method for controlling insect pests [30]. In order to successfully alter a gene's function, genome editing technology actually leverages the cell's own internal processes. Genome editing makes sure that the DNA sequence of a specific target genome is altered via the addition, deletion, and/or substitution of DNA bases [31].

3.1. CRISPR-Cas Genome Editing in Insect

CRISPR-Cas can be used in agriculture to regulate insect pests and safeguard crops. A two-step method that involves modifying the target insects and releasing them back into the wild can be used to successfully edit an insect's genome [32]. One of the earliest documented uses of the CRISPR-Cas system in insects was in *Drosophila* fruit flies, when effective modifications of the yellow gene were made [33].

The *BmBLOS2* gene was the focus of another reported successful application of the method in silkworm [34], which was followed by several successful applications. In a case study by Garczynski et al. [35], the codling moths genome was edited using the CRISPR-Cas gene editing technology in order to alter the viability and production of eggs by targeting a particular gene (*CpomOR1*). Worldwide, the codling moth is a significant pest of pomes fruit. A member of the pheromone receptor subfamily, the *CpomOR1* gene product is an odorant receptor. In the early-stage eggs of the codling moths, single-guide RNAs (sgRNAs) were created targeting at nucleotides of the *CpomOR1* gene. It was discovered that alterations, including insertions and deletions, were successfully introduced. By mating males with females who have *CpomOR1* gene alterations, the study tried to produce stable populations of edited codling moths by raising the young moths to adulthood. It has been discovered that the modified females' fecundity and fertility are compromised, causing them to produce non-viable eggs. The result was the regulation of fruit pomes by insects. However, it is yet unclear exactly how *CpomOR1* affects the fertility and reproduction of codling moths. In another case, it was claimed that the migratory locust underwent a targeted heritable mutation as a result of the CRISPR-Cas technique. Locusts are a dangerous agricultural pest that have impact on a wide variety of crop plants. Their swarming behavior can result in very serious crop damage over large areas at once, frequently leading to significant financial loss. Li et al. [36] study's involved engineering the guide-target RNA's sequence to prevent the odor receptor co-receptor gene from being expressed (*Orco*). *Orco* gene mutants have been found to exhibit faulty electrophysiological responses to a variety of smells, which prevents them from attracting aggregation pheromones in crowded conditions.

Although the transgenic Bt technology is well established and widely utilized, the development of insect resistance to Bt insecticidal proteins (ICPs) has become a significant concern. In order to avoid this, efforts are being made to build receptors in a way that will enable effective resistance management. By altering the *Helicoverpa armigera* genome, it is possible to successfully knockdown cadherin receptors that are functionally connected to *Cry1Ac* toxin tolerance [37]. Base replacement in the encoding genes of the mid-intestinal receptor has demonstrated how the genome of insects can change their resistance to insect pests. Modifying *Cry* protein binding receptors can be used to edit insects for plant vulnerability. Unique detoxifying enzymes produced by insects are crucial for resolving the chemical defense response in many plant species. A possible alternative would be to focus on the polyphagous bugs' detoxifying genes. Insect susceptibility resulted from targeting and deleting insecticidal detoxifying genes, such as gossypol-inducing cytochrome P450 [38]. The polyphagous insect *H. armigera's* susceptibility to phytotoxins was revealed by CRISPR-Cas-mediated deletion of the *CYP6AE* gene cluster, which also made crops resistant to insects and showed the importance of these enzymes in the detoxification of several toxic phytochemicals [39]. The most long-lasting answer has consistently been this one.

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The modification of target genes that can prevent chemical contact and mating pair recognition, which are crucial for efficient interactions between plants and insects, is another method of controlling insects using CRISPR-Cas. Olfactory receptors *(ORs)* in insects are critical for identifying host plant and mating pair odorants. The *Or83b* gene mutation in Drosophila prevented the host from being detected [40]. Similar to this, the CRISPR-Cas method's deletion of the *Orco* gene from *Spodopthera litura* affects the choice of mating partner and host plant [32]. Implementing such technology would be a smart move to keep insects away from plants and prevent pest damage. In insects, female adults release pheromones that males pick up on. Males select mature females based on pheromone cues. A CRISPR-Cas9-based odorant receptor 16 *(OR16)* knockout in *H. armigera* prevented males from detecting pheromone signals and mating with immature females, which led to the dumping of infertile eggs and helped in controlling insects [41]. Another strategy for controlling insects is to use CRISPR-Cas9 to remove growth genes like the *abd-A (Abdominal A)* gene from a variety of insects, including *Spodoptera litura* [42], *Spodoptera frugiperda* [21], and *Plutella xylostella* [43], which showed abnormal gonads, disarmed prolegs, and lack of body segment functions. The CRISPR-Cas9 technology was used to modify numerous more genes in a variety of insect pests (Table 1).

