

JOINT EVENT

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## The various roles of CsRDR1 family genes in cucumber defense against different viruses

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RNA-dependent RNA polymerases (RDR) play an important role in virus protection and plant gene control. Their activity is based on the synthesis of double stranded RNA that is a template for the gene silencing system. Four *RDR1* (*RDR1a*, *b*, *c1*, *c2*) genes were identified in cucumber having different expression levels before and after infection by various viruses. The *CsRDR1a*, *b*, *c* genes have a homology of 60-58% at the amino acid level, while *CsRDR1c1* and *c2* are almost identical (97% homology), but have different promoter sequences. A high expression level of *CsRDR1b* was characterized in cucumber strains with partial resistance to a number of viruses, while *CsRDR1c1* and *c2* not expressed in healthy plants. Cucumber plants infected by several different viruses showed a moderate expression level of *CsRDR1b* and a dramatic level of *CsRDR1c1*, *c2*. The expression level of *CsRDR1a*, *CsRDR2* and *CsRDR6* did not increase following virus infection. The differential expression of *CsRDR1b* and *CsRDR1c1*, *c2* indicates dissimilar control of these genes. The relationship between the high levels of expression of *CsRDR1b* in resistant cucumber cultivars reinforces the assumption that this gene is unique for virus resistance. Using the *CRISPR/Cas9* system, we created a variety of mutants in the *rdr1b* and *rdr1c1* and *c2* genes. Knockdown of these genes increased virus accumulation. Plants with a homozygous mutation in the *rdr1b* genes (deletion of 34 nucleotides) and *rdr1c* (2 and 1 nucleotides in the *rdr1c1* and *rdr1c2* genes respectively) showed increased susceptibility to *Cucumber mosaic virus* 4 dpi, especially *rdr1c* mutants plants which collapse 6 days after infection. The viral symptoms increased also after *Cucurbit vein yellowing virus* infection, whereas the susceptibility to *Zucchini yellow mosaic virus* infection was less. Notably, *CsRDR1b* expression in healthy *rdr1b* mutants was less than in non-mutant plants. The control of *CsRDR1b* and *CsRDR1c1*, *c2* is under study in our laboratory to help understand the silencing mechanism of plants defense.

### Recent Publications

1. Leibman D, Kravchik M, Wolf D, Haviv S, Weissberg M, Ophir R, Paris H S, Palukaitis P, Ding S W, Gaba V and Gal-On A (2017) Differential expression of cucumber RNA-dependent RNA polymerase 1 genes during antiviral defense and resistance. *Molecular Plant Pathology* 19(2):300-312.
2. Chandrasekaran J, Brumin M, Wolf D, Leibman D, Klap C, Pearlsman M, Sherman A, Arazi T and Gal-On A (2016) Development of broad virus resistance in non-transgenic cucumber using CRISPR/Cas9 technology. *Molecular Plant Pathology* 17(7):1140-53.
3. Leibman D, Wolf D, Saharan V, Zelcer A, Arazi T, Shibolet M Y, Gaba V, and Gal-On A (2011) A high level of transgenic viral small RNA is associated with broad *potyvirus* resistance in cucurbits. *Molecular Plant-Microbe Interactions* 24(10):1220-1238.

### Biography

Diana Leibman completed her PhD in 2012 from the Hebrew University in Jerusalem. She works as a Research Engineer in the Agricultural Research Organization, The Volcani Center. The research topics in which she is involved are: The role of RNA-dependent RNA polymerase 1 in plant defense against viruses; CRISPR/Cas9 genome editing technology for crop improvement; development of resistance to RNA and DNA viruses in cucurbits and tomato by transgenic approaches; identification of plant genes associated with disease symptom development to plant virus infection and; genetic engineering of attenuated ZYMV-AