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Review

# Importance of Biotechnology in Controlling Insect Pests

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## Abstract

Agriculture is considered an important source of national income for a lot of countries and most of inhabitants of these countries are dependent on farming for their food. Insects have been one of the most important sources of food production damage, accounting for 20% to 30% of global production losses. Approximately 67,000 insect species are thought to inflict harm to plantations and tropical regions, which are often the poorest in the world and are hit hardest by insect pests. Agriculture spends billions of dollars each year on insect control around the world. Pesticide use has resulted in pesticide-resistant insects, a drop in beneficial insect populations, and a slew of other negative effects for humans and the environment. These concerns have led researchers to consider a different approach in order to produce more environmentally friendly insect control tactics that use both synthetic and natural chemicals. The use of bio-pesticides in pest management has been increasing in recent years. There are three types of the ecofriendly management agents bio-pesticides have been identified as follows: bio-control organisms (pathogenic microorganism, predators and parasites); plant-incorporated protectants (transgenic Bt toxin); and biochemical pesticides (botanical pesticides and other natural compounds. Most of chemical companies such as Monsato, Dupont, and Ciba Geigy, are now promoting biotechnology as a strategy to ensure high productivity, these companies invests millions of dollars in biotechnology research to develop genetically modified crops, animals, and microorganisms to combat pests, produce fertilizer, and improve efficiency. Where they uploaded this businesses and scientific institutions are the slogans that biotechnology is the future of agriculture.

**Keywords:** biotechnology, genetically modified

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## 1 INTRODUCTION

Agriculture is considered an important source of national income for a lot of countries and most of inhabitants of these countries are dependent on farming for their food[1].

Insects have been one of the most important sources of food production damage, accounting for 20% to 30% of global production losses. Approximately 67,000 insect species are thought to inflict harm to plantations and tropical regions, which are often the poorest in the world and are hit hardest by insect pests[2].

Agriculture spends billions of dollars each year on insect control around the world[3]. Despite this investment, insect damage can cause up to 40% of a crop to be lost, especially in poorer countries[4].

Insects that are resistant to pesticides have developed, and numbers of beneficial insects have decreased, and a slew of other negative effects for humans and the environment[5-8]. Consumers prefer chemical management because of its rapid effects on the death of insects, but the extensive and excessive use of special chemical insecticides in agriculture leads to several problems of these, different types of viruses are different[9] and chronic diseases such as cancer, hepatic and renal failure in human and other environmental pollutions as air and water pollution which causing adversely or negative impacts on public health[10].

These concerns have led researchers to consider a different approach in order to produce more environmentally friendly insect control tactics that use both synthetic and natural chemicals.

The use of bio-pesticides in pest management has been increasing in recent years. There are three types of the ecofriendly management agents bio-pesticides have been identified as follows: bio-control organisms (pathogenic microorganism, predators and parasites); plant-incorporated protectants (transgenic Bt toxin); and biochemical pesticides (botanical pesticides and other natural compounds)[11].

Most of chemical companies such as Monsato, Dupont, and Ciba Geigy, are now promoting biotechnology as a strategy to ensure high productivity, these companies invests millions of dollars in biotechnology research to develop genetically modified (GM) crops, animals, and microorganisms to combat pests, produce fertilizer, and improve efficiency. Where they uploaded this Businesses and scientific institutions are the slogans that biotechnology is the future of agriculture[12].

## 2 BIO-AGENTS ARE BIOLOGICAL AGENTS THAT ARE USED TO CONTROL INSECT PESTS

Bio-agents include botanicals, microbiological agents, natural enemies, parasitoids, and parasites. Bio control agents come in a variety of forms[13] and biopestides[14], pesticides, which are frequently interchanged, have been used to control pests.

Natural enemies of insect pests, also known as predators and parasitoids are examples of biological control agents. Predators are mostly free-living animals that consume a huge number of preys over the course of their lives. Given that many major crop pests are insects, many of the predators used in biological control are insectivorous species, parasitoid eggs are laid on or in the body of an insect host, which is then consumed by developing larvae. The host is eventually assassinated. Wasps and flies are the most common insect parasitoids, and many of them have a very limited host range[15].

Microbial control refers to the employment of microorganisms or diseases, such as bacteria, viruses, fungi, nematodes, and protozoa, to improve insect management and reduce pest populations to levels that do not cause damage[13]. The microorganisms largely harmless and nonpathogenic to people, animals, and other species when utilized in microbial insecticides. Microbial insecticides frequently target a single insect species or group with their harmful effects. So the microbial insecticides become main component in the integrated pest management programs[16].

Research[17] showed that, botanical insecticides are one approach for controlling insect pests and protecting crops. Botanical pesticides have several advantages, including low human toxicity, low persistence and bioaccumulation in the environment, and selectivity toward beneficial insects. Botanical pesticides (essential oils, flavonoids, alkaloids, glycosides, esters, and fatty acids) contain a wide range of chemical properties and modes of action, including repellents, feeding deterrents/antifeedants, toxicants, growth retardants, chemosterilants, and attractants. So it is preferable to use the botanical insecticides instead of synthetic insecticide and these botanical insecticides are recognized by organic crop producers in industrialized countries[18].

## 3 DEFINITION OF BIOTECHNOLOGY

Biotechnology is “any technological application that uses biological systems, living organisms, to make or modify products or processes for specific use”.

However, in the context of controlling insect pests, management of insect pests is the deliberate and regulated manipulation of biological processes. It is feasible to effectively control such insect pest species by selecting suitable organisms with a certain biological aptitude from the vast array of living things that have evolved[19].

## 4 APPLICATIONS OF BIOTECHNOLOGY AND ITS IMPORTANCE FOR CONTROLLING INSECT PESTS

### 4.1 Tissue Culture (TC) Techniques

TC is a method of vegetative propagation based on biotechnology. The plants are derived from stem, root or leaf tissues and the technology generally aids in mass production of desired crop varieties. TC is also useful in regeneration of GM cells into whole plants.

### 4.1.1 The Advantages and Disadvantage of TC

The advantages and disadvantage of this method are stated as follows[20]:

Advantages:

(1) Controlled environment and controlled development of the plants that enable very rapid multiplication rate;

(2) Clean conditions for plant development that produce micro-plants free of many pests and diseases;

(3) The small size of the propagated plants saves nursery space and plant transport costs.

Disadvantages:

The main disadvantage of TC plants is their high production costs. This difficulty limits the number of plant species in commercial TC propagation.