Table 1. CRISPR-Cas genome editing in insects for insect pests management.

Name of insects	Target gene	Editing	Outcome
Drosophila melanogaster	*yellow*	Knockout, Knock-In	Generated designer flies [33]
	LUBEL	Knockout	Reduced survival rate [44]
	Chitin synthase 1	Substitution	Controlled insect population and resistance to various insecticides [45]
	Nicotinic acetylcholine receptor α6	Substitution	Controlled insect population and resistance to various insecticides [46]
	Scsa	Knockout	Reduced normal growth [47]
	kdr	Knockout	Reduce insecticide resistance [48].
Drosophila suzukii	*White (w), Sex lethal (Sxl)*	Knockout	Controlled insect population and resistance to various insecticides [27]
Spodoptera exigua	*Ryanodine receptor*	Substitution	Controlled insect population and resistance to various insecticides [49]
Spodoptera littoralis	*Orco*	Knockout	Reduced survival rate [32]
Spodoptera litura	*Slabd-A*	Knockout	Defected body segmentation and pigmentation [42]
	SlitPBP3	Knockout	Destroyed pest insect mating [50]
Spodoptera frugiperda	*Sfabd-A*	Indel	Defected body segmentation [21]
Helicoverpa armigera	*OR16*	Knockout	Destroyed pest insect mating [41]
	Tetraspainin	Knockout	Resistance to *Bt* toxin *cry1Ac* [51]
	HaABCA2	Knockout	Resistance to *cry2Aa* and *cry2Ab* [52]

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	CYP6AE	Knockout	Regulation of detoxification enzymes [39]
	nAchR	Knockout	Resistance to insecticide [23]
	HaCad	Knockout	Resistance to *Bt*toxin *Cry1Ac* [37]
Plutella xylostella	*Abdominal-A*	Knockout	Defected body segmentation [43]
	PxABCC2 PxABCC3	Knockout	Resistance to *cry1Ac*protoxin [53]
	Pxabd-A	Knockout	providing novel ideas for pest management [54].
	PxCHS1.	Knockout	Described the resistance management strategies of major agricultural pests [55].
Dendrolimus punctatus	*DpWnt-1*	Knockout	Defected anterior segmentation and appendage development [56]
Danaus plexippus	*clk*	Knockout	Defined the role of the clk gene in the control of migration behavior [57].
Bombyx mori	*BmBLOS2*	Knockout	Generated designer flies [34]
	BmOrco	Knockout	Impaired olfactory sensitivity [58]
Locusta migratoria	*Orco*	Knockout	Generated loss-of-function for managing insect pests [36]
Tribolium castaneum	*EGFP*	Knockout, Knock-In	Controlled insect pest and resistance to insecticides [59]
Gryllus bimaculatus	*Dop1*	Knockout	Destroyed appetitive reinforcement [60]
Rhopalosiphum padi	*ß-1-3glucanase*	Knockout	Reduced callose deposition in maize [61]
Ostrinia furnacalis	*ABCC2*	Knockout	Resistance to*Bt cry1Fa* toxin [62, 63]
Cydia pomonella	*CpomOR1*	Knockout, Knock-In	Affected egg production and viability [35]

