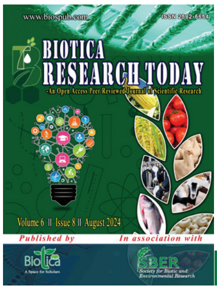

Bio. Res. Today 2024, 6(8):419-422

Biotica Research Today

e-ISSN: 2582-6654

August, 2024

Article ID: RT1709

Popular Article

Genetically Modified Insects

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Open Access

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Conflict of interests: The author has declared that no conflict of interest exists.

How to cite this article?

Patil, B.J., Katti, P., Supreeth, G.S., 2024. Genetically Modified Insects. *Biotica Research Today* 6(8), 419-422.

Abstract

Technological and scientific advancement has recently proved advantageous for raising crop production and improving public health. Genetic engineering technology is one example of such progress. In this regard, transgenic crops are an effective model. However, insect genetic manipulation is becoming increasingly significant. Applications of genetically modified insects are numerous and include managing agricultural pests, managing disease vectors in humans and animals, improving the efficiency of beneficial insects like silkworms and honey bees and industrial entomology. This technology is even now in its infancy in India. It is imperative that need-based research be done and that a safety framework is established in this field.

Keywords: Biotechnology, Genetic engineering, Sterile insect technique, Transgenic insects

Introduction

Insect pest management is mainly relying on chemical pesticides. Nevertheless, this strategy is facing mounting pressure globally and is detrimental to the ecosystem. Furthermore, it might be challenging to identify novel and effective pesticide molecules and the expense and time required for registration are rising. This has sparked an investigation into novel pest management techniques. Sterile Insect Techniques that are basically genetic approach of pest control are getting lot of importance in the recent past. An insect that has undergone genetic modification, either by "mutagenesis" or more precisely "transgenesis" or "cisgenesis," is known as a genetically modified insect (GM). Using genetically modified insects is a new and creative way to combat crop pest losses and insect-borne diseases that affect humans. An overview of genetically engineered insects and their possible uses is given in this article.

Strategies Involved in using Genetically Modified Insects under Field Conditions

Under field situation, several strategies are available for using GM insects for their effective usage. Based on the tendency of development or spread by the genetic character, the strategies are planned as: (i) Self-limiting strategies and (ii) Self-sustaining strategies (involves non-GM methods like *Wolbacia* infestation). Similarly, based on the favorable outcomes, again the strategies are divided as: (i) Population suppression, (ii) Population replacement and (iii) Others like flying needles. Also, the processes by which the heritable changes are done also used for strategies development and these involve: (i) Classical genetics, (ii) Methods using recombinant DNA and (iii) Paratransgenesis (Figure 1).

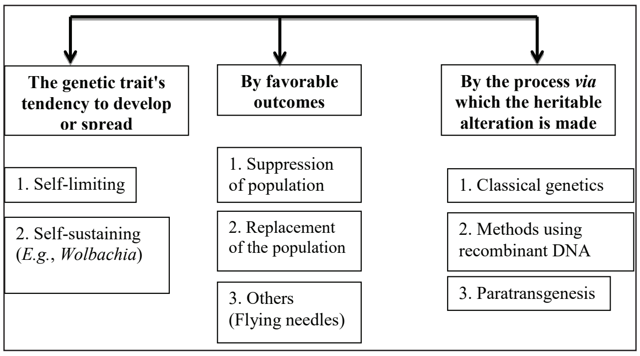

Figure 1: Flowchart showing the strategies of using GM insects under field conditions

Why GM Insects Approach is needed?

1. Community approach of pest management
2. In vitro synthesis of pharma products;
3. To develop virus-resistant insects;
4. For benefit of public health; and

Article History

RECEIVED on 05th August 2024 RECEIVED in revised form 25th August 2024 ACCEPTED in final form 26th August 2024

5. To improve economic aspects of insects like pest resistance, pollination efficiency in honey bees and good quality silk production (Gopinathan, 1992).