Plant TC techniques are revolutionising pest management strategies for dealing with host plant resistance by mass-producing planting that is free of diseases materials. TC technologies allow for the rapid growth of disease-free plants in a controlled and aseptic environment in a short amount of time. It gives up a world of possibilities for the creation, conservation, and application of genetic variation for plant improvement[21,22].

For mass multiplication and plant conservation, plant TC techniques have been used[23-28]. These *in vitro* techniques (TC) were also frequently utilised in secondary metabolite production[25,29]. TC is also used to screen disease resistance in plants *in vitro*[30-33].

### 4.2 Transgenic Plants (GM Plants)

Transgenic refers to genetic engineering approaches that involve the transfer of a foreign gene from an unrelated species. This area of genetic engineering encompasses the majority of the GM products of the last two decades.

Over the last two decades, significant progress has been made pest resistance, herbicide tolerance by modifying genes from various and unusual origins are being inserted into microbes and crop plants; and improved nutrient uptake and nutritional quality; enhanced understanding of gene activity and metabolic pathways; higher photosynthetic rate, sugar, and starch synthesis; increased defectiveness of bio-control agents; and drug and vaccine manufacturing in crop plants[21].

Biotechnology is the term used to describe the development of DNA-based technologies. The genetic make-up of organisms can be altered through the use of modern agricultural biotechnology or genetic engineering for the production or processing of agricultural goods. By inserting nucleic acids into virus, bacterial plasmid, or other vector systems outside of cells to enable their absorption into the host in which they do not naturally occur but are capable of ongoing proliferation, genetic engineering creates new combinations of heritable material[34].

The use of transgenic plants expressing plant defence chemicals is one such method. Biotechnology has the potential to give a far wider range of novel insecticidal genes that would otherwise be outside the limits of conventional breeding. In the year 1987 first transgenic plant was developed that expressed an insecticidal gene produced in it. This transgenic tobacco plant produced cowpea trypsin inhibitor (CpTI) at levels of up to 1% of the soluble protein and had enhanced protection against *Heliothis virescens* (Lepidoptera: Noctuidae)[35,36].

Since 1996, when biotech crops were first commercially planted, more than 70 countries from all over the world have either planted or imported biotech crops. The six founding countries of biotech crops, the United States, China, Argentina, Canada, Australia, and Mexico, planted these crops on 1.7 million hectares in 1996. Biotech crops were accepted by 70 nations in 2018, with 26 countries planting them and 44 importing them. In 2018, 26 countries, 21 developing and 5 industrial grew a total of 191.7 million hectares of biotech crops. Biotech crops have grown 113-fold in the 23 years since they were first planted, covering 2.5 billion hectares globally, making biotechnology the world's fastest-growing crop technology[37].

#### 4.2.1 Production of GM Crops

Genetically crops that have been modified are created in a variety of ways:

(1) the most difficult part of the transgenic procedure is identifying and pinpointing genes for plant characteristics. It's not enough to find a single gene linked to a trait; scientists must also determine how the gene is regulated, what other effects it might have on the plant, and how it interacts with other genes in the same biochemical system are all things to consider;

(2) However, after a gene has been discovered and cloned (amplification in a bacterial vector), it must go through a series of steps before being put into a plant;

(3) Transforming plants are heritable changes in a cell or organism caused by the absorption and introduction of foreign DNA. Plant cells and tissues can be transformed in one of two ways: the gene gun approach, which has proven to be particularly effective in changing monocot plants such as corn and rice;

(4) The Agrobacterium method which is considered preferable to the gene gun. When a portion of bacterial DNA is integrated into a plant chromosome, *Agrobacterium tumefaciens* is able a part of its DNA is used to infect plant cells, effectively hijacking the plants' cellular machinery and using it to assure the bacterial population's reproduction;

(5) After the gene insertion process, choose the successfully converted tissues to be moved to a selective medium containing an antibiotic, nutrition, and hormones. Only plants that express the selected marker

gene and carry the transgene of interest will survive; and

(6) Regeneration of complete plants in a series of selective media providing nutrients under controlled environmental conditions (a process that is known as TC)[38]. This process is most commonly used to develop insect resistant crops, which are referred to as GM organisms (GMOs)[39].

Transgenic plants have been produced by addition of one or more following

(1) Bt endotoxin from *Bacillus thuringiensis (B. thuringiensis*)

(2) Protease inhibitors (PI)

(3) Amylase inhibitors

(4) Lectins

(5) Enzymes

#### 4.2.2 Bt Endotoxin Gene

The area of GM crops expanded from 4 to 44 million ha between 1996 and 2000[40]. The two key features introduced are herbicide resistance and insect protection, with insect protection obtained by introducing genes encoding for shortened enterotoxins produced by *B. thuringiensis* strains. Herbicide tolerance and insect protection are the two main features introduced in soybean, maize, cotton, and potato, which are four of the most important crops changed (Bt) [41]. *B. thuringiensis*, biological soil organisms that release a lethal endotoxin. Lepidopterans, Coleopterans, Dipterans, and other related species are among the many pest creatures for which Bt toxins are quite effective, but mammals and the majority of other non-target organisms are not harmful to them using genes from *B. thuringiensis* that encode endotoxins to create transgenic plants with increased resistance to Lepidopteran insect pest larvae is now a well-established method[42].

(1) *B. thuringiensis* var. *israelensis* was active against Blood sucking dipteran insects, such as Mosquitoes and Blackflies that transmit of animal diseases;

(2) *B. thuringiensis* var. *tenebrionis* was active against the larvae of Coleoptera;

(3) *B. thuringiensis* var. *berliner* was active against Lepidopteran;

(4) *B. thuringiensis* var. *thuringiensis* was active against both Lepidoptera and Coleoptera.

A number of unique Bt isolates have been identified to be active against several insect orders in recent years. (Hymenoptera, Homoptera and Orthoptera)[43].

#### 4.2.3 Mode of Action of Bt Toxins

Gut proteases solubilize and activate the 3d-Cry protoxin in sensitive larvae, resulting in a 60kDa toxic fragment containing the three-domain toxin[44]. The activated toxin undergoes a complex series of binding events with the many Cry-binding proteins found in insect guts, resulting in pore formation and membrane insertion[45-47]. Cry toxins enter the midgut cells of larvae through the apical membrane, killing the larvae and harming the cells[48]. Given that 3d-Cry toxins stimulate the formation of nonselective channels that are permeable to cations, anions, and neutral solutes, and that water causes cell swelling and lysis, colloidal osmotic lysis of midgut cells was hypothesise[49].

