

RNA interference and artificial micro RNA based strategies for engineering insect resistance against fruit borer (*Helicoverpa armigera*) of tomato

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Biotic stresses, including insect pests cause a colossal loss of crop yield. The traditional approaches to control insect pest have met with certain limitations. Therefore, transgenic strategies, including RNA interference (RNAi) have gained relevance for crop protection against insect pests. Indeed, RNAi-based strategies have proven to be novel and potential alternative for insect pest control, besides many other potential applications for crop improvement. Thus, we have been exploiting RNAi technology for engineering some important crop plants like tomato for resistance against important insect pests, particularly *Helicoverpa armigera*, the notorious pest that attacks more than 50 crop plants. RNAi strategy essentially involves the expression of double-stranded RNA (dsRNA) in transgenic plants targeting some vital genes of insect pests, which are absolutely essential for their growth and development. We have identified chitinase (*CHI*) gene of *H. armigera* as a novel RNAi target as this gene is vital for insect metamorphogenesis. Several RNAi lines of tomato plants expressing dsRNA of *H. armigera CHI* gene were developed by *Agrobacterium*-mediated genetic transformation, and insect bioassays on tomato leaves and fruits of RNAi lines showed enhanced insect resistance. Further, we are also using the most recent and novel artificial miRNA technology to develop tomato resistant to *H. armigera*. The amiRNA technology uses the endogenous precursor miRNAs (pre-miRNAs), where the native miRNA/miRNA* sequences are replaced with the desired amiRNA/amiRNA* sequences. This leads to an efficient and specific silencing of the target gene. For the control of *H. armigera*, ecdysone receptor (*EcR*) gene, which plays a vital role during insect development and metamorphogenesis was targeted by amiRNA. The feeding bioassays with *E. coli* expressing amiRNA-*HaEcR* showed decreased target gene transcripts and also affected the *H. armigera* growth and reproduction when compared to controls. Further, the amiRNA-*HaEcR* construct was prepared and used for tomato transformation by *A. tumefaciens*, and the confirmed amiRNA transgenic tomato lines showed the expression of the intended amiRNA, and these lines have exhibited resistance against *H. armigera*. These studies suggest that RNAi and amiRNA strategies can be effectively used for the control of crop insect pests.