Nuclear voyage of a plant virus protein

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The nucleolus is a prominent sub-nuclear domain and is the site of transcription and processing of pre-rRNAs and biogenesis of pre-ribosomal particles. In addition, the nucleolus participates in other aspects of cell function such as stress responses and the cell cycle. The nucleolus is structurally and functionally linked to Cajal bodies (CBs) that contain small nuclear and small nucleolar ribonucleoprotein particles (snRNPs and snoRNPs) as well as a range of different proteins, including nucleolar protein fibrillarin. CBs are dynamic structures which can move within the nucleus. The multifunctional nature of the nucleolus and CBs has recently been extended to include RNA silencing; the production of heterochromatic small interfering RNAs involved in transcriptional silencing occurs in CBs or other processing foci in the nucleolus. A number of animal and plant viruses have a nucleolar phase in their life cycles. However, the specific role of the nucleolus and other sub-nuclear bodies in virus infections has remained elusive.

Our recent work has provided the first model describing the molecular mechanism of the involvement of sub-nuclear bodies, CBs and the nucleolus, in biological functions of a plant virus. We have shown that the ability of *Groundnut rosette virus* (GRV) to move long-distances through the phloem strictly depends on its interaction with CBs and the nucleolus. Umbraviruses differ from most other viruses in that they do not encode a capsid protein (CP) such that conventional virus particles are not formed in infected plants. The absence of a CP in umbraviruses, however, is compensated for by the ORF3 protein that facilitates long-distance movement of viral RNA in the form of viral RNP particles through the phloem, the specialised vascular system used by plants for the transport of assimilates and macromolecules. After entering the plant cell, GRV establishes translation and replication of viral RNA (Fig. 1). Once the ORF3 protein has been translated it enters the nucleus and targets Cajal bodies (Fig. 2). Targeting of CBs by the ORF3 protein may utilise elements of existing CB-trafficking pathways, for example, those used by snRNPs trafficking from the cytoplasm to CBs. Interaction of the ORF3 protein with CBs further leads to the re-organisation of CBs into multiple Cajal body-like structures (CBLs), which may involve either fragmentation of CBs into multiple bodies or the redistribution of CB components into new structures containing the ORF3 protein. In CBLs the ORF3 protein interacts with fibrillarin. This interaction prompts further CBLs movement to and fusion with the

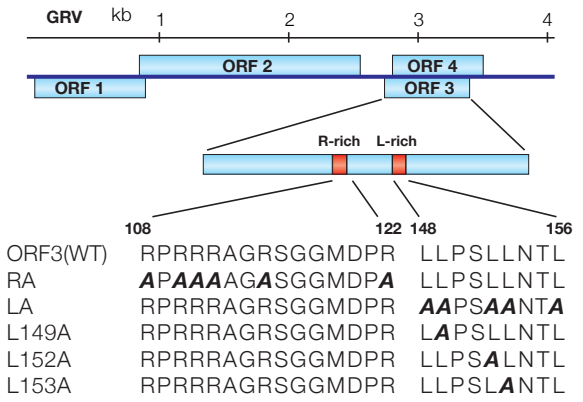

Figure 1 GRV ORF3 protein domains involved in nuclear localisation. Schematic representation of the GRV genome with wild-type and mutant ORF3 protein sequences of the R-rich and L–rich domains.

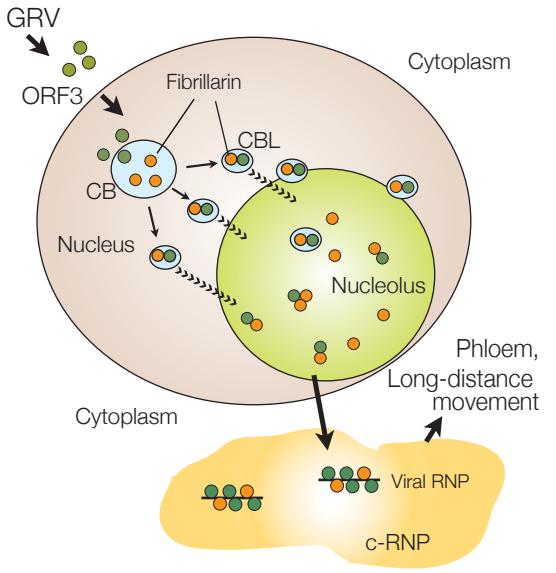

Figure 2 Model of GRV infection and the role of the ORF3 protein, fibrillarin, Cajal bodies and the nucleolus.

nucleolus. The last stage of the nuclear voyage of the ORF3 protein is its nuclear export leading to formation of virus RNP particles in cytoplasmic inclusions. During this stage the ORF3 protein causes the re-localisation of some of the fibrillarin pool to the cytoplasm where viral RNPs containing ORF3 protein, fibrillarin and viral RNA accumulate. Fibrillarin, an RNA-binding protein, needed for RNP formation, may bind the viral RNA or act as a chaperone to permit or catalyze the regular assembly of proteins around viral RNA. When produced in companion cells, the viral RNPs are able to migrate into the phloem sieve elements where they are transported to the rest of the plant to generate a systemic infection.

Thus the interaction of the GRV ORF3 protein with fibrillarin triggers all the consequent molecular and cellular events to establish a systemic infection. How the ORF3 protein re-programmes trafficking pathways and molecular interactions for successful infection will have implications for other plant viruses which interact with the nucleolus and will aid our understanding of interactions between the nucleolus, CBs and their components.