##### 4.2.3.1 Effects of GM Cotton on Insect Pests

Cotton insect pests *Helicoverpa armigera* (*H. armigera*), *Earias spp., Spodoptera spp*. and *Pectinophora gossypiella* are among the Bt cotton target insect pests. They cause problems on cotton plants by feeding on squares, blooms, and bolls, resulting in considerable yield reductions in severe cases[50].

Cotton bollworm *H. armigera* Hubner (Lepidoptera: Noctuidae), is among the most harmful pests to cotton and many other field crops worldwide[51-53]. In India, this insect is estimated to cost $350 million in crop losses each year, farmers are expected to spray 15-20 times during the growing season. Farmers in Pakistan rely extensively on chemical control to combat this pest, and widespread use of insecticides, especially Pyrethroids, has resulted in pesticide resistance in this pest[54,55].

*H. armigera* is resistant to transgenic Bt cotton cultivars[56-60]. It shown to be quite effective in eliminating this pest in Australia, producing 80-90% death. China has more than 90%[61] and 40-50% in India[62]. However, other researchers have found that Bt cotton does not effectively reduce *H. armigera*[36].

Pink bollworm *Pectinophora gossypiella* Saunder (Lepidoptera: Gelechidae) wherever cotton is grown, it is the most significant pest[63,64]. Because of its mysterious feeding habits, this insect is nearly impossible to control. This pest can be efficiently controlled by Bt cotton carrying Cry1Ac[59].

Bt-cotton has enhanced cotton yields by up to 60% in India alone, while cutting pesticide applications in half. This has resulted in an increase in revenues of up to $11.9 billion every year[65].

Spotted bollworm *Earias spp*. (Lepidoptera: Noctuidae). It is a major cotton pest in the Indo-Pak subcontinent, causing damage to fruiting bodies as well as square, flower, and boll shedding[66]. Although transgenic Bt cotton is primarily used to control the cotton bollworm, *H. armigera*, it also has a substantial influence on other bollworm species such as *Earias insulana* and *Earias vittella*. Because it is a pest of the early to midseason in cotton, transgenic Bt cotton can effectively control it during the early to midseason, when toxin expression is strong. Cry1Ac was shown to be very poisonous to spotted bollworms, with $LC_{50}$ values ranging from 0.006 to 0.105g/mL of food and 0.88ng/$cm^2$ in leaf-dip bioassays[67].

Another study in Pakistan looked at the spotted bollworm infestation trend finding a minimum infestation of 3.36% in transgenic variety "IR-FH-901" compared to 10.5% infestation in conventional variety "FH-900" in different plant parts of transgenic Bt and conventional cotton cultivars[68].

#### 4.2.3.2 GM Cotton's Effect on Major Sucking Insect Infestations

Whitefly, jassid, thrips, aphid, and cotton mealy bug are among GM cotton's non-target sucking insect pests. These pests are extremely damaging to cotton plants throughout the seedling and vegetative stages, sucking the plant's sap, weakening it and, in severe infestations, causing wilting and leaf shedding. According to the results of a field investigation, transgenic Bt cotton was particularly successful against specific chewing Lepidopterous pests and reduced the use of insecticides[69]. Non-target sucking insect pests, on the other hand, may become substantial insect pests as a result of the reduced use of pesticides in Bt cotton, which can expand the sucking insect pest complex[70]. The majority of research investigations have found that transgenic Bt cotton has a greater population of sucking insect pests such as jassid, whitefly, aphid, and thrips[71]. Some other research studies conducted in Pakistan[72]. There were no substantial differences in the populations of between transgenic Bt and non-Bt cotton, sucking insect pests such as whitefly, jassid, and thrips because Bt cotton lacks resistance to sucking insect pests, effective management of these pests necessitates the application of pesticides and other control methods on a constant basis[73,74].

#### 4.2.3.3 Effects of GM Corn on Insect Pests

In 1997, Bt corn was commercially planted for the first time in the United States, By 2009, it has been planted in 11 countries, including Canada and Europe (Spain). It amounted for 85% of total corn area in the United States at the time, 84% in Canada and 20% in Spain, in Egypt planted 1,000 hectares of Bt corn in 2012, down from 2,800 hectares in 2011[75]. In 2016, 60.6 million ha of GM corn were planted worldwide (in 16 countries). The crop was created to resist the European corn borer, *Ostrinia nubilalis* (Hübner) (Lepidoptera: Crambidae) as well as the corn earworm, *Helicoverpa zea* (Boddie) (Lepidoptera:Noctuidae). In addition to *Heliothis zea* (Lepidoptera:Noctuidae), the maize rootworm *Diabrotica virgifera* (LeConte) (Coleoptera: Chrysomelidae), and *O. nubilalis*, later in the 2000s[75].

In spite of Egypt's announcement in 2009 that any agricultural import must have a certificate from the country of origin stating that the product is not GM[76], the country takes a permissive approach to GMOs, and its public policy does not oppose growing, importing, and exporting GM crops. According to recent news reports, Egypt ranks third in Africa in planting and importing GM crops. Since 2010, GM crops have been planted without restrictions in ten different Egyptian provinces, including one thousand hectares of GM corn in 2012. In 2008, Egypt became the first North African country to grow GM crops, and it is now one of the five countries worldwide to introduce biotech crops to other countries.

In Egypt, Massoud[77] tested three corn hybrids expressing the gene Cry1Ab transgenic corn plant was more effective against Lepidopteran insects.

#### 4.2.3.4 Effects of GM Potato on Insect Pests

Potato aphids are the most common insect pests[78], with the polyphagous *Myzus persicae* (Hemiptera: Aphididae) being the most common and investigated. Aphids cause direct damage to plants by piercing and sucking the phloem. The fact that *Myzus persicae* is a vector for over a hundred plant viruses, with roughly twelve directly harming potato crops, including many leaf-roll viruses, is even more harmful[79].

Jenny et al.[80] showed that aphid fitness differed far more between normal potato varieties than between DeSiree and the GM events, according to the findings. It's important to compare distinct GM events to the non-transformed kind because insertion can have unintended effects.

