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SPECIFIC INTERFERENCE THROUGH RNAI OF INSECT GENES

POTENTIALLY INVOLVED IN VECTOR TRANSMISSION OF PLANT VIRUSES

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Transmission by vector organisms is the most usual method of dissemination of plant viruses. Homopteran insects like aphids and whiteflies are vectors of numerous viruses, and therefore their control is considered among the most promising strategies to fight against viral diseases. However, the effectivity of insecticide treatments is frequently not sufficient for an adequate control of non-circulative viruses, which are acquired and inoculated in short periods of time. As an alternative, we propose to interfere with the virus transmission process itself. The interference could be designed considering the mechanisms of transmission, and particularly the specific molecular interactions among the virus and the insect. Using aphid-transmitted potyviruses as experimental model, we are exploring the use of RNAi-based strategies to target specific vector genes that might be essential for transmission. Potyviruses are transmitted in a non-persistent manner with a viral auxiliary factor, the HCPro protein, mediating the reversible retention of virions to the stylet of the vector. Using purified HCPro as bait, new aphid proteins specifically interacting with the potyviral HCPro have been identified. The involvement in virus dissemination of these candidates could be validated functionally using RNAi to knock down their expression in aphids. Two procedures for RNAi are being evaluated: the first one is based on feeding insects in artificial diets supplemented with dsRNAs generated *in vitro*, and the second one takes advantage of unrelated plant virus-based expression vectors containing fragments of the targetted genes to induce accumulation *in planta* of specific siRNAs. Aphids fed on the artificial diets with dsRNAs, or on plant tissues producing siRNAs were subsequently analyzed by quantitative RT-PCR to measure accumulation of the targetted gene transcripts. Parameters like concentration of triggering molecules, duration of treatments, and developmental stage of the aphid at the time of application are being considered to optimize the effect. In addition to provide new tools for understanding the mechanisms of virus transmission, the final goal of this research will be to design novel strategies for virus control based on targetting specifically the expression of insect vector elements essential for transmission.