

Agroinfiltration in Fruit Crops: A Fast-Track to Genetic Enhancements

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Introduction

In the context of plant genetic manipulation, stable integration of foreign genes into the plant genome is not always a prerequisite for gene expression. Instead, the inherent cellular machinery responsible for gene expression and protein translation can effectively operate on transiently introduced genetic elements, such as genes carried by vectors. This property is harnessed in a technique known as agroinfiltration, which stands as a remarkably versatile approach that expedites research in laboratory settings while also serving as an exceptional platform for large-scale production of valuable compounds within plants.

Agroinfiltration, facilitated by recombinant DNA techniques, offers the distinctive advantage of leaving the host plant's genome entirely unaltered. Consequently, there is typically no need to generate a subsequent generation of plants, as the plant materials involved in the agroinfiltration process are usually discarded after utilization. This method thus permits precise and temporary genetic modifications, ensuring that the original genetic makeup of the plant remains intact while enabling rapid and efficient gene expression for research and industrial purposes.

Advanatges

Agroinfiltration stands as a valuable research tool renowned for its capacity to swiftly screen and evaluate genes of interest. In contrast to the protracted timeline associated with the development of transgenic plants harboring stably integrated genes, which often spans several months, agroinfiltration expedites large-scale screening processes within a mere few weeks. This rapidity represents a significant advantage, particularly in research contexts where timely results are imperative.



Moreover, agroinfiltration serves as an effective means to screen for plants displaying desirable phenotypes, such as disease resistance. The technique facilitates the straightforward application of pathogen-specific genes, allowing for the swift assessment of disease resistance traits. This expedites the identification of promising plant candidates for further study or crop development. Beyond its research utility, agroinfiltration shines as a platform for the high-capacity production of valuable compounds, including pharmaceutical and other recombinant proteins. This platform's notable advantage lies in its ability to rapidly accumulate substantial quantities of proteins, presenting a compelling option for industries requiring large-scale protein production. Its efficiency in this regard holds the potential to drive advancements in the pharmaceutical and biotechnology sectors.

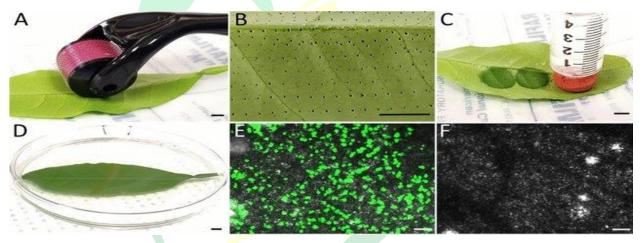


Fig 1: Simplified Citrus Leaf Gene Insertion: Using a Microneedle Roller, Source: Acanda *et al*, 2021

- Step a and b: Creating Surface Wounds with a Microneedle Roller
- Step c: Gentle Pressure Application for Citrus Leaf Agroinfiltration
- Step d: Incubation for Transient Gene Expression in Agroinfiltrated Citrus Leaves
- Step e: Seeing Green: GFP Expression in Citrus Leaf Cells
- Step f: Control Group: Agroinfiltration with Empty Plasmid

Scientific description

Transient expression through agroinfiltration in plants represents a straightforward process characterized by its simplicity. The primary time-consuming phase in this procedure revolves around the cloning of a transgene construct into a vector designed for the temporary delivery of genetic material into plant cells. Typically, the vector finds its home in



Agrobacterium cells, and the act of infiltration can be executed using one of two methods: direct infiltration employing a syringe or the use of a vacuum chamber.

What sets agroinfiltration apart is that it does not entail the integration of any DNA sequences into the plant's genome. Instead, the delivered gene enjoys a temporary and localized expression within the plant cells. This approach can be further categorized into two methods: "sensu stricto," where non-germline tissues, such as leaves, are infected, leading to localized expression; and agro-infection, where similar tissues are exposed to a virus-based vector, thereby enabling the gene to disseminate throughout the entire plant. This versatility in methods allows researchers to tailor their approach to meet specific research objectives, offering a powerful tool for plant genetic studies and biotechnology applications.

References

Acanda, Y., Welker, S., Orbović, V. *et al.* A simple and efficient agroinfiltration method for transient gene expression in *Citrus. Plant Cell Rep* **40**, 1171–1179 (2021).