3.2. CRISPR-Cas-Mediated Gene Drive in Insects

Genome editing using CRISPR-Cas creates a gene drive that is effective enough to propagate changed genes across generations until they are released for mating. Gene drive is a technique for rapidly distributing altered genes throughout an insect species' entire population. Gene drives based on CRISPR-Cas may cause sterility or mortality in targeted insect species through gene disruption, which ultimately led to population collapse and even elimination due to severe recessive lethal changes [64]. A species will completely disappear as a result of this over the course of 15-20 generations. By selectively harming the X chromosome, the gene drive will alter the male sex ratio. This causes the Y chromosome to be more common in the most viable sperms, resulting in a greater proportion of male progeny and a progressive decline in the number of females [64]. Therefore, releasing insect strains with undesirable features including lethality, infertility, biased sex ratio, insecticidal sensitivity, etc. is a successful method for controlling insect

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pests. For instance, it should be assumed that the Bt resistance management in *H. armigera* is a sustainable method since in this case, gene deletion would only affect the species of *H. armigera* that is resistant to Bt toxins [51].

3.3. CRISPR-Cas Technology in Genome Editing of Crop Plants

Technologies like CRISPR-Cas can improve plant quality to preserve crops and help them survive specific biotic and abiotic challenges [6, 62]. Maintaining healthy plants is a part of the Integrated Pest Management program because insects are drawn to unhealthy, diseased plants. Plants can be modified using CRISPR-Cas systems so that they produce or do not produce particular enzymes that can deter insect pests from coming into touch with the plant or attract specific insect predators to feed on the bug species that are attacking the plant [65]. The process of genome editing is quickly increasing its potential and chances for giving crop plants insect resistance traits. The lack of a clearly defined source of resistance in the gene pool, however, has led to less research into altering plants for pest management. The goal of several efforts to alleviate this bottleneck is to collect genes from uncharacterized crop plant accessions and wild relatives. However, due to poorly understood resistance characteristic genetics in uncharacterized accessions, significant advances could not be made [66]. On the other hand, a transgenic method was used to introduce insect resistance genes into crops from more remote origins, like Bt genes of bacterial source. These transgenic plant species, however, encountered severe political, moral, and social opposition because of a lack of scientific understanding [67]. In this situation, the main challenge for modern agriculture is to develop an environmentally sound breeding strategy for crops that can accomplish two breeding objectives: to produce de novo tolerance in the absence of the proper R-genes and to track the dynamics of pests by destroying insecticide resistance, killing, or inducing insect sterility. Any insect will choose to lay eggs on the host plant if feed is available for the young. Plant volatile blends are combinations of volatiles that serve as cues for insects to select hosts and oviposition sites. Insects use their highly adaptable olfactory systems to detect suitable plants to serve as hosts by detecting volatile secondary chemicals in plants. According to research by Beale et al., altering volatile mixtures by genome editing can kill insects on host plants while making the plants resistant to them. When plants become infested with aphids, the sesquiterpene hydrocarbon (E)-β-farnesene (Eβf) is released, which reduces the populations of other hosts' ability to eat while luring *Diaeretiella rapae*, a parasitic wasp that has been shown to dominate the aphid population in transgenic plants [68]. The genetic engineering of plant volatile blends may be a different strategy for insect management. However, care should be made to ensure that the change doesn't have a negative impact on the species of beneficial insects.

It is also possible to enhance host immunity to pests by editing important plant immunity genes, such as genes regulating targets' interactions with insect effectors and resistance genes (R-genes). Although S genes make plants vulnerable to stress, R genes evaluate a plant's susceptibility to insect pests or diseases [69]. The editing of R and S genes for the development of plant species with insect resistance is emerging as a dependable method. For their growth, immunity, and behaviors that have been observed in rice, insects are dependent on important chemical components contained in plants [22]. Genetic engineering in plants has been demonstrated for insect pest resistance by knocking off S genes from the plants. Tryptamine 5-hydroxylase encoding *CYP71A1* gene deletion using CRISPR-Cas causes tryptamine conversion to serotonin in plants, which reduces plant hopper growth. Rice was changed by Lu et al. [22] using the CRISPR-Cas9 technology to make it resistant to the striped stem borer and the brown plant hopper *(Nilaparvata lugens) (Chilo suppressalis)*. The simultaneous deletion of two endogenous phytoene dehydrogenase (PDS) genes in *P. tomentosa* Carr., *PtoPDS1* and *PtoPDS2*, using the CRISPR-Cas9 technique resulted in the effective generation of endogenous gene mutations in the populus [70, 71]. By enhancing endogenous defenses, CRISPR-Cas genome editing techniques also make it possible to increase the population's resistance to

