W6-6

Virus Family: Geminiviridae

Category: Resistance

Title: RNAi-mediated resistance to *Bean golden mosaic virus* in genetically engineered common bean

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Abstract: BGMV is transmitted by the whitefly Bemisia tabaci (Gennadius) in a persistent, circulative manner, and causes the golden mosaic of common bean (Phaseolus vulgaris L.). The characteristic symptoms are yellow-green mosaic of leaves, stunted growth or distorted pods. Control measurements have been primarily focused on controlling the vector by contact or systemic insecticides, with the concomitant problems of development of insecticide resistance, low cost-benefit ratio, and environmental concerns. The disease is the largest constraint to bean production in Latin America and causes significant yield losses (40-100%) in South and Central America, Mexico, and the United States. In Latin America, BGMV causes severe yield losses, particularly during the warmer months when B. tabaci population is higher, leading to a great reduction of summer plantings of common bean. Extensive screening of common bean germplasm for resistance to BGMV has revealed no genotypes with high levels of resistance. The resistance often is unsatisfactory and commercial cultivars are susceptible under early, moderate, or severe infection. Several strategies have been employed for genetically engineering resistance to viruses in transgenic plants. For begomoviruses, most of them have involved the expression of truncated defective genes and antisense RNA. Here, we explored the concept of using RNAi construct to silence the AC1 viral gene and generate highly resistant transgenic common bean plants. AC1 encodes a complex, multifunctional protein (Rep) that acts as a rolling-circle replication initiation factor, which is the only protein strictly essential for viral genome replication and is capable of regulating its own expression. Twenty transgenic common bean lines were obtained using the biolistic process with an intron-hairpin construction to induce post-transcriptional gene silencing against the AC1 gene. Two lines (named 5.1 and ahas 3.2) presented high resistance (approximately 93% of the plants were free of symptoms) upon inoculation at high pressure (about 500 viruliferous whiteflies per plant during the whole plant life cycle) and at a very early stage of plant development. Transgene-specific siRNA were detected in both inoculated and non-inoculated transgenic plants. A semiquantitative PCR analysis revealed the presence of viral DNA in transgenic plants exposed to viruliferous whiteflies for a period of 6 days. However, when insects were removed, no virus DNA could be detected after an additional period of 6 days. Geminiviruses cause severe disease problems on several other crops throughout the world, such as cassava, cowpea, mungbean, pepper, melon, tomato, cotton blackgram, lima bean, soybean, potato, eggplant, pepper, chili pepper, watermelon, squash, and papaya and have been considered the "pest of the century". The strategy presented here could be extended to achieve resistance against geminiviruses from other plant species.