In Egypt, Hussein et al.[81] found that Cry3Aa can cause some effects on the cotton leaf worm *Spodoptera littoralis* (*S. littoralis*) (Boisd.) (Lepidoptera: Noctuidae), while Cry1Ae, Cry1Ab and Cry3Aa transgenic potato plant were more effective against *Phthoremia operculella* (Zeller) (Lepidoptera: Gelechiidae)*, S. littoralis* and *Agrotis ipsilon* (Lepidoptera: Noctuidae)[82-84].

#### 4.2.3.5 Effects of GM Soybean on Insect Pests

For more than a decade, the strategy of genetically modifying soybeans to produce resistance to Lepidopterans by the introduction of Bt toxins has been applied. As a result, a variety of strategies have been used to successfully deliver Cry genes into soybean embryos[85]. As a result, resistance to *Helicoverpa zea* (*H. zea*)*, Pseudoplusia includes* (Lepidoptera: Noctuidae), *Anticarsia gemmatalis* (Lepidoptera: Noctuidae) and *Elasmopalpus lignosellus* (Lepidoptera: Pyralidae) was demonstrated in a GM soybean containing a combination of 229-M from the strain PI 22948 and a synthetic Cry1Ac gene[86].

Further research combined the synthetic Cry1Ac gene with both quantitative trait loci (QTLs), 229-H and 229-M, to create transgenic soybeans that were resistant to two Lepidopteran insect pests (*H. zea* and *Heliothis virescens* (*H. virescens*))[87].

The development of a GM soybean with a third quantitative trait locus (QTL), QTL-G, as well as the cry1Ac resistance gene was discussed in later articles. QTL-M was found to have the greatest impact on *Photedes includens* and *H. zea* resistance. *H. zea* larvae were more resistant to QTL-G[88,89]. When compared to the other two resistance genes, QTL-H was less effective[87,89]. As a result, adding another QTL to boost pest resistance in soybean cultivars could be an attractive technique for biological control[89].

Transgenic soybean lines expressing the *B. thuringiensis* toxin Cry1Ac have recently been evaluated in the field for potential resistance to lepidopteran pests. Bt toxins have been shown to be effective in soybean as a resistance mechanism against *Anticarsia. gemmatalis*, *Photedes includens*, and *Hypena scabra* (Lepidoptera: Erebidae)[90].

In Egypt, *S. littoralis* larvae fed transgenic material looked to be smaller than control larvae, according to the study[91]. The accumulation of greater phenoloxidase activity in insect tissues resulted in a reduction in insect size and weight. The higher mortality observed in L2 was due to a significant decrease in the acetylcholine esterase activity that leads to accumulation of acetyl cholin at higher levels which causes paralysis and death.

#### 4.2.3.6 Effects of GM Tomato on Insect Pests

In Egypt, Saker et al.[92] showed that, using the Agrobacterium-mediated transformation approach, a transgenic tomato (cv. Money maker) overexpressing the Bt (Cry 2Ab) gene was created. The expression and incorporation of the transgene into the tomato genome were validated using molecular and biochemical analyses. Obvious effects of Cry 2Ab were judged by the mortality of the American bollworm *H. armigera* (Hu bner) and the potato tuber moth *Ph. operculella* (Zeller) when fed on Bt tomato. These findings suggest that all transgenic lines have a large amount of Bt protein, and that plants expressing the Cry 2Ab gene could be exploited to control endemic lepidopteran insect pests. In comparison to the other two target species, the larvae of *S. littoralis* appear to be more tolerant. After 7 days of feeding on transgenic plants, the mortality of the 1st and 2nd instars was 47.30% and 36.30%, respectively. After 7 days of feeding on transgenic plants, the proportion of larval mortality in the 3rd and 4th instar larvae was decreased, ranging between 20-44% and 18-32%, respectively.

The insecticidal toxin from the bacterium *B. thuringiensis* has been inserted into a tomato plant when field tested they showed resistance to the tobacco hornworm (*Manduca sexta*) (Lepidoptera: Sphingidae), tomato fruit worm (*H. zea*), the tomato pinworm *Keiferia lycopersicella* (Lepidoptera: Gelechiidae ) and the tomato fruit borer (*H. armigera*)[93].

Bt technology has also been applied to the control of the tomato leaf miner *Tuta absoluta* (Lepidoptera: Gelechiidae) when the *cry1Ac* gene was introduced into tomato plants, *Bt*-expressing tomato lines were better protected against the leaf miner[94].

#### 4.2.4 Bt Crops: Are They Safe?

Organizations such as Monsanto, which manufactures GM crops with Cry toxins, argue that the toxins are only active against specific insects and have no significant environmental impact, mammals, or human[95].

#### 4.2.4.1 Safety to Beneficial Insects

Lady bugs (Coccinellids), green lacewings (*Chrysoperla spp*.) and lady beetles (Coccinellids) were not poisoned by pollen containing Cry toxins in laboratory trials, according to reference[95].

Furthermore, field investigations demonstrated beneficial arthropods were found in considerably greater numbers in Bt crops than in chemical pesticide-treated crops.

Head et al.[96] detected no remnants of Bt protein in their bodies when aphids were fed Bt transgenic maize or Bt protein-containing artificial meals.

Dutton et al.[97] discovered that the Cry1A protein class had no direct effect on the larvae of lacewings. Given that the green lacewing is a predatory generalist, that feeds on aphids and other insect eggs in addition to lepidopteran larvae in the field, Bt crops are unlikely to pose a threat to this useful predator.

In Egypt, Al-Deeb and Wilde[98] found Bt corn for corn rootworm control had no deleterious effects on beneficial insects.

The detrimental effects of Cry2Ab on *Chrysoperla carnea* larvae fed on lepidopteran larvae have been described by Romeis et al.[99] were attributable to a drop in prey quality rather than a direct harmful effect.

Feeding studies in the lab were carried out by Rose et al.[100] honeybees given Cry1Ab sweet corn pollen for 35 days had no influence on their weight or survival.

In field studies, there were no negative impacts on bee weight, foraging activity, or colony performance in Bt pollen cakes were provided to colonies foraging in sweet corn areas for 28 days. Exposure to Bt pollen had no effect on brood growth. The mortality rate between the treated and control groups was minimal when the 2nd instar larvae were fed pure Bt toxins mixed in with their food at quantities far greater than those they would be exposed to in the wild.

"According to Duan et al.[101]" when the 2nd instar larvae were fed pure Bt toxins, Bt toxins mixed in with their food at concentrations were significantly higher than those to which they would be exposed in the wild.

