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Genetic Modification of Plants: An Emerging Technology

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ABSTRACT

Genetically modified (GM) plants can be created by adding specific DNA sequences obtained from the same plant species or different species. Which aims to achieve higher yields, disease and pest resistance, herbicide tolerance, production of antibodies, and other pharmaceutical molecules by manipulating gene expression to alter the protein properties. Insect resistant plants reduce the damage caused by pests and diseases. Herbicide glyphosate or glufosinate-tolerant GM plants gave promising results in combating weed. The properties of plants such as metal uptake, transport, accumulation, and detoxification of organic pollutants can be enhanced by genetically manipulating the fast-grown and introducing the responsible gene from the hyper accumulative species. GM plants can be used to produce cost-efficient and manageable drugs, vaccine, and biopharmaceuticals, if certain limitations are to be considered such as quality of final products, techniques for extraction and processing of biopharmaceuticals, and biosafety. Despite all these benefits, its adverse effects on the environment and human health have always been a matter of concern. The main limitation includes a horizontal transfer of the transgene to other species which may code for the specific antibiotic and herbicide resistance. It might be the possible transmission of resistance from the food products to the whole human population via intestinal bacteria. To address this several methods, need to be adopted to either keep away or eliminate marker genes from the transformed plants before growing in the field. Many scientists have come up with strategies to generate marker-free transgenic plants to give us safe and reliable GM technology.

Keywords:
Genetic Modification; Insect Resistance; Herbicide Tolerance; Plantibody; Selectable Markers

1. Introduction

Genetic modification of plants can be characterized as the alteration in structure, composition, or development of plants by inserting specific DNA sequences acquired from similar plant species or even from different varieties. The ultimate goal is to alter the biological properties of proteins by manipulating the expression of genes encoding it. For centuries plant-breeders and Farmers have been changing the genes of crop plants. Nonetheless, genetic modification varies from traditional plant breeding in terms of precise gene transfer and quite rapid technology, as it can circumvent biological barriers for genetic exchange and recombination across incompatible organisms by generating transgenes (Halford and Shewry, 2000; Holst-Jensen, 2009).

Transgenic plants are developed by two types of transformation method, *Agrobacterium*-mediated transformation method and Direct gene transfer methods. In direct gene transfer methods, DNA transferred by employing chemical, electrical or physical methods (Tsaftaris et al., 2000). Rapid advances in biotechnology have enabled the incorporation of gene sequences into the plant genome that offers new characteristics, such as pest resistance, herbicide tolerance, and so on. A large number of such Genetically Modified (GM) plants are being created, and many of them have hitherto been released into the market. Plant genetic engineering can suit many objectives including the improvement in agronomic traits like higher yield, disease and pest resistance, the production of antibodies and other pharmaceutical molecules, or the application of genetically altered plants in cross-breeding programs. (Elenis et al., 2008; Maessen, 1997).

The characteristics of crops pursued by genetic manipulation are not entirely different from those aimed by conventional plant breeding. However, due to the inclusion of direct gene transfer across phylogenetically distinct species in genetic manipulation, most traits that were traditionally difficult to develop were easily developed (Matin Qaim, 2009). There are three types of GM crops to be defined i.e. The first-generation GM crops which focus on improvements in agronomic traits, such as pests and disease resistance, herbicide tolerance, delayed ripening, that primarily benefit farmers and also the seed companies who have trailblazed the work. The second-generation GM crops include traits such as improved nutritional value, prime quality, good shelf life, and hypo-allergenic nature, which benefit consumers. While the development of third-generation GM crops is in progress to produce therapeutic drugs, vaccines, biosensors, and industrial products. (Yonekura-Sakakibara and Saito2006; Stewart and McLean, 2005; Naranjo and Vicente, 2008).

