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Tomato cultivar tolerant to Tomato leaf curl New Delhi virus infection induces virus-specific siRNA accumulation and defense associated host gene expression

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Tomato leaf curl New Delhi virus (ToLCNDV) infection causes significant yield loss in tomato. Availability of conventional tolerance source against this virus is limited in tomato. To understand the molecular mechanism of virus tolerance in tomato, the abundance of viral genomic replicative intermediate molecules and virus-directed short interfering viral RNAs (siRNAs) by host plant in a naturally tolerant cultivar H-88-78-1 and a susceptible cultivar Punjab Chhuhara at different time points after agroinfection were studied. We report here that less abundance of viral replicative intermediate in tolerant cultivar may have a co-relation with a relatively higher accumulation of virus-specific siRNAs. To study defense-related host genes expression in response to ToLCNDV infection, suppression subtractive hybridization technique was used. A library was made from tolerant cultivar H-88-78-1 between ToLCNDV-inoculated and *Agrobacterium* mock inoculated plants of this cultivar at 21 day post-inoculation (dpi). A total of 106 non-redundant transcripts were identified and classified into 12 different categories according to their putative functions. By reverse northern analysis and quantitative real time-PCR (qRT-PCR), we identified differential expression pattern of 106 transcripts, out of which 34 transcripts were up-regulated (>2.5 fold induction). Of these, 8 transcripts showed more than four-fold induction. qRT-PCR analysis was carried out to obtain a comparative expression profiling of these 8 transcripts between Punjab Chhuhara and H-88-78-1, upon ToLCNDV infection. The expression patterns of these transcripts showed a significant increase in differential expression in the tolerant cultivar mostly at 14 dpi and 21 dpi in comparison to that in the susceptible cultivar as analyzed by qRT-PCR. Our study reveals that changes in host gene expression that occurred during ToLCNDV interaction were associated with tolerant characteristics of cultivar H-88-78-1. A strong correlation of siRNA accumulation with ToLCNDV tolerance was also observed in cultivar H-88-78-1. The probable direct and indirect relationship of siRNA accumulation and up-regulated transcripts with ToLCNDV tolerance mechanism is discussed.

Keywords: *Tomato leaf curl New Delhi virus* (ToLCNDV), short interfering viral RNAs (siRNA), gene expression, suppression subtractive hybridization (SSH), reverse northern, quantitative real time-PCR (qRT-PCR)

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