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The process of genetic engineering in plants

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Abstract:

Genetic engineering in plants has emerged as a revolutionary tool in agriculture, enabling the modification of plant traits to enhance crop productivity and sustainability. This review provides an in-depth examination of various genetic engineering techniques, their applications, and the implications for agriculture and the environment. The study highlights key findings and future directions in plant genetic engineering.

Keywords:

Agriculture, Bombardment, CRISPR, Genome, Transgenic.

1. Introduction:

Addressing the escalating global population and the growing importance of maximizing crop yields on limited land is crucial.(4) This challenge is exacerbated by increasing demands for food safety, sustainability, reduced agricultural inputs, and pesticide reduction. In response to these pressures, genetic modification of crops has emerged as a viable solution.(13) The advent of genetic engineering, facilitated by advances in molecular and cellular biology, enables the incorporation of desired features from diverse species into crop plants.(6) This technology marks a significant departure from traditional plant breeding, allowing the transfer of genes between species rather than restricting modifications to similar plant types.(14) "Genetic engineering" refers to the purposeful manipulation of genes, involving processes like gene transfer and alterations in gene sequences.(2) Specifically, genetic engineering utilizes recombinant DNA technology to induce targeted changes in plant or animal gene sequences, with the ultimate goal of introducing novel features.(18) Commonly known as genetic modification, this technique involves manually introducing additional DNA to an organism, resulting in the creation of genetically modified organisms (GMOs).(3) Recombinant technology is seen as a potential solution to agricultural challenges posed by biotic and abiotic stressors.(17)

2. Genetic engineering techniques in plants/gene transfer strategies:

2.1. Vector-mediated gene transfer:

(A.) Agrobacterium-Mediated Gene Transformation

2.2. Direct or vectorless DNA transfer:

(A.) Gene Gun or Particle Bombardment (B.) Electroporation

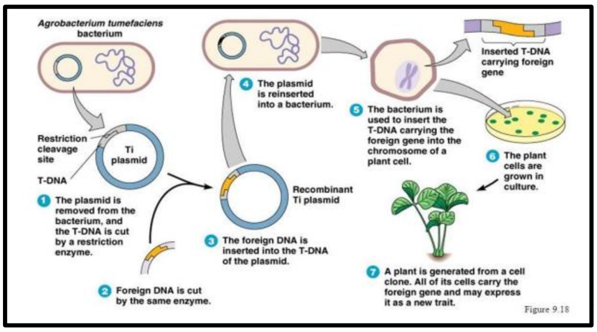
(C.) Microinjection

2.3. Vector-mediated gene transfer:

(A.) Agrobacterium-Mediated Gene Transformation

One approach to genetic engineering exploits Agrobacterium tumefaciens, a naturally occurring soil bacterium acting as nature's genetic engineer. This bacterium induces tumorous growth, known as 'Crown gall,' on wounded plants by integrating a DNA fragment (T-DNA) into the plant genome. The T-DNA, carrying genes causing tumor growth and producing compounds for bacterial nutrition, resides in a plasmid, enabling its isolation and modification. Modified

Agrobacterium can be used to infect plant material, leading to the regeneration of genetically modified plants from tumor-like clumps (callus) through the application of plant hormones.(8)



2.4. Direct or vectorless DNA transfer:

(A.) Gene Transfer Using Gene Gun or Particle Bombardment Technique

Particle bombardment, known as biolistics, involves coating DNA onto microscopic gold particles and introducing them into plant cells with helium gas bursts. This technique has proven successful, especially in producing genetically modified cereals. (8)

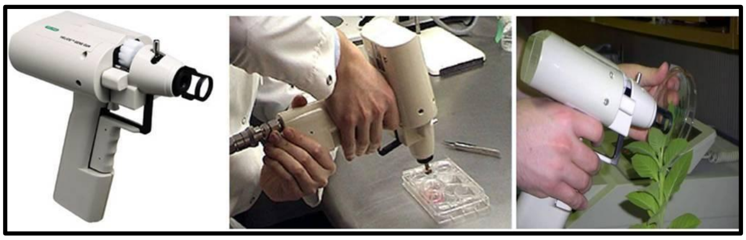


Figure. 2: Gene gun HeliosTM by BioRad is used to transfect cells in cultures and plant leaves