According to the study[102], aphid counts in Bt cotton fields in 36 locations across six northern Chinese areas had dropped considerably. This decline was connected to an increase in Coccinellid, Chrysopid, and Spider populations, according to the researchers. Furthermore, larger predator Insect biological control on surrounding cotton, corn, and peanut crops was greatly influenced by populations of Bt cotton.

Dahi[103] found that Bt cotton that produces Cry1Ac and Cry2Ab had no effect on the numbers or quantity of common predators in cotton fields in Egypt.

Whether *Chrysoperla rufilabris* eaten cabbage looper, *Trichoplusiani*, or fall army worm, *Spodoptera frugiperda,* that had consumed Bt or non-Bt plants, there were no differences in any of the fitness metrics (larval survival, development time, fecundity, and egg hatchability).

In Egypt, Feeding *Carlo Carena* larvae on Aphids grown on Bt maize till pupation or adult emergence was validated by Moussa et al.[104], they added that studies of microbial diversity, the physical and chemical qualities of soil, as well as the organisation of the soil microbial population, were not changed by transgenic popular.

### 4.2.5 PI for the Control of Insect Pests

Insects' guts contain proteases, which are enzymes that aid in protein digestion. Insect digestion is affected by PI, which are chemicals that block proteases. To create transgenic plants, the protease inhibitor gene is extracted from one plant and cloned into another.

e.g. Transgenic apple, rice, tobacco containing PI.

e.g. CpTI is a PI isolated from cowpea and cloned into tobacco. This transgenic tobacco is resistant to *H. virescens.*

Plants protect themselves directly by constitutively expressing PI[105] in reaction to mechanical damage or insect attack, the body produces PI[106,107]. They may also emit volatile compounds in response to insect injury, which act as powerful attractants for insect herbivore predators[108]. Following injury, volatile compounds like Methyl Jasmonate because neighboring unwounded plants to produce proteinase inhibitors, effectively protecting the local population against insect attack[109].

When compared to transgenic tobacco plants expressing only a Bt-toxin, *H. armigera* was less harmful to transgenic tobacco plants that expressed both a Bt-toxin and a CpTI[110].

There are many transgenic crops expressing plant proteases inhibitors genes that controlled many insect pests (Table 1) such as CpTI controlled *Chilo suprssalis* (Lepidoptera: Crambidae)*, Sesamia inferens* (Lepidoptera: Noctuidae) *Spodoptera litura,* (*S. litura*), *Helicoverpa armigera, Sitotroga cerealla* (Lepidoptera: Gelechiidae) and *H. armigera.*

#### 4.2.5.1 PI' Mechanism of Action in Lepidopteran Insects

When added to artificial diets or expressed in transgenic plants, PI enhance mortality[112] and larvae of a variety of insect pests, including Coleoptera, grow and mature slowly[113], Orthoptera[114] and Lepidoptera[115,116] ingested PIs have different pathways for mediating their effects on insect physiology according to the insect species[117]. Proteinase inhibitors bind to digestive proteases in insects, blocking proteolysis, which prevents protein digestion[105].

### 4.2.6 Amylase Inhibitor Gene

Amylase is a carbohydrate-digesting digestive enzyme found in insects. Amylase inhibitors impact insect digestion.

Transgenic tobacco and tomato have been developed that express an amylase inhibitor and are resistant to Lepidopteran pests. A case in point is provided by the inhibitors of α-amylases found in the common bean, *Phaseolus vulgaris*. Bean seeds contain at least two different α-amylase inhibitors called αAI-1 and αAI-2[118,119].

### 4.2.7 Lectins Genes

Lectins are proteins that bind to carbohydrates, and when insects consume them, they link to chitin in the peritrophic membrane of the midgut, preventing nutrients from being absorbed. e.g. *H. virescens* is resistant to transgenic tobacco expressing the pea lectin gene, Lectins, also known as carbohydrate-binding proteins are found across the plant kingdom and are stored in plant tissues as defensive proteins in many species. They are especially numerous can account for up to 1% or more of total protein in seeds and other storage[120,121].

Some lectins, such as the *Wheat germ* agglutinin are

**Table 1. GM Plants Express Protease Inhibitor Genes[111]**

| Inhibitor | Crop Plant | Crop Pests |
|---|---|---|
| Cowpea trypsin inhibitor (CpTi) | Tobacco<br>Rice<br>Potato<br>Strawberry<br>Tobacco<br>Cotton<br>Wheat | *H. virescens*<br>*C.suprssalis*<br>*S. inferens*<br>*Lacanobia oeraceae*<br>*Otiorynchus suscatus*<br>*S. litura*<br>*H. armigera.*<br>*S. cerealla* |
| CpTi and Snowdrop lectin potato inhibitor II | Sweet potato<br>Tobacco<br>Rice | *Cyclas formicarius*<br>*Manduca Sexta*<br>*S. inferens* |
| Tomato inhibitors I and II | Tobacco | *M. Sexta* |
| Sweet potato trypsin inhibitor (TI) | Tobacco | *M. Sexta* |
| Soybean TI | Rice | *Nilaparvta lugens* |
| Barley TI | Tobacco | *A. ipsilon*<br>*S. litura* |
| Nicotinia alat protease inhibitor (PI) | Tobacco | *H. armegera* |
| Serpin Type serine PI | Tobacco | *Bemesia tabaci* |

highly toxic to mammals[122]. While the pea lectin could be used as transgenic resistance factors against various insects' pests; these include Hemiptera such as aphids[123] and the rice brown plant hopper *Nilaparvata lugens* (Stål) (Hemiptera: Delphacidae)[124], Coleoptera such as bruchid beetles and Colorado potato beetle *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae)[125], A gene called *Allium cepa* agglutinin has also been shown to have insecticidal properties and is used to combat sap sucking insects[126].

*Galanthus nivalis* agglutinin (GNA, snowdrop lectin), a lectin which is exhibiting a strict specificity for alpha-d-mannose belongs to a group of lectins isolated from bulbs of species in the plant family Amaryllidaceae[127]. In Egypt, GNA is toxic towards Colorado potato beetle, *Leptinotarsa decemlineata* and against some lepidopteran insects include *S. littoralis* and *Agrotis ipsilon*[128].

### 4.2.8 Enzyme Genes

Plants have been cloned with the chitinase enzyme gene and the cholesterol oxidase gene, both of which have insecticidal capabilities. Cholesterol oxidase: A screening approach assaying culture filtrates of different bacterial species led to the discovery of a protein from Streptomyces that was highly insecticidal to larvae of the coleopteran pest cotton boll weevil *Anthonomus grandis* (coleopteran: Curculionidae)[129].