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insects. The golden promise barley variety's two beta-1- 3 glucanase genes were altered by CRISPR-Cas9, which reduced the amount of callus that formed in sieve tubes. Therefore, the aphid *Rhopalosiphum padi* cannot access the phloem sap and has adversely affected its growth as well as disrupted its predilection for particular hosts [60]. On the basis of a plant's outward appearance, insects can also recognize and target certain plants. It has been found that variations in plant color can influence insect host preferences. This was confirmed in red leaf tobacco made by altering the anthocyanin pathway. By changing the color of the leaf, gene editing for insect pest tolerance in plants has been demonstrated. This prevents the insect from recognizing the host plant. The red color of the leaves was a result of an excess of anthocyanin coloring. The *Helicoverpa armigera* and *Spodoptera litura* were discouraged by this color change [72]. This study demonstrates that CRISPR-based editing for pest management, where the insects are unable to recognize the host plant, may be resolved by altering the anthocyanin pathway (Table 2).

Table 2. CRISPR-Cas genome editing in crops for insect pests management.

Name of crops	Target gene	Editing	Outcome
Rice	*OsCYP71A1*	Deletion	Resistant to the striped stem borer and the brown plant hopper [22].
Paulownia tomentosa	*PtoPDS1, PtoPDS2*	Deletion	Enhance endogenous defenses and increase resistance to insects [70, 71].
Barley	*beta-1-3 glucanase*	Alteration	Resistant to aphid infestation [61].
Tobacco	Anthocyanin pathway	Alteration	Discourage insect attack [72].
Solanum pimpinellifolium	Six different genes	edit	Resistant to insect pests [74].

3.4. Utilization of Crops Wild Relatives for Insect Resistance by CRISPR-Cas Technology

The insertion of foreign genes into the plants is one of the key regulatory problems associated with transgenics that can be overcome by gene editing. The cultivated crops' forebears and close relatives, known as crop wild relatives (CWRs), are robust to biotic and abiotic stress but have low yields. After domesticating wild species and breeding plants, however, the cultivable germplasms and crops have large yields and can meet other human needs, but they cannot withstand insect assault. Using CRISPR-Cas9 genome editing, we may effectively delete or modify the genes that cause an insect's susceptibility, or we can introduce unique features from CWRs into the cultivated species to create new cultivars that are insect-resistant [69].

Two steps can be taken to implement this. First, the de novo domestication of crops with insect resistance's wild cousins. Gene editing techniques can be used to alter desired agronomic traits that are caused by genes. There is evidence that the wild tomato *Solanum pimpinellifolium* is resistant to spider mites and other arthropod insect pests [73]. Multiplex CRISPR-Cas editing of six different genes in *S. pimpinellifolium* resulted in the production of a high-yielding tomato with insect and pest tolerance in a single generation [74]. This method, based on properties and molecular pathways, can be carefully applied to additional CWRs. De novo domestication of the CWRs may therefore be a ground-breaking method for the development of crops with improved characteristics.

Second, using genes found in CWRs that are insect resistant, genome alter the cultivated crops. By altering the genomes of cultivated crops for insect tolerance from wild species, the first study of variation in the sequences of individual insect-sensitive genes across vulnerable cultivated germplasms and resistant wild cousins using multiomic

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techniques may be accomplished [69]. The resistance genes can be successfully used for gene editing after being validated against related insects. These present chances for developing resistance in the gene pool of cultivated crops to control insect pests [75]. It has been suggested that commercially valuable crops can produce insect-resistant phenotypes utilizing CRISPR-Cas gene editing based sequence variation by using either over-expression or silencing techniques. However, this has not yet been demonstrated.

