

GENOME ORGANIZATION OF *CHERRY LEAF ROLL VIRUS* AND FUNCTIONIONAL ANALYSES OF VIRUS-ENCODED PROTEINS

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Abstract

Cherry leaf roll virus (CLRV) infects many stone and some small fruits. Recently, the complete genome was determined revealing the genome organization of the bipartite positive stranded plant RNA virus (*Secoviridae* family, genus *Nepovirus*). Now we aim to characterize the functions of CLRV-encoded proteins. As a prerequisite, it is necessary to understand the processing of mature proteins from polyproteins encoded by vRNA1 (P1) and vRNA2 (P2). The viral proteinase and putative cleavage sites will be analyzed *in vitro* in order to determine specific sites utilized during proteolytic processing of P1 and P2.

CLRV is transmitted by pollen and seed. Systemic infection of a host plant is achieved by cell to cell movement via plasmodesmata and long-distance transport through the vascular system. Members of the family *Secoviridae* are transported as virions, thus requiring the coat protein (CP). Further, the viral movement protein (MP) inducing tubular structures by multimerization within plasmodesmata is necessary for passage of virus particles to adjacent cells. Virus-like particles (VLPs) have been observed within tubules in anther cells and pollen grains of CLRV-infected birch and walnut. However, the underlying interactions of CLRV-CP and MP involved in cell to cell movement have never been investigated. We applied the yeast two-hybrid system (YTHS) to address this question. Dimerization of the MP (385 aa, 42 kDa) and the CP (512 aa, 54 kDa) could be shown as well as the specific protein interaction of both viral proteins. Additionally, binding of the viral MP to a plant protein (At-4/1) which is localized at plasmodesmata was demonstrated. At-4/ has been shown to interact with the tubuli-forming MP of *Tomato spotted wilt virus* (TSWV) and facilitates intra- and intercellular trafficking. This suggests that CLRV and TSWV are utilizing the same cell-to-cell transport mechanism in their host plants.

Keywords: CLRV, genome organization, cell-to-cell movement