Background of Genetically Modified Insects

New World Screw-worm, *Cochliomyia homnivorax*, was the biggest threat for the cattle industry. According to Raymond Bushland and Edward Knipling's theory, sterilizing the males of this pest will inhibit the mating with the females and results in ceasing of oviposition (Joe, 2010). Several mutagens *viz*., ethyl methyl sulphonate, UV light and gamma irradiation were employed in sterile insect approach. E.F. Knipling was the pioneer of utilizing genetically engineered insects as pest management tools in genetic control (Joe, 2010).

Methods of Genetic Pest Management

1. Release of Insects carrying Dominant Lethal
2. Incompatible Insect Technique
3. Sterile Insect Technique
4. Maternal Effect Dominant Embryonic Arrest
5. X-Shredder

Sterile Insect Technique (SIT)

Sterile Insect Technique is one of the biological based insect pest management strategy where, sterlized males were released into the wild to mate with the wild females. These infertile males will compete with wild males and deprive them of females. This mating with sterile male will lead to absence of oviposition and ultimately resulting in collapse of population.

Release of Insects Carrying a Dominant Lethal (RIDL)

This is a variant of the sterile insect technique and includes release of genetically modified insects that are homozygous for a dominant lethal trait, rather than irradiating them (Alphey, 2014). These insects carrying the dominant lethal genes will mate with the wild females leading to expression of lethal traits in off-springs causing them to die prior to the maturity.

The Incompatible Insect Technique (IIT)

IIT and SIT are closely comparable approaches; however, IIT employs male *Wolbachia*-infected mosquitoes, which stave of them from copulating with wild-type females and generating healthy offspring, instead of releasing sterilized insects. One biological process that endosymbiotic bacteria, such as *Wolbachia*, may exhibit is called cytoplasmic incompatibility (CI).

Maternal Effect Dominant Embryonic Arrest (MEDEA)

MEDEA is a gene based technique which has both toxin and antidote. A female carrying MEDEA will express this lethal toxin in the progeny leading to their death. Contrastingly, if offspring have the same MEDEA, then antidote is produced leading the survival of progeny. Hence, if a female has both one MEDEA allele as well as non-MEDEA allele, leading to the 50% inheritance of the same to the progeny, half of the progeny having with MEDEA will survive and death of other half having non-MEDEA (provided MEDEA inherited from male).

Examples: malaria and dengue fever mosquitoes, diamondback moth, *Plutella xylostella* and medfly, *Ceratitis capitata*.

X Shredder (Chromosome Shredder)

Distorting the sex-ratio by using the X-chromosome shredding was found to be highly efficient as compared to the release of sterile males. In this process, X-chromosome bearing gametes are eliminated during male spermatogenesis leading to male biased population.

Example: Malaria mosquito, *Anopheles gambiae*.

Methods of Genetic Transfer

Physical Methods

1. Microinjection: Liquid material of tiny or nearly macroscopic level will be injected using a glass micropipette. Intercellular space may sometimes be included and the live cells are often targeted.

2. Lipofection: Also called as liposome transfection, where introducing genetic material is introduced into cell through liposomes, which are basically vesicles made up of phospholipid bilayers and will easily merge with the cell membrane.

3. Electroporation: The desired DNA or chromosomes are transferred into the bacteria through the pores, which will be opened momentarily when exposed to electric pulse

Biological Methods

1. Transposable Elements (Transposons or Jumping Genes)

Transposons were primarily reported by Barbara McClintock during her early career and earned her a Nobel Prize. Transposons are movable DNA segments that shift from one chromosomal position to another and are not fixed at a single genomic location (Liao *et al*., 2000). They move by means of a transposition mechanism. They have the power to change the genome's DNA content and, in the process, bring about mutations. The widely employed transposons in insect transformation are *piggyBac*, Mos 1, (mariner) Minos and Hermes. Transposons in *Drosophila* are *P* system and *hobo*.

2. Paratransgenesis

With this method, the flora in an insect's gut can be changed or entirely new flora can be added, yet the insect's genome remains unaltered. The kissing insect (*Rhodnius prolixus*), which spreads *Trypanosoma cruzi*, is one such instance. By manipulating the bacteria often present in the kissing bug's stomach, scientists are able to create an anti-trypanosome peptide called cecropin. This peptide does not harm the insect but is lethal to the pathogen. Green Fluorescent Protein (GFP) was used as marker for gene transfer.