Opportunities for GM Crops are multifarious. Against the backdrop of declining natural resources, increasing productivity in global agriculture is important to ensure adequate accessibility of food as well as other raw materials for a rising population. GM crops can also be expected to bring environmental benefits and, like new seed technologies, can play an important role in poverty eradication and rural income growth in developing countries. (Qaim, 2009). Nevertheless, the term "genetically modified organisms (GMOs)" has become a contentious issue because its beneficial impacts to both producers and consumers are reaped by substantial biomedical threats and adverse effects on environmental (Zhang et al., 2016). In this review, we have summarized the great potential of GM technology towards agricultural benefits as well as pharmaceutical and industrial benefits and also highlighted some of the complexities of these new technologies.

2. Cost-benefit analysis of selected GM Crops

The safety concerns, cost of production and benefits or socio-economic impacts have always been a matter of intense discussion since genetically modified plants have been in the market (Majeed, 2018). It is necessary to analyse its cost and benefits to adopt or reject any new technology. Thus, to adopt GM technology, it is essential that this will help the producers economically. Because of that the cost of GM seeds compared to conventional seeds, cost of pesticides, labor and other relevant inputs needs to be minimal. Occasionally the economic benefits received from GM crops depend on the ability of farming, agriculture factors and geographical conditions. So, it may also be that this technique does not benefit farmers or they do (Flannery et al., 2004;

Kalaitzandonakes, 2003; Fulton and Keyowski, 1999). Many studies have shown that GM crops benefit farmers in terms of higher yield and savings in production, while some studies show its negative impacts related to high cost of technology and complicated methods (Pray et al., 2002; Huang et al., 2008; Morse et al., 2004; Qaim and Traxler, 2005; Sexton and Zilberman, 2011; Stone, 2012; Smale et al., 2009; Glover, 2010).

A meta-analysis of the impacts of GM crops by Klümper and Qaim (2014), which mainly concentrates on the herbicide tolerant (HT) soybean, maize and cotton and insect-resistant (IR) maize and cotton, shown that by adopting GM technology, effective control of pest and stopping the crop losses, there is a 21% increase in the crop yield. The quantity of pesticide used reduces by 37% and cost incurred on it by 39%. Although GM seeds are more expensive, its return can be found in savings from chemical and mechanical pest control. Thus, on an average GM technology benefits farmer by 69%. Increase in yield and reduction in use of pesticide are larger for insect-resistant crops compared to herbicide tolerant crops. Even yield and farmers benefits are larger in developing countries than developed countries (Klümper and Qaim, 2014).

The advantage of GM herbicide-tolerant (HT) plants is that it provides less expensive methods for the weed control and thus higher yield. The impacts of GM HT crops depend on the cost of herbicides used for the GM HT systems, amount of herbicides applied, the cost of GM HT technology and level of weeds present in fields. The use of GM HT crops facilitates a reduction in tillage techniques. It leads to farm level gain to farmers by reduction in cost of production and nil spent on weed control due to avoiding expensive herbicides. Since 1996-2016, the cost of technology for the production of GM HT soybeans including cost of GM seeds was 2-4 \$/hectare in Argentina, 7-25 \$/hectare in Brazil, 50-60 \$/hectare in Romania, 15-57 \$/hectare in US, 20-40 \$/hectare in Canada, 4-10 \$/hectare in Paraguay and the average gross farm income benefits obtained from the GM HT soybeans was 22.5 \$/hectare in Argentina, 32 \$/hectare in Brazil, 104 \$/hectare in Romania, 34 \$/hectare in US, 21 \$/hectare in Canada, 16.5 \$/hectare in Paraguay. On the whole, in 2016, GM HT technology leads to increase in gross farm income by \$ 4.37 billion, while there is an increase of \$54.6 billion from 1996 to 2016. From the overall gains obtained through GM HT soybeans, 45% of income is due to higher yield and 55% of income is due to cost savings. Cost of technology for the production of GM HT maize was 15-30 \$/hectare in US, 17-35 \$/hectare in Canada, 16-33 \$/hectare in Argentina, 9-18 \$/hectare in South Africa, 10-32 \$/hectare in Brazil and average gross farm income benefits obtained from maize was 28 \$/hectare in US, 15 \$/hectare in Canada, 108 \$/hectare in Argentina, 5 \$/hectare in South Africa, 38 \$/hectare in Brazil. On an average, gross farm income benefits increase by \$ 2.1 billion in 2016 and \$ 13.1 since 1996 to 2016. 34% of overall gains are due to higher yield and rest due to cost savings. GM HT cotton increase gross farm income by \$ 1.92 billion since 1996 while \$ 130.1 million in 2016. Other GM HT crops like canola and sugar beet deliver total benefits of \$ 559 million in 2016 and \$ 6.44 billion since 1996 to 2016. GM insect resistant (IR) crops reduce the loss of crops caused by insect and therefore increase in yield. Cost of technology for the production GM IR maize and cotton was 17-32 \$/hectare (insect resistance to corn boring pests)/ 22-42 \$/hectare (insect resistance to corn rootworm and 111 \$/hectare respectively in US, 10-30 \$/hectare and 240 \$/hectare respectively in Argentina. The Cost of technology for the production of GM IR cotton was 207 \$/hectare in India. Aggregate income benefits obtained from the GH IR maize and cotton was \$ 38,509 million and \$ 5,430.5 million respectively in US, \$ 1,108.8 million and 921 million respectively in Argentina.

