Transgene silencing: New insights into an old puzzle

The use of transgenic organisms has become routine in biology in the last decade. The technology is now being put to practice for creating grains with increased protein content, fruits and vegetables with enhanced nutritional value and flowers with exotic colours. A common means of doing so is to try and overexpress the plant's own proteins. Often however, instead of producing large quantities of proteins, the manipulation leads to exactly the opposite result: it causes suppression of the host gene(s) as well as of copies inserted as transgenes ("co-suppression") and results in a strong reduction in both host and transgene steady-state mRNA levels. This could occur as a result of methylation of DNA sequences, particularly promoter elements, leading to a decrease in transcription. In a number of cases however, the (trans)genes are apparently transcribed at normal rates in the nucleus, indicating that the suppression is post-transcriptional. When the transgenes contain a cDNA derived from the genome of an RNA virus, plants carrying the transgene can be resistant to the virus (Lindbo et al 1993). Although the effect has been documented in several systems, the mechanism by which it occurs remains unclear.

One model invokes a threshold level of RNA accumulation, following which, all homologous RNA in the cytoplasm is degraded specifically (Dougherty and Parks 1995). Another proposes that ectopic pairing of homologous DNA is involved in the initiation of silencing (Baulcombe and English 1996). There is also evidence that co-suppression is not cell autonomous, and can be transmitted between cells—perhaps through the entire plant. Palauqui et al (1997) address questions about the existence of a silencing message and its propagation within the plant. The work involves a series of elegantly designed grafting experiments with tobacco plants that exhibit co-suppression of nitrate reductase (Nia) host genes and transgenes, which results in chlorosis (yellowing of leaves). The authors demonstrate that when a normal non-suppressed scion (upper vegetative tissue) is grafted on to a suppressed stock (lower vegetative tissues and the root system), co-suppression is induced in the scion. Transmission is gene specific and requires the presence of a transcriptionally active non suppressed transgene in the scion. Moreover, it can occur in the absence of the roots of suppressed stocks. Strikingly, the information which triggers the de novo co-suppression is mobile and can be transmitted through as much as 30 cm of a non transgenic interstock segment. The phenomenon has been termed 'systemic acquired silencing' (SAS) by analogy with systemic acquired resistance, a mechanism that offers plants broad resistance to pathogen attack. Palauqui et al (1997) suggest that accumulation of Nia mRNA is required for both spontaneous and graft induced silencing.

A more piece of recent work (Palauqui and Vaucheret 1998) shows that transgenic lines accumulating Nia mRNA above the level of wild-type plants, can undergo graft induced silencing even if they are unable to spontaneously trigger co-suppression. In addition, non-transgenic mutants that over accumulate host Nia mRNA on account of metabolic deregulation, also display graft induced silencing. This implies that whereas the presence of a transgene is necessary for the initiation step, it is dispensable for the RNA degradation step of silencing.

What is the identity of the agent that carries the signal for co-suppression? Increasing evidence favours that this may be at least in part, an RNA molecule derived from the suppressed gene or its transcripts. Likely candidates include bits of transcript produced during RNA degradation, prematurely terminated transcripts, and copy RNA (cRNA) molecules produced from sense transcripts by endogenous RNA dependent RNA polymerases. A ribonucleoprotein (RNP) complex composed of cRNA molecules and plant proteins could be responsible for transmitting the signal into surrounding cells through plasmodesmata, the intercellular channels that connect plant cells (Jorgensen et al 1998). This hypothesis is consistent with the finding that plasmodesmata can facilitate cell to cell trafficking of proteins and their transcripts, thereby regulating plant growth and development (Lucas et al 1995). The identity of the SAS signal however remains unknown. Its characterization could be vital in understanding plant pathogen interactions. More significantly, it could reflect the existence of a complex information network that forms a basis for precise processing and transmission of information, central to plant development and physiology.

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