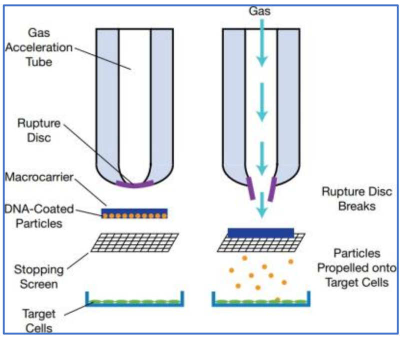


Figure. 3: Gene gun or particle bombardment delivery device can deliver transgenes to cells.(7)

(B.) Gene Transfer by Microinjection Technique

Microinjection utilizes injection needles to deliver DNA into tissues, causing minimal damage to cells directly in contact with the injected DNA. This method allows precise and efficient delivery of DNA into intact cells adjacent to wounded sites, increasing the chances of integrative transformation. (8)

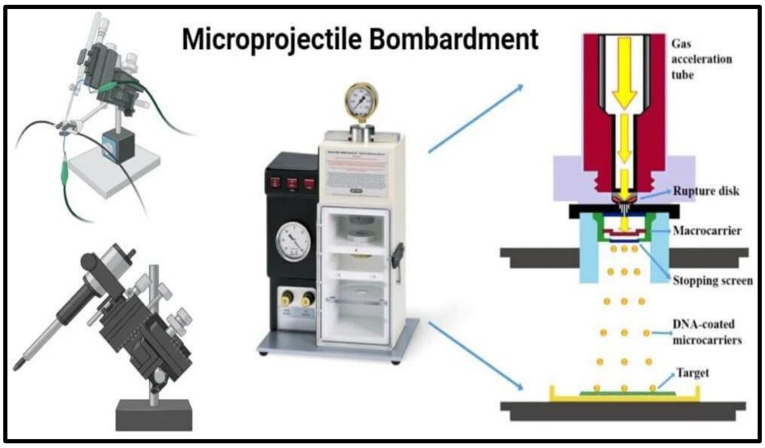


Figure. 4: Microprojectile Bombardment

(C.) Electroporation

Electroporation involves using electrical pulses to temporarily permeabilize cells, facilitating the entry of DNA into cells. Successful gene transfer through electroporation has been demonstrated in various plants, making it applicable to both monocot and dicot protoplasts.(8)

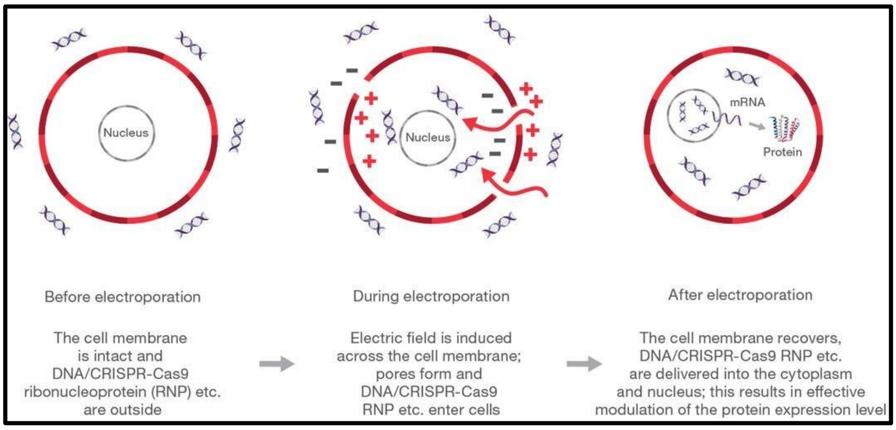


Figure. 5: Physical transfection methods; electroporation

3. Gene editing technologies

3.1. Clustered regularly interspaced short palindromic repeats system (CRISPR/Cas):

The CRISPR/Cas system, derived from bacterial immune systems, involves two components: CRISPR-associated protein 9 (Cas9) and a single guide RNA (sgRNA). These components collaborate to perform genome editing by creating double-strand breaks in target DNA. CRISPR/Cas9 has become a preferred gene editing method due to its versatility, simplicity, and ease of use. However, concerns exist regarding off-target effects and the potential environmental impact, especially with the proposal to use CRISPR/Cas for creating "gene drive" mechanisms. (15)