Aggregate income benefits obtained through GM IR maize in Brazil was $ 6,222.9 million. Aggregate income benefits obtained through GM IR cotton in India was about $ 21,121.7 million. On the whole, gross farm income benefits obtained through GM IR maize was $ 4.81 billion, and through GM IR cotton was $ 3.7 billion in 2016, while $ 50.6 billion through GM IR maize and $ 54 billion since 1996 to 2016. Aggregate impact of GM soybeans, maize, canola and cotton results in net increase in gross farm income by $ 18.2 billion in 2016 while increase by $ 186.1 billion since 1996 to 2016. Farmers of the developing countries gets more income benefits compared to developed countries that is 55% of the total global income benefits in 2016. Over the period of 1996 to 2016 they get income benefits up to $ 96 billion. Cost of GM technology was 29% of the total technology benefits. In developing countries, it was 20% of the total technology gains while in developed countries it was 36% of the total technology gains (Brookes and Barfoot, 2018). Economic benefits obtained from the GM crops since 1996 to 2016 was $ 80.3 billion in USA, $ 23.7 billion in Argentina, $ 21.1 billion in India, $ 19.8 billion in Brazil, $ 19.6 billion in China, $ 8 billion in Canada, $ 1.7 billion in Paraguay, $ 4.8 billion in Pakistan, and $ 7.1 billion in other countries (ISAAA, 2017).

3. Application of Genetically Modified Plants

Given the low agricultural productivity, malnutrition, poverty, hunger, food security issues in many developing countries, technological advances such as GM technology, are likely to address some of these issues (Adenle, 2011). There is mass production of GM crops in developing countries which provides compelling proof of benefits to small scale farmers and, indeed, the rapid growth of GM cultivated area in these countries speaks of the value proposition for small scale growers and also, genetically modified plants have been used globally for industrial chemicals, pharmaceuticals, crop improvement and beyond (Lewis, 2016). Thus, GM plants offer potential applications for herbicide tolerance, pest resistance and industrial products.