4. Limitations and Future Perspectives

Like other biotechnological techniques, genome editing techniques modify a gene specifically through cellular and in vitro mechanisms. In the course of evolution, genome modification is beyond of our control. However, when the genome is altered experimentally, it may be primarily for the benefit of humans. Its application to crop improvement should likewise be limited to breeding objectives that are both absolutely important and challenging to achieve within the confines of the current heterogeneity. Like any modern technology, there are still a number of legal questions about gene editing that the scientific community needs to address. In order to fully utilize this innovation's potential for the advancement of global agriculture and the eradication of neophobia in society, it is essential to adopt a realistic viewpoint supported by legislative bodies that uphold scientific norms. CRISPR-Cas-based deliberate dissemination of genetic components into wild species of insects that alter the population's sex ratio or contribute to lethal mutations is a precise and environmentally sustainable method of battling pests. However, the emergence of insect resistance in response to CRISPR-mediated gene drive could be a serious and ongoing problem at both experimental and theoretical scales [76, 77]. Multiplex gene editing, however, can overcome resistance [78]. Therefore, it is crucial to address insect resistance issues in order to reach an agreement on ethics and science in favor of this technology.

Additionally, because the engineered insect pests have the power to change the entire population or environment, the introduction of CRISPR-Cas-edited insects bearing gene drives into the ecosystem is linked to a number of biosafety concerns. Prior to their release, stringent risk assessments of non-target outcomes are also required. Unexpected post-release impacts on beneficial insects can have a negative influence on food chains and alter the composition of communities [67]. Additionally, the disease can become worse due to the possibility of gene transfer between the target organisms and their non-target relatives. If the risks are appropriately managed in light of unanticipated environmental repercussions, gene-driven technology could prove effective in the targeted extermination of insect pests, insect vectors for viruses, or alien insect species. Utilizing the terminator genes that permit the programmed life of modified insects and the use of tagged insects to monitor gene flow may seem to be a crucial step to the biosafety use of gene drives in the context of risk management. Additionally, another option for the management of invasive pests is the use of robotic equipment and artificial intelligence to physically eliminate individual pests [79]. Robotics may not be as effective, though, when dealing with tiny insects, uneven terrain, and hidden eggs.

Insect resistance to invasive pests has been successfully achieved via CRISPR-Cas-based deletion of vulnerable genes. The fundamental problem associated with S gene deletion, which also adds to the associated fitness penalty, is pleiotropic effects in the plant. However, it is possible to ensure insect resistance without affecting plant performance by altering the binding effector factor rather than the gene itself [80]. The CRISPR-Cas approach of creating insect resistance in crop species will therefore develop as a successful tool for supplying genetic traits in farmed varieties in a shorter amount of time. It is true that CRISPR-Cas-enabled genome editing technology is a fast evolving technique and thus the scope of its application in agriculture is expanding [81, 82]. However, thorough understanding of the gene and genome activities of the target species is required prior to its full adoption for the generation of insect pest resistance and plant protection. As Bt technology developed through recombinant DNA technology has

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revolutionized in management of insects in many economically important crops including cotton, maize, soyabean and brinjal [83], the ease and multiplexing manner of CRISPR technology would also replace the currently used recombinant DNA technology for the insertation R gene(s) in a faster manner.

5. Concluding remarks

Despite being relatively young, the genome editing techniques centered on CRISPR-Cas have already changed insect functional genomics. In order to create plants resistant to insect infestations, we can now easily change, remove, and add DNA practically anywhere we want in any crop or insect species thanks to CRISPR-Cas. Therefore, this technology needs to be enhanced in order to produce crop plants that are resistant to insect pests. Sincere and proactive measures in this regard are required in addition to protecting our crops from significant output losses brought on by insect pest infestation. However, the global legislative bodies will eventually decide what happens to genome-modified goods created by CRISPR in crop development efforts. Either product-based regulation or process-based regulation is used by regulatory regimes for innovative crop cultivars. The degree to which CRISPR-based crops are regulated will affect their cost of production and the rate at which they are adopted by commercial enterprises. The set of product-based legislation on crops created using CRISPR-Cas genome editing would be classified similarly to products created by classical mutagenesis, eliminating them from the restrictions imposed on products made via genetic modification. This would surely have an impact on the hopeful public perception of this technology and help the majority of nations adopt it. Many countries have given green pass to CRISPR edited products that carry no transgene(s). It is expected that the CRISPR-Cas technology would lead a new green revolution in agriculture if timely deregulation for adoption of CRISPR products and technological know how are shared by an open science practice.

Patents: This is a review article. Therefore, no patent filing is associated with it.

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