3.2. Zinc finger nucleases:

Zinc finger nucleases aim to deliberately alter the genetic makeup of organisms by changing DNA sequences. This technique involves the use of zinc finger proteins to recognize specific DNA sequences, coupled with the FokI enzyme to induce DNA breaks. Successful implementation involves transforming plant cells, followed by the selection of transgene-free lines. (15)

3.3. Oligonucleotide directed mutagenesis (ODM):

ODM aims to introduce small, predetermined changes to specific gene sites, either altering gene function or preventing its production. This method utilizes synthetic oligonucleotides with sequences nearly identical to the target gene, inducing site-specific DNA changes through the cell's own DNA repair mechanism. (15)

3.4. Cisgenesis and intragenesis:

Cisgenesis involves inserting DNA sourced from the same or closely related species, recreating the exact sequence of a gene from a donor organism. Intragenesis, on the other hand, utilizes a composite gene sequence from different genes of closely related species.(15)

4. Transgenic plant development:

The first transgenic plants were reported in 1983, marking the beginning of genetically engineered plant production in laboratories. Transgenic plants are created by altering the genetic makeup in a laboratory, typically by adding one or more genes to a plant's genome, focusing on the nucleus of plant cells.(1)

4.1. Application in agriculture:

Genetically modified (GM) technology has been employed to develop various crop plants, offering benefits such as improved production yield, reduced transportation costs, and enhanced nutritional content. Commercially produced GM crops in countries like the USA and Canada have primarily focused on conferring resistance to pests, insects, and viruses, as well as tolerance to specific herbicides. While these traits benefit farmers, the impact on consumers, beyond potential cost reduction, is limited. Future developments may introduce GM crops addressing malnutrition concerns within the next decade.(1)

4.2. Role of genetic modification in crop improvement:

Genetic modification of crops not only enhances nutritional content but also provides essential nutrients for healthy human living.(9) Examples include Bt maize crops and Golden Rice, addressing Vitamin A deficiency and potentially saving lives.(19) While potential risks of GM crops remain, especially in unproven scenarios, their deployment is deemed ethical when considering severe nutrient deficiencies and life-threatening conditions.(20)

4.3. Application of genetic engineering:

Genome editing has been utilized to enhance nutritional quality, resistance, and productivity in various plant species, resulting in transgene-free genetically modified plants.(21) Field tests have demonstrated the effectiveness of genetically modified apples, preventing apple scabs.(5) Additionally, genetic engineering has been applied to alter plant products' primary and secondary compositions, producing crops with desired qualities such as alkaloids, antibiotics, enzymes, flavonoids, pigments, and proteins.(10)

4.4. Environmental impact:

The negative impact of GM crops on the environment and ecosystems is a significant concern. Studies have highlighted potential risks, such as the impact of Bt toxins on non-pest species like the monarch butterfly, gene transfer leading to herbicide-resistant "superweeds," and cross-pollination between GM and non-GM crops. Contamination of indigenous seeds poses threats to biodiversity and wildlife. (8)

5. Future scope:

The acceptance of transgenic crops depends on positive public perception, compliance with regulations, rigorous biosafety research, and successful field trials. Challenges include addressing consumer concerns, defining clear guidelines and regulations, and managing potential lawsuits.(11,12) Despite these challenges, successful developments in agronomic traits and ongoing research in developing countries are optimistic indicators. The need to feed a growing world population, adapt to climate change, and produce plant-based industrial and pharmaceutical products should drive further research in GM plants.(16)

6. Conclusion:

Genetic engineering holds promise for improving agricultural production and environmental sustainability. The commercialization of transgenic plants is expected to expand, encompassing a wider range of plant species and traits. While potential benefits include increased stress tolerance, improved post-harvest processing, and modified food quality, caution is warranted. Rigorous testing, public education, and clear labeling of genetically modified foods are essential to making informed decisions. The precautionary principle should guide the release and regulation of GMOs, ensuring thorough understanding of their effects before production, sale, or consumption.

7. References:

(1) Abdul Aziz, Mughair & Brini,Faical & Rouached Hatem & Masmoudi Khaled. (2022). genetically engineered crops for sustainably enhanced food production systems. Frontiers in Plant Science, November. DOI 10.3389 fpls.2022.1027828. Pubmed ID 36426158.

(2) Altman A, Hasegawa PM. Eds (2011). Plant biotechnology and agriculture: prospects for the 21st century. Academic press.