3.1. Insect Resistant Genetically Modified Plants

It was estimated that about 30 % of crops damaged by pests and diseases. If we reduce these causes of damage, we can improve the yield and productivity of the crops. For the protection of crops, Integrated Pest Management (IPM) is more desirable instead of relying on harmful chemical pesticides. But, due to a lack of efficient tools and techniques, it would not be generally accepted by the farmer. Insect resistant transgenic plants overcome this problem and provide more strength and stability to IPM. The first-generation insect resistant transgenic plants generated by introducing crystal protein (Cry protein) from the bacterium *Bacillus thuringiensis* (Bt) to the plants. It was reported that about 30 different varieties of Bt, produce more than 50 Cry proteins and each Cry protein is selective for various groups of insects. During transformation, the Cry gene inserted singly or in conjugation with other non-Bt insecticidal genes. Nowadays, such types of Bt genes are inserted which codes for the two Bt proteins. In commercially available Bt-crops, Cry protein is expressed in all parts of plants. When insect larvae feed on the transgenic plants, Cry protein is ingested along with plant tissue, which damages the midgut epithelium by making it porous which leads to the death of the insects. In the year 1996, three insect resistant crops i.e. Bt-corn against European corn borer, Bt-potato against Colorado potato beetle and Bt-cotton against tobacco budworm and bollworm were commercialized in the U.S.A. Due to benefits obtained from transgenic insect resistant plants such as increasing yield and effective control of insects, the global area under the transgenic insect resistant crops increase from 1.1 million hectares in 1996 to more than 11.5 million hectares in 2000. But for the commercial release of any transgenic insect resistant plant, we need to take into consideration several biosafety measures such as cross-pollination, effects on non-target organisms, insect resistance, toxicity, allergic effects, etc. (Manjunath, 2002). Several studies have reported decreased use of pesticides due to the cultivation of Bt crops designed to resist specific insect pest. Many crops have been genetically engineered to express insecticidal Cry protein from Bacillus thuringiensis, such as maize, tobacco, rice, etc. However, papaya, potatoes, and squash have been genetically engineered to resist viral pathogens such as cucumber mosaic virus which infects various plant species (Reddy, 2015).

Transgenic eggplant having a wild type of Bt genes shows not as much resistance to insects because of the less expression level of the transgene. Arpaia and co-workers reported the formation of transgenic eggplant commercial F1 hybrid 'Rimina' by introducing the mutagenized gene of *B. thuringiensis* Berl. Var Tolworthi (Cry III B) via *Agrobacterium tumefaciens* mediated transformation. This transgenic eggplant shows the sufficient level of the transgene expression to control the Colorado potato beetle and provide complete protection from the Colorado potato beetle (Arpaia et al., 1997).

3.2. Herbicide-Tolerant Genetically Modified Plants

Weed is one of the major reasons responsible for the loss of plant yield. The use of herbicide-tolerant transgenic plants for weed control is a better option rather than hand weeding, mechanical tilling, hoeing, and chemical methods. Other than herbicide-tolerance, it has many advantages as they are agronomically and economically responsible. Herbicide-tolerant GM crops are those that have been genetically modified so that they are not harmed when sprayed with wide-spectrum herbicides while the weeds that infest the crop are destroyed. Many herbicide products, including Roundup, contain glyphosate as an active ingredient that kills plants by inhibiting EPSPS involved in shikimate pathway of plants. Animals get aromatic amino acids from their diet so they do not have this shikimate pathway. The trait for herbicide tolerance was developed because at that time the herbicides used on the crop were extremely toxic and not really efficacious against many weed species. Therefore, development of herbicide tolerant crops can reduce both environmental and health risks, and also give the farmers an agricultural edge ((Reddy, 2015).

Glyphosate herbicide resistant soy and canola are produced in 1996 and cotton in 1997. According to the 2012 report of International Service for the Acquisition of Agri-biotech Applications (ISAAA), global cultivation of transgenic plants containing at least one herbicide-tolerance trait was 100.5 million hectares. Glyphosate (N-[phosphonomethyl] glycine) is commonly used as herbicide for agricultural practices. Glyphosate is also readily absorbed by some important crop plants other than weeds. Thus, herbicide resistant transgenic plants were created. The introduction of transgenic glyphosate-tolerant soybean (Roundup Ready® or RR soybean) was very successful and readily accepted by the farmers all over the world. Thus, herbicide-tolerance plant acreage increases over the years. For the creation of glyphosate-tolerance crops, either we can introduce a transgene code for a glyphosate-insensitive target enzyme which deactivates the glyphosate (Feng et al., 2010; Pollegioni et al., 2011). Glyphosate solely acts on 5-enolpyruvulshikimate-3-phosphate synthase (EPSPS) and strongly inhibits EPSPS. Glyphosate-tolerance crops engineered by introducing glyphosate-insensitive EPSPS. Glyphosate tolerant EPSPS is found in many microorganisms such as *Agrobacterium sp*. CP4, *Achromobacter sp*. LBAA etc. These are not affected by glyphosate and retain their catalytic activities in the presence of glyphosate. By using this technique many glyphosate-tolerant transgenic plants were generated. Some of these are Roundup Ready® soybean, Roundup Ready® cotton, Roundup Ready® canola, Roundup Ready® sugar beet, and Roundup Ready® alfalfa. Other than glyphosate-tolerant, glufosinate-tolerant plants also generated and gave promising results (Huang et al., 2015).