### 4.3 RNA-based Technologies for Insect Control in Plant Production

RNA interference (RNAi) is a biological process in which sequence-specific short RNA (sRNA) molecules suppress gene expression at the transcriptional or post-transcriptional level by causing inhibitory chromatin modifications or decreasing the stability or translation potential of the targeted mRNA. The first commercial products have been developed using RNAi mechanisms. Given their enormous potential, RNAi techniques will almost certainly be used extensively in agriculture, horticulture, and forestry in the near future more research is needed, however, to increase the efficacy of RNAi-based plant protection methods and assess the impact of these methods[130].

James et al.[131] showed that the expression of *B. thuringiensis* insecticidal proteins, the majority of which permeabilize the membranes of sensitive insects' gut epithelial cells, is required for commercial biotechnology solutions for controlling lepidopteran and coleopteran insect pests on crops. They show that ingestion of double-stranded dsRNAs supplied in an artificial diet promotes RNAi in a variety of coleopteran species, most notably *Diabrotica virgifera virgifera* LeConte, the western corn rootworm[132].

However, insects in the orders Lepidoptera (moths and butterflies), Diptera (flies and mosquitoes), and Hemiptera (aphids, hoppers, and stinkbugs) respond to ingested dsRNA in different ways than beetles[133]. A transgenic corn crop created by Monsanto (now bayer crop science) that expresses a hairpin dsRNA targeting the snf7 gene in the Western corn rootworm, *Diabrotica virgifera virgifera*, was the first commercial RNAi product targeting an insect pest[134,135]. In order to delay the evolution of resistance, this novel RNAi construct is stacked with two *B. thuringiensis* Cry proteins (Cry3Bb1 and Cry34/35Ab)[136]. This device will be known as Smart Stax Pro, and it was certified by the US Environmental Protection Agency in 2017[137]. It is projected to be available for commercial use by the end of the decade. Smart Stax Pro is regarded as a watershed moment in the application of RNAi technology in agriculture.

The dsRNA must prevent destruction by nucleases from the insect's salivary glands, midgut, and hemolymph once it has been ingested. Saliva degrades dsRNA in Hemipterans *Lygus lineolaris*, a tarnished plant bug, and *Acyrthosiphon pisum*, a peach aphid (Homoptera: Aphididae)[138,139].

The tobacco hornworm, *Manduca sexta* (Lepidoptera: Sphingidae), and the German cockroach, *Blatella germanica* (Linnaeus) (Blattodea: Blattellidae), both showed after 1 and 24 hours, dsRNA degraded in the hemolymph, respectively[140]. Midgut fluids degrade dsRNA in the silkworm *Bombyx mori* (Lepidoptera: Bombycidae), *Schistocerca gregaria*, a desert locust (Orthoptera: Acrididae), and *Leptinotarsa decemlineata*, a Colorado potato beetle (Coleoptera: Chrysomelidae )[141,142]. dsRNA degraded in *Bombyx mori* after only 10min of exposure to midgut nucleases, the efficiency of nucleases in insect stomachs varies depending on the species.

### 4.3.1 The Benefits, Drawbacks and Biosecurity Concerns of Insect Control Based on RNAi the Benefits of Using dsRNA to Manage Insects

RNAi-based approaches may provide a more environmental friendly method of controlling insect pests while decreasing the use of traditional pesticides. Furthermore, because RNAi could be quite particular, it's a good idea to employ it[143-145].

RNAi is also a natural process occurring in almost all eukaryotes, which is an added benefit. sRNAs are produced by both animals and plants to regulate the expression of endogenous genes and transposable elements, as well as to combat viral infection. RNAi-based pest management is thought to be non-toxic because humans absorb sRNAs on a daily basis[146].

Moreover, the mechanism of RNAi activity has been thoroughly investigated. To silence the target gene, neither host-induced gene silencing nor synthesised dsRNA require the production of a transgenic protein. This is beneficial since it lowers the risk of toxicity in animals after ingestion or exposure. Furthermore, because dsRNAs decay quickly in soil and water (half-lives of less than 30 or 72h, respectively), topical administration of dsRNA may not represent an environmental risk[147,148].

RNAi has been shown to minimize predation on plants and increasing the survival and fecundity of various insects in laboratory and greenhouse studies. These findings show that RNAi-based pest management techniques could have a wide range of agronomic implications.

#### 4.3.1.1 dsRNA-Mediated Insect Control has Drawbacks

RNAi-based insect pest control strategies have a lot of drawbacks to consider, despite their enormous potential:

(1) Public opposition to GMOs is a major challenge, especially for host-induced gene silencing -based initiatives.

(2) Furthermore, environmental RNAi varies greatly between insect groups and species. As a result, not all insects are receptive to environmental RNAi, which is a big worry.

(3) Furthermore, the insect's life stage and whether it has been starved can have an impact on Activity of dsRNase and, as a result, dsRNA instability[149].

#### 4.3.1.2 Biosafety Aspects of RNAi-based Crop Protection

While multiple published studies show that insect control with RNA is feasible, Large-scale use will necessitate a deeper understanding of how to mitigate the drawbacks, as well as the resolution of a wide range of scientific, administrative, and safety problems[150-152]. Off-target effects occur when sRNAs made from dsRNA silence genes in a cell or organism that are not the intended target are a major problem[153]. The occurrence of off-target binding sites within the genomes of target animals is a significant possibility due to the tiny size of sRNAs[154-157].

### 4.4 Genome Editing Technologies and New Breeding Techniques

In recent years, newer genetic modification techniques (nGMs) and new breeding technologies (NBTs) are being developed in a variety of ways at a breakneck pace created and employed to more precisely manipulate the genetic make-up of a plant variety in order to add new features or make changes to existing ones these techniques have shown to be effective hastened the development of crop varieties with a variety of valuable characteristics[158,159]. The introduction of these nGMs or NBTs has sparked global debates about their regulation, with existing GMO regulatory systems covering nGMs to varying degrees[160].

The capacity of insects to recognize and attack host plants is heavily influenced by their visual appearance. Insect host choice has been discovered to be influenced by changes in plant pigmentation. This behavior has been observed a transgenic plant developed by changing the anthocyanin pathway in red leaf tobacco[161]. Overproduction of anthocyanin pigmentation caused the transgenic tobacco plant's leaves to turn red, according to the study. The herbivores, *S. litura* and *H. armigera*, were deterred by the change in leaf colour, confirming the importance of leaf colour and appearance in insect host recognition. In the field of genome editing for biotic stress tolerance, this method has proven to be effective.

