APLICAÇÃO TÓPICA DE MOLÉCULAS DE RNA DE FITA DUPLA VISANDO O GENE DO NUCLEOCAPSÍDEO CONFERE PROTEÇÃO CONTRA GROUNDNUT RINGSPOT VIRUS

TOPICAL APPLICATION OF DOUBLE-STRANDED RNA MOLECULES TARGETING THE NUCLEOCAPSID GENE CONFERS PROTECTION AGAINST GROUNDNUT RINGSPOT VIRUS

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Resumo:

Groundnut ringspot virus (GRSV) is a devastating plant pathogen that causes huge crop losses worldwide. In Brazil, the disease is known as 'vira-cabeça do tomateiro', being particularly destructive to tomato plants. The control of this disease is frequently carried out by the use of resistant cultivars. However, due to the emergence of resistance-breaking strains, new disease management strategies are necessary. RNA interference-based approaches represent attractive and promising tools against viral infections in plants. Furthermore, non-transgenic and transgenic approaches by expression of double-stranded RNA (dsRNA) molecules homologous to genomic regions of viruses have been successfully developed for viral management. In this work, dsRNA molecules deriving from the nucleocapsid (N) gene sequence of GRSV were produced *in vitro* and topically applied onto tomato seedlings (cv. BRS Sena) to test for protection against the virus. The dose of 200 µg/plant of the GRSV-dsRNA suspension was applied by gently rubbing one fully expanded true tomato leaf with abrasive. After 24 h, primed plants were challenged by mechanical inoculation of GRSV (t1). Four other treatments were included in this experiment: t2) Plants inoculated with dsRNA only; t3) Plants inoculated with the virus without dsRNA application; t4) mock-inoculated plants (phosphate buffer); and t5) untreated plants. Each treatment consisted of 12 plants and the trial was replicated thrice. GRSV infection was confirmed 14 days post-inoculation (dpi) by indirect-ELISA and symptom evaluation. As expected, plants from t2, t4 and t5 did not show any symptom of virus infection, while those from t3 presented curling of upper leaves and necrotic spots 8-dpi, with infection rates >91%. In contrast, t1 plants showed the first symptoms after 12-dpi, and at lower infection rates, which ranged from 0% to 66%. In general, symptoms were milder in infected plants that were treated with dsRNA (t1) than in those that were untreated (t3), suggesting that some level of dsRNA protection occurred in these plants. The topical application of GRSV-dsRNA significantly reduced infection rate by GRSV and, therefore, this non-transgenic approach is a promising biotechnological tool for plant viral protection.

Palavras-chave: Groundnut ringspot virus; Viral management; dsRNA; Tomato; Biotechnological tool