3.3. Abiotic Stress Resistant Genetically Modified Plant

Phytoremediation is the technology that uses the plants to clean off the hazardous contaminants including heavy metals, metalloids, or organic pollutants from the soil, water, and air. Trees are the first to be selected for phytoremediation because of their more expanded roots, high water uptake, fast growth, a large amount of production of biomass, and longer life cycle. For example, poplar plants, which are used for the removal of elements like citrazine, tricholoroethylene, and selenium (Burken and Schnoor, 1997; Newman et al., 1997; Pilon-smits, 1998). The properties of plants such as metal uptake, transport, accumulation and detoxification of organic pollutants can be enhanced by genetically manipulating the fast-grown species and introducing the responsible gene from the hyper accumulative species (Raskin, 1996; Pilon-smits and Pilon, 2002; Terry et al., 2003; Van Huysen et al., 2004; Ellis et al., 2004; LeDuc et al., 2004). Transgenic poplar plants that overexpressing modified bacterial mercuric reductase (merA) or Y-glutamyl cysteine synthetase (Y-ECS) used for the accumulation of heavy metals contamination of mercury (Hg) or cadmium (Cd) respectively (Cherian and Oliveira, 2005).

An increase in heavy metal concentration in the soil causes hazardous effects to human, animal, and plant health. Human activities such as mining, industrial emission, disposal of sewage and industrial wastes, fertilizers and pesticides, etc., are responsible for the contamination of soil with heavy toxic metals. Genetically modified tobacco expressing antisense sequence of proline dehydrogenase gene shows tolerance to several heavy metals such as nickel (Ni), cadmium (Cd), and lead (Pb). Genetic modification of heavy metal hyperaccumulator species *Thlaspi caerulescens* L. by introducing foreign catalase gene, modify its accumulation properties. Introduction of heavy metal responsive gene *Phaseolus vulgaris* stress-related gene (PvSR2) from the French bean to tobacco, increase the cadmium tolerance in the tobacco plant (Safavi and Asgari, 2011).

3.4. Genetically Modified Plant as a Source of Biopharmaceutical Products

Plants used as biofactory for the production of drugs, vaccines, and biopharmaceuticals. Biopharmaceuticals derived from the plants are cost-efficient, easy to maintain, and easy to produce to a high level and bring easier than animals. There are three types of drugs we can derive from the plants, Antibodies, biopharmaceutical medicines, and vaccines (Walmsley and Arntzen, 2000). Currently, molecular farming opens up a new field in worldwide for the creating a transgenic plant used as a bioreactor for the production of medicinal proteins (Daniella et al., 2001; Ma et al., 2003; Larrick et al., 1998; Jaeger et al, 2000). The technology of producing antibodies in plants is referred to as a 'plantibody'. Plants as a source of antibodies rather than microorganism is a better option because of safety concerns related to human health and product and low cost of production. However, some limitations must be considered such as the quality of final products, techniques for the extraction and processing of biopharmaceuticals, and biosafety (Hashemzade et al, 2014).

Transgenic plants are used to make edible vaccines for some infectious diseases (Kant et al., 2011; Vianna et al., 2011; Yoshida et al., 2011; Sharma and Sood, 2011; Twyman et al., 2012). The first edible vaccine was created in 1990 by introducing gene for surface protein A from *Streptococcus mutans* in tobacco plant. Edible vaccines can be a better alternative to traditional vaccines. Edible vaccines have many advantages over traditional vaccines such as deep-rooted cultivation, cost-effective production, easy to store, maintain and distribute, least health concerns due to human pathogens and toxins, oral delivery, etc. Significant success has been achieved to express edible vaccines in plants like tobacco, tomato, maize, rice, etc. (Kumar et al., 2013).