In response to the attack of biotic stress factors, plants have created a variety of methods. While resistance genes (R genes) determine a plant's ability to fight pests and diseases, susceptible genes (S genes) make them succumb to the stress[162]. Editing of susceptible genes for the development of insect resistant plants is emerging as a reliable strategy. Insects rely on important chemical components from plants for their development, immunity, and behavior. The fact that serotonin, a plant-derived neurotransmitter, is required for larval immunity and behavior was proposed[163].

### 4.5 Sterile Insect Technique

The sterile insect technique (SIT) involves the introduction of radiation-sterilized insects as part of an environmentally friendly, area-wide integrated pest management programme. It was first used against the new world screwworm, *Cochliomya hominivorax* (Coquerel) (Diptera: Calliphoridae), in the 1950s in the United States. Since then, it has been refined in many ways, and its use extended to include at least 15 other pests, most of them Lepidopterans and Tephritids[164]. The Joint Food and Agriculture Organization of the United Nations/International Atomic Energy Agency Programmed on Nuclear Techniques in Food and Agriculture, based in Vienna, Austria, has directed subsequent research and development of SIT, as well as its international promotion SIT is presently utilized on each of the world's six continents[165].

SIT is constantly being developed and improved around the world, and its use against other pests, such as mosquitoes, is constantly being researched and sugarcane borers in southern Africa[166-169]. SIT entails mass-rearing the target species, sterilising them with ionizing radiation (where possible, only the males), and releasing millions of sterilized insects into the target region every week[170]. Males who have been sterilized mate with viable "wild-type" females, producing only infertile eggs as a result. Providing that certain cultural measures, such as insect control, plant removal, or stripping of immature fruit, and on-farm cleanliness are carried out at the same time, the size of successive generations of the pest is thus systematically reduced[171].

In general, induced lethal mutations by sterilizing doses of gamma irradiation may cause death at any stage of development, but for the sake of simplicity and convenience, harmful deadly mutations are induced at the time of egg hatch[172], Lethal mutations, on the other hand, can occur at any stage of development. The pupal and adult products of irradiated male flies can be utilised to evaluate the effective dosage sterility.

Full sterility in fruit fly males usually lower the quality and it's typically better to lower the dose in order to achieve a greater induction of sterility in field females by having more competitive males[173].

Moreover, Lux et al.[174] reported that using routine irradiation as commonly used in the mass rearing of the sterilized male facilities, reduces the mating performance nearly two-fold.

Collins et al.[175] reported a range of 20-70 Gray (Gy) of gamma irradiated full grown pupae in *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae ) to reduce the target sterilization dose below that of current dose range (70-75Gy) at the same time as retaining an adequate safety margin above radiation doses at which residual fertility can be expected.

Also, Guerfali et al.[176] suggested a dose range of 50-145Gy of gamma irradiated full grown pupae to see if a theoretical model for producing sterility in a wild population was supported at higher doses. They recommended that this experiment needs to be repeated at least in a semi field cage.

### 4.6 GM Insects

Many insects, including agricultural pests like the Mediterranean fruit fly *Ceratitis capitata* (Diptera: Tephritidae) and disease vectors like mosquitoes, have been genetically altered in the last ten years thanks to novel gene manipulation approaches.

Researchers are getting ready to release some GM insects into the wild, with the 2006 release of a GM pink bollworm moth (a pest of cotton) in the United States being the first use of GM insects in a plant pest control program[177].

Insects that have been GM are created by adding new genes into their DNA.

#### 4.6.1 GM Insect Strategies' Potential Benefits

GM insects are seen as a tool to supplement existing management strategies by proponents. GM insects are thought to have a number of distinct advantages:

(1) They would only target a single insect nuisance species, avoiding harming beneficial insects;

(2) Pest populations inaccessible to traditional control measures could be removed by leveraging insects' natural proclivity to discover one another;

(3) GM insects could lessen the requirement for

insecticides and any related hazardous residues in the environment;

(4) GM insects would safeguard everyone in the release region, regardless of socioeconomic position, if they were deployed in disease control program; and

(5) Disease control employing GM insects would necessitate less community participation, making it less sensitive to individual failure to participate in a control program.

They have expressed many reservations concerning the release of GM insects, including the following:

(1) New insects may fill the ecological niche left by the insects suppressed or replaced, possibly resulting in new public health or agricultural problems;

(2) The new genes engineered into the insects may "jump" into other species, a process called horizontal transfer, causing unintended consequences to the ecosystem; and

(3) Releases would be impossible to track and irrevocable, as would any environmental damage.

Chen et al.[178] developed a novel silkworm strain with great resistance to infection by the *Bombyx mori* nuclear polyhedrovirus.

By replacing this editing approach generated a male-only breed in which the sex ratio of silkworms may be adjusted utilising W chromosomal insertion by combining the silkworm fibroin heavy chain gene with the major ampullate spidroin-1 gene from the spider *Nephila clavipes*[179].

Female lethality can be caused by inserting a cassette containing an embryonic fatal gene KO function into a specific position on the W chromosome. Males generate higher-quality and quantity silk than females, so this technique will be valuable not only in the silk industry, but also in the creation of environmentally acceptable lepidopteran pest management[180].

### 4.7 Other Types of Biotechnology as Modified Bio-Agent Using for Controlling Insect Pests

#### 4.7.1 Induction Mutation (Induction Mutation Ultraviolet (UV) Rays and Ethylmethansulfonate Treatments)

The main problems with *B. thuringiensis* products for pest control are their narrow activity spectrum and high crystal sensitivity to UV degradation. The researchers looked at a UV-resistant mutant (Bt-m) of *B. thuringiensis* subsp. *kurstaki* that produces a dark brown pigment called melanin. The larvicidity of Bt-m against *H. armigera* was higher than that of its parent. Bt-m spore survival and insecticidal activity were both higher than the parent after irradiation at 254nm and 366nm[181].

Also, in Egypt mutants of *B. thuringiensis* var. *kurstaki* HD-73 (parent strain) producing a melanin was obtained after treatment with the mutagenic agent ethyl-methane-sulfonate against potato tuber moth *Ph.operculella*[182].