(3) Bruce D, Bruce A (2014). Engineering genesis: ethics of genetic engineering in non-human species. Routledge.

(4) Bruinsma J (2009). June. The resource outlook to 2050: by how much do land, water and crop yields need to increase by 2050. In Expert meeting on how to feed the world in (Vol. 2050, pp. 24-26).

(5) Clark, E. A., 2006. Environmental risks of genetic engineering. Euphytica, 148(1-2), Pp. 47-60. https://doi.org/10.1007/s10681-006-5940-x

(6) Conner AJ, Jacobs JM (1999). Genetic engineering of crops as potential source of genetic hazard in the human diet. Mutation Research/Genetic Toxicology and Environmental Mutagenesis, 443(1-2), pp.223-234.

(7) Demirer, G. S.; Landry, M. P. (2017). Delivering genes to plants. Chemical Engineering Progress. 113(4).

(8) Jan, Masrat & Shrivastava, Mukta. (2017). genetically modified plants: a systematic review. International Journal of Recent Scientific Research. 08. 16471-16481. 10.24327/ijrsr.2017.0804.0155.

(9) Kettenburg, A., Hanspach, J., Abson, D. (2018). From disagreements to dialogue: Unpacking the golden rice debate. Sustain Sci. 13 (5), 1469-1482. doi: 10.1007/s11625-018-0577-y

(10) Lee, J. M., and an, G., 1986. Industrial application and genetic engineering of plant cell cultures. Enzyme and Microbial Technology, 8(5), Pp. 260-265.https://doi.org/10.1016/0141-0229(86)90019-0

(11) McDougall, P. (2011) The Cost and Time Involved in the Discovery, Development and Authorisation of a New Plant Biotechnology Derived Trait. Consultancy Study for Crop Life International by P McDougall, Midlothian, 1-24.

(12) Monsanto (2005) Annual Report for Fiscal Year Ended August 31, Form 10-K. U.S. Securities and Exchange Commission, Washington DC. http://www.monsanto.com/investors/documents/pubs/2005/mon_2005_10-k.pdf

(13) National Research Council (2010). The impact of genetically engineered crops on farm sustainability in the United States. National Academies Press.

(14) Phillips MJ (2013). Agricultural biotechnology issues. In US Programs Affecting Food and Agricultural Marketing (pp. 443-470). Springer, New York, NY.

(15) Raina, Meenakshi & Pandotra, Pankaj & Salgotra, Romesh & Ali, Sajad & Mir, Zahoor & Bhat, Javaid & Ali, Ajaz & Tyagi, Anshika & Upadhyay, Deepali. (2018). Genetic Engineering and Environmental Risk. 10.1007/978-3-319-64501-8_4.

(16) Singh, Hari & Singh, Bharat. (2014). Genetic Engineering of Field, Industrial and Pharmaceutical Crops. American Journal of Plant Sciences. 05. 3974-3993. 10.4236/ajps.2014.526416.

(17) Snow AA, Andow DA, Gepts P, Hallerman EM, Power A, Tiedje JM, Wolfenbarger LL (2005). Genetically engineered organisms and the environment: current status and recommendations 1. Ecological Applications, 15(2), pp.377-404.

(18) Sprink T, Eriksson D, Schiemann J, Hartung F (2016). Regulatory hurdles for genome editing: process-vs. Product-based approaches in different regulatory contexts. Plant cell reports, 35(7), pp.1493-1506.

(19) Swamy, B., Samia, M., Boncodin, R., Marundan, S., Rebong, D., Ordonio, R., et al. (2019). Compositional analysis of genetically engineered GR2E "Golden rice" in comparison to that of conventional rice. J. Agric. Food Chem. 67 (28), 7986-7994. doi: 10.1021/acs.jafc.9b01524

(20) Vega Rodríguez, A., Rodríguez-Oramas, C., Sanjuán Velázquez, E., Torre, A., Armendáriz, C., Iruzubieta, C. (2022). Myths and realities about genetically modified food: A risk-benefit analysis. Appl. Sci. 12 (6), 2861. doi: 10.3390/app12062861

(21) Zhang, Y., Iaffaldano, B. and Qi, Y., 2021. CRISPR ribonucleoproteinmediated genetic engineering in plants. Plant Communications, 2(2), and 100168. https://doi.org/10.1016/j.xplc.2021.100168