As the field of creating transgenic plants, expressing proteins for pharmaceutical drugs is emerging, we can produce the pharmaceutical molecules such as single-chain variable fragments (ScFv) or single-chain antibodies in plants. Human epidermal growth factor receptor-2 (HER-2) is an oncogene responsible for the cause of breast cancer. Galeffi and co-workers described the first production of ScFv against the HER-2 antigen in plants. The transient and stable system used for the expression of ScFv-αHER-2 in *Escherichia coli* and plants. How efficiently these antibodies work on clinical specimens was analyzed by the Flow Cytometry. Its positive result suggests that in future ScFv-αHER-2 produced in plants may use to develop anticancer drugs (Galeffi et al., 2005).

4. Safety Concerns Regarding Genetically Modified Plants

In addition to the recorded advantages of GM plants, there are few documented cases of possible negative impacts of GM plants. Major concerns about GM plants can be categorised into four different broad sections: agricultural concerns, food safety and health concerns, environmental concerns, and socioeconomic issues. Environmental concerns posed about GM plants include the adverse effects on non-target organism, invasiveness, unexpected variability and development of resistive pest and diseases (Oh and Ezezika, 2014). Other major challenge that could obstruct access to GM plants deal with issues of intellectual property rights (IPR). GM technology is currently being implemented to the production of major commercial crops like maize, cotton, and soybeans, clearly making the biggest profits for biotech companies. However, these biotech companies are often reluctant to invest in developing countries (Adenle, 2011). Given a number of concerns, it is prerequisite to enable the assessment of GM plants on rational and scientific basis using accurate, reliable scientific and technical information, effective biosafety regulatory frameworks, policies, legal tools and decision-making processes. Therefore, risk assessment of GM plants on biodiversity is done by following these four steps: hazard identification, Exposure assessment, hazard characterization, and risk characterization, which can be followed by risk management (Yogo, 2016).

4.1. Effects of Genetically Modified Plants on Soil Microorganism

It has been possible to create new transgenic plants with improved characteristics such as high yield, pest and disease resistance, etc. through recent advances in biotechnology and genetic engineering. But its adverse effects on the environment and human health are a matter of concern, especially effects on our soil resources which contain large soil microbial communities. The main limitation and concern for cultivating transgenic plants is the horizontal transfer of the transgene to other species. According to many scientific data, the successful transfer of transgene borne antibiotic resistance genes to bacteria may be inescapable. If the origin of the transgene sequence was prokaryotic than it was easy for them to integrate with the bacterial genome and thus increase the chances of transfer of gene to bacteria. Demanèche et al., (2008) assessed the risk of transfer of antibiotic resistance gene from the transgenic plant to the soil bacteria. They selected the transgenic corn Bt176. This is produced by inserting the bla gene with a bacterial promoter that encodes for the TEM 116 beta-lactamase and provides resistance to ampicillin. They collected the soil samples from where the transgenic Bt176 corn and non-transgenic corn was grown in the separate field for the 10 years and then assessed for the diversity of bla genes in the soil bacteria but like all other previous studies they did not observe any evidence that confirm the transfer of bla TEM116 gene from transgenic plant to the bacteria. Even if it gets transferred and remains undetected, doesn't affect the soil bacterial community structure. Risk due to the transfer of bla TEM116 to soil bacteria is neglected because an abundance number of genes are already present in the soil bacteria and undergo constant evolution. However, bla TEM116 gene acquired from the transgenic plant would not have selective advantages relative to other resistant bacteria. Biao Liu and co-workers presented the review on the effects of transgenic plants on soil microorganisms. Transgenic plants directly or indirectly affect the soil microbes as its direct effects are due to the accumulation of transgene protein in the environment and indirect effects are due to change in metabolic pathways that lead to the formation of modified plant proteins and root exudates. Pest and insect resistant chemicals may have toxic effects on non-target soil microorganisms. Transgene proteins such as Bt toxin, T4 lysozyme enters in the soil by root exudates and deposition of leaf litter in soil and maintain their biological activity in the soil. Root exudates secreted from the transgenic plants responsible for the change in rhizospheric activities and thus affect the microorganism's lives in its rhizosphere by increasing or inhibiting their growth (Bardgett et al., 1999; McGregor and Turner, 2000; Saxena et al., 1999). Herbicide resistant rice plant containing the P450 cyP1A1 gene exuded metabolites of herbicide from the roots. But still, it is difficult to say that the above -said effects on soil ecology are solely due to transgenic plants. Because transgenic plants take a long time to make changes in the soil. Several transgenic plants show transient changes and, in some cases, it shows adverse effects only at the specific growth stages or when viable plants are present in soil (Demanèche et al., 2008; Liu et al., 2005; Mina et al., 2007).