#### 4.7.2 Protoplast Fusion

In Egypt, Shereen et al.[183] using protoplast fusion technique between two native strains *Bacillus Subtillus* subsp. *subtilis* strain (Bs1) and *Bacillus licheniforms* strain (Bl) to increase the effect of bio-control on *Tuta absoluta* they were isolated fifty six of fusants products and studied their effect on mortality of *Tuta absoluta* under lab conditions, obtained results showed as follow; for the first attempt, the best Fusan time was 40min that was registered high mortality percentage of tomato leaf miner ranged from 74% to 100% for fusants product F7, F8 and F9 respectively compared with their parents.

To create *Pseudomonas fluorescens* hybrids with insecticidal activity, protoplast fusion was performed between a Gram-negative strain *Pseudomonas fluorescens* with plant growth promoting activities and a Gram-positive *B. thuringiensis* var. *kurstaki* HD 73 with insecticidal activity[184].

#### 4.7.3 Transformation (Transgenic Microorganism)

Shuttle vectors are frequently used to quickly make multiple copies of the gene, A cry1Ab gene isolated from a native *B. thuringiensis* strain (LM-466), showing a relevant activity against *Tuta absoluta* larvae, was cloned into the shuttle vector pHT315[185]. Hydroxylamine was used to treat the replication region of the pHT1030 *B. thuringiensis* plasmid. Several copy-number mutants were chosen, and shuttle vectors with several cloning sites were created as a result. The cloning of a delta-endotoxin-encoding gene in *B. thuringiensis* was made possible by these recombinant plasmids, which are extremely stable. With a copy-number of about fifteen per equivalent chromosome, a plateau in delta-endotoxin synthesis is reached, according to a comparison of gene expression levels and vector copy-number.

## 5 ADVANTAGES, DISADVANTAGES AND SAFETY OF BIOTECHNOLOGY

### 5.1 Advantages

Since their introduction in 1995/1996, Bt-protected crops, particularly corn and cotton, have proven significant benefits. Insect protection provided by these items is often superior to that provided by conventional chemical insecticides. As a result, Bt-protected crops require fewer treatments of externally applied pesticides, resulting in a significant reduction in overall chemical pest control product use and the preservation of beneficial insect populations[186].

(1) Reducing rates of infectious disease;

(2) Reducing water consumption and trash generation;

(3) Generating higher crop yields with fewer inputs;

(4) reducing the amount of agricultural chemicals used by crops and limiting product run-off into the environment;

(5) Using transgenic crops that require fewer pesticide treatments; and

(6) Developing crops with enhanced nutrition profiles that solve vitamin and nutrient deficiencies; creating meals that are devoid of allergies and poisons improving the nutritional value of foods and agricultural oils in order to promote cardiovascular health[186].

### 5.2 Disadvantages

Biotechnology has benefited the globe in many ways, but it also has drawbacks, and some people are concerned about its potential harmful consequences. Concerns have been raised in agriculture that GM crops could spread genetic material to wild, unmodified plants. For example, a herbicide-resistant crop could transfer some of its features to a weed, resulting in a herbicide-resistant weed Another concern about agricultural biotechnology centers on the uncertainty of GM crops' long-term biological viability[187].

### 5.3 Safety

Before being used in commercial agriculture, Bt-protected plants are intensively researched. According to these investigations, Cry and marker proteins are not toxic to humans and pose no significant concern for allergenicity.

(1) Except for the presence of the Cry and flag proteins, Bt-protected plants are nearly identical to non-Bt counterparts;

(2) Food and feed obtained from Bt-protected crops are safe to eat, based on the previous two statements;

(3) Except for certain insects that are closely related to the target pest, cry proteins are virtually harmless to all target animals; and

(4) The Cry and marker proteins, as well as the Bt-protected plants themselves, offer no known environmental concerns.

The data on Bt-protected crops has been assessed by a number of regulatory agencies throughout the world. They have judged that these items are safe and suitable for introduction into commercial agriculture, in accordance with their regulatory mandates. The nearly 40-year history of safe usage of Cry proteins in Bt microbiological products around the world backs up these assertions. Companies such as Monsanto, which create GM crops with Cry toxins, claim that the toxins are solely effective against certain insects and do not harm the environment, mammals, or human.

## 6 CONCLUSION

The potential of biotechnology for increased crop output has been shown by the widespread and successful adoption of GM biotech crops around the world. However, the development of insect resistance to Bt-cotton has occasionally caused worries about the fabric's weak resilience in light of potential insect resistance growth. However, the future of GM crops depends on the search for novel genes that, by working differently, could provide equivalent or more resistance in transgenic plants, which has led to the identification of several genes from various sources. When studied, many of these demonstrated a sizable potential for use in crop protection. The use of stacking genes, modified Bt-toxins, spider/scorpion venom peptides, vegetative insecticidal proteins, lectins, endogenous resistance mechanisms, and innovative techniques are thus future trends and opportunities for biotechnological applications to mediate crop protection against insects. The advantages and risks of using GM insect-resistant crops, particularly for developing nations and resource-strapped small-scale farmers, must be considered while utilising such tactics. The majority of research right now is concentrated on crucial genes and metabolic pathways that are fundamental to the biology of the insect pest and host-plant interactions. Being a complicated phenomenon, the host-pest connection, the discovery of these genes and the precise function of their function.

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**Conflicts of Interest**
The authors declared no conflict of interest.

**Author Contribution**
Sadek HE, Ebadah IM and Mahmoud YA contributed equally to this work including idea conceptualization, review topic critical discussion and development, writing, editing and approved the final version.

### Abbreviation List

| | |
|---|---|
| *B. thuringiensis* | *Bacillus thuringiensis* |
| CpTI | Cowpea trypsin inhibitor |
| GM | Genetically modified |
| GMOs | Genetically modified organisms |
| GNA | Galanthus nivalis agglutinin |
| Gy | Gray |
| H. armigera | Helicoverpa armigera |
| H. virescens | Heliothis virescens |
| H. zea | Helicoverpa zea |
| NBTs | New breeding technologies |
| nGMs | Newer genetic modification techniques |
| PI | Protease Inhibitors |
| QTL | Quantitative trait locus |
| RNAi | RNA interference |
| *S. littoralis* | *Spodoptera littoralis* |
| *S. litura* | *Spodoptera litura* |

*SIT, Sterile Insect Technique*
sRNA, Sequence-specific short RNA
TC, Tissue culture
UV, Ultraviolet

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