

Virus induced gene silencing: Tool to identify host genes affecting viral pathogenicity

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Introduction

Virus induced gene silencing (VIGS) is a mechanism which makes use of a dsRNA to provide defense against viral pathogens. In this method a dsRNA gene segment homologous to the host mRNA is carried by a viral vector which transfers it inside the plant. The viral gene of interest is identified by plants after which plant's defense mechanism induces VIGS to degrade these gene segments. In this way the plants becomes transgenic for the particular gene and is protected from further attack by these viral pathogens. In VIGS, transcript and not the protein is involved in protection of plants. Also, resistance is more at lower levels of transcripts than at higher levels. TMV was the first virus which was used as a vector for VIGS.

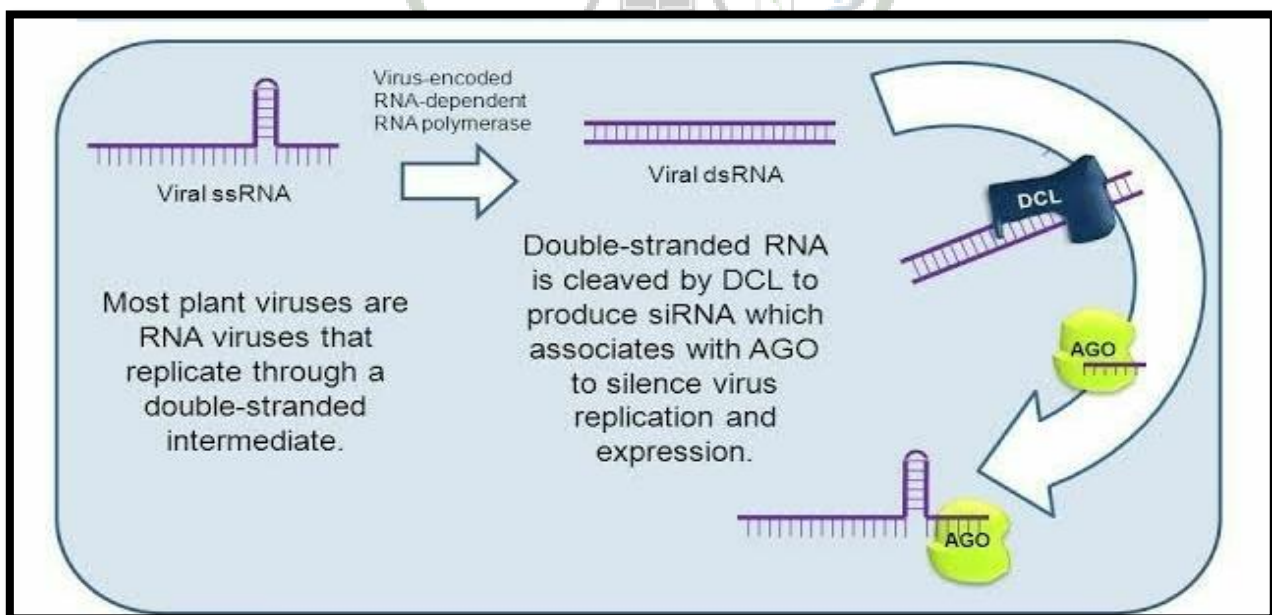


Figure 1:-Mechanism of VIGS

Mechanism of VIGS

There are so many theories regarding the mechanism of VIGS. However the most accepted theory states that VIGS operates via dsRNA gene segment. The transgene having the sense transcript which is a dsRNA produces antisense RNA indirectly. These dsRNA target sequence are produced by either DNA transcription or RNA replication. The presence of enzyme RdRp also supports the above mentioned mechanism. The dsRNA gene segment of the viral vector targets the homologous segment present in host and destroys it. This produces a observable effect on the phenotype of the plant .However, VIGS is not always induced by the transgenes. It is activated only when the transgene crosses a certain "Threshold Level". This character has 100% transferability.