4.2. Marker Free Genetically Modified Plants

Selectable marker genes used for the identification and selective regeneration of the transformed plant tissue from the non-transformed plant tissue. Selectable marker genes codes for the specific antibiotic and herbicide resistance. The use of such an antibiotic and herbicide resistance gene for the selection is a matter of concern. If such genes enter in the environment or the food chain will become a threat to the ecosystem or human health. It might be possible the transmission of resistance from the food products to the whole human population via intestinal bacteria. By keeping in mind public concern and human health safety, genetic marker-free plants should be made. Several ways are adopted to either keep away or eliminate marker genes from the transformed plants before growing in the field. Hypothetically, it should be possible to use more efficient regeneration methodologies instead of selectable markers for the screening of desirable genes. Use such selectable genes that have the least negative effects on the ecosystem or human health. Separate introduction of two transgenes, one of them carrying a gene for desired character and other for selection purpose. Site-specific recombination, transposable elements, or homologous recombination also used to remove the selectable marker from the complete transgene integrated with the host genome after a successful selection process (Doshi, 2010).

Chong-Pérez and Angenon gave strategies for generating marker-free transgenic plants. It involves the introduction of two transgenes delivered by two separate DNA molecules which get segregated in progeny. After growing on selective medium containing antibiotic, survived tobacco callus cells are transferred to non-selective medium and allowed to lose selectable marker gene by homologous recombination. In site-specific recombination process, site-specific recombinases cleave the DNA at a particular site. Generally, between the recognition sites in a direct repeat and remove the undesirable transgenic part from the nuclear genome of plants (Chong-Pérez and Angenon, 2013).

5. Conclusion

GM crops have proven to be agronomically and economically responsible as they benefit not only growers but also consumers and the country's main economy. GM crops can overcome many current challenges in contemporary agriculture, and can also make significant contributions in the pharmaceutical and industrial sectors. However, we need to consider several biosafety measures such as toxicity, effects on non-target organisms, allergic effects, etc. In conclusion, we can say that, despite the controversies, GM technology is reliable, safe, effective, and essential to satisfy the future food need and predilections.

Author Contribution

The first and second author made equal contribution to this paper. Krishnaben Desai performed literature search and drafting of manuscript, while Nainesh Modi read the manuscript thoroughly, gave appropriate comments, and gave final approval.

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Abbreviations

GM: Genetically Modified, USA: United States of America, IPM: Integrated Pest Management, Cry protein: Crystal protein, Bt: Bacillus thuringiensis, EPSPS: 5-Enolpyruvulshikimate-3-Phosphate Synthase, ScFv: Single-Chain Variable Fragments, HER-2: Human Epidermal Growth Factor Receptor-2, GUS: β-Glucuronidase, ACC: 1-Aminocyclopropane-1-Carboxylic Acid, ECS: Embryogenic Cell Suspensions, PCR: Polymerase Chain Reaction, DNA: Deoxyribonucleic Acid, T-DNA: Transfer DNA, bla: Beta-Lactamase

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