Aspects of GM Insects' Regulations for Pest Management

International Initiatives

In a variety of industries, including GM crops, chemicals and medicines, a phased testing strategy is thought to be a suitable means of evaluating risk for novel technologies (Romeis *et al*., 2008). Below is a step-by-step guide for evaluating genetically modified insects.

a) Testing in a lab under controlled usage circumstances,

b) Field testing in a confined space,

c) Field release,

d) Pilot operational assessment.

Other regulatory initiatives include Cartagena Protocol on Biosafety (CPB) and European Food Safety Authority.

Applications of Transgenic Insects

A wide range of potential applications of transgenic insects are given in table 1.

Table 1: List of potential uses of transgenic insects

Insect species	Possibility of using Genetically Modified insects	Present status
Pink bollworm (*Pectinophora gossypiella*)	Improvements to the sterile insect method	Open field and programmatic scale trials
Yellow fever/ dengue mosquito (*Aedes aegypti* [Skuse])	Controlling dengue transmitting vectors	Open field trials
Mediterranean fruit fly (*Ceratitis capitata*)	Protein production	Laboratory research
Olive fly (*Bactrocera oleae*)	Sterile insect technique development	Laboratory research and contained field trials
Cabbage looper moth (*Trichoplusia ni*)	Production of recombinant antibodies	Laboratory research
Bombyx mori and other silkworms	Protein production	Laboratory research and potential contained commercial production
Honey bee (*Apis mellifera*)	Insecticide resistance	Laboratory research

Current Status of Transgenic Insects

1. Oxitec's Friendly™ Fall Armyworm: Carrying a Self-Limiting Gene

Friendly™ fall armyworms are produced exclusively by males thanks to a self-limiting gene. These Friendly™ males will locate and mate with wild females once they are released into the field. Their progeny get the self-limiting gene, which keeps the female offspring from living long enough to procreate.

2. Earliest Release of Genetically Modified in the United States, 2021

The males that have undergone genetic engineering possess a gene that is passed down to their progeny and causes the early death of female offspring. Instead of dying, male progeny will get the gene and pass it on to succeeding generations. The population of *Aedes aegypti* should decrease as more females perish.

3. First Ever Field Release of a Genetically Engineered, Self-Limiting Agricultural Pest Insect: Diamondback Moth (2020)

In an innovative move to safeguard crucifer crops, a genetically modified strain of *Plutella xylostella* (OX4319L) that is "self-limiting" and capable of producing male-only cohorts of moths for field releases was created. Upon release in the field, the mating between wild females and self-limiting males lead to inhibition of female progeny production.

Status of Transgenic Insects in India

1. First Ever Genetically-Modified Silkworm Awaits a Nod from GEAC

Andhra Pradesh State Sericulture Research and Development Institute (APSSRDI) has developed the first-ever genetically modified silkworm and awaits approval from the Genetic Engineering Appraisal Committee (GEAC) at the national level to go for further trials like Baculovirus-resistant transgenic silkworm.

2. GM Mosquito Trials to Control Dengue, Chikungunya Launched in India

On January 23, in Dawalwadi, Badnapur in the Jalna district of Maharashtra, open-air caged experiments were initiated to show the effectiveness of genetically modified mosquitoes in suppressing wild female *Aedes aegypti* mosquito populations that spread dengue, chikungunya and Zika.

3. Government may Swat GM Mosquitoes in India

Scientists have doubts regarding the technology's effectiveness. The introduction of GM *Aedes aegypti* mosquitoes through open field trials is scheduled to be denied by the government. By disrupting the life cycle of mosquitoes that carry the dengue, chikungunya and zika viruses, the new genetically modified technology intends to stop their spread in India.

Possible Risks of GM Insects

1. Instability of the introduced genes.
2. Horizontal gene transfer in the ecosystem.
3. Gene silencing.
4. Disruption of ecosystem services.
5. Poor fitness.
6. Environmental risks.

Conclusion

Transgenic crops have transformed agriculture by improving crop yield, safeguarding crops from pests and extending the shelf life of produce after harvest. Similarly, modern technology has progressed to the point that it is now possible to genetically alter insects in order to accomplish objectives related to industrial Entomology, public health and pest management. This technology is still in its infancy in India. Thus, further research in this aspect is necessary.

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