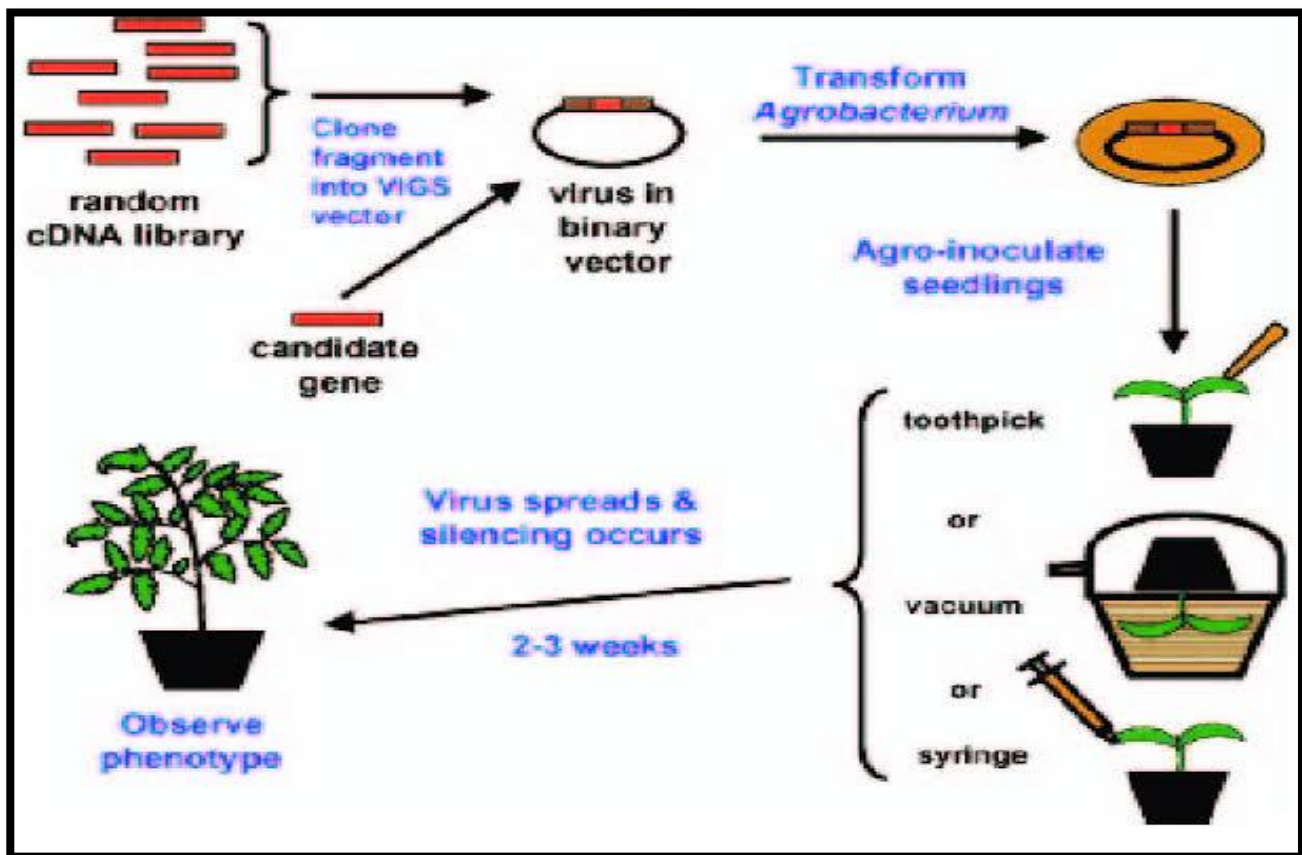


Figure 2:-Steps in VIGS

Image reference:-Burch-Smith TM, Anderson JC, Martin GB, Dinesh –Kumar SP. Applications and advantages of virus-induced gene silencing for gene function studies in plants. Plant J 39:734-746.

Commonly used VIGS Techniques

PVX derived VIGS for potato silencing:-These are agroinfection vectors. An example is Pgr106. This technique involves construction of PVX-Derived vectors followed by transformation and infection by *Agrobacterium tumefaciens*.

TRV derived VIGS for Arabidopsis silencing:-This is also *Agrobacterium* mediated method of introducing and producing infection in plants. It is most commonly used out of all the methods because it is easy to apply on plants.

Arabidopsis One step TYMV derived silencing:-This method involves silencing of inverted repeated sequences of host. It also allows for direct transfer of plasmid DNA into plant.

Limitations of VIGS

- Υ VIGS sometimes completely suppress the phenotype.
- Υ VIGS can also miss phenotype that is masked by functional redundancy between gene family members.
- Υ Uniform silencing sometimes does not occur.
- Υ Inoculation of plant with virus can alter the plant's development.
- Υ Might suppress the non-target genes thus hindering important functions performed by plants.
- Υ Leads to non-stable gene silencing.

Applications of VIGS

- Υ VIGS is also applied in studying important metabolic pathways of plants which is usually difficult to detect by traditional methods.
- Υ VIGS is being used during tissue culture to silence some genes to allow uninterrupted growth of callus.

Y VIGS is also used in studying mechanism of different types of abiotic stress in plants and how to overcome that. Ex:-It was used to study factors involved in stress tolerance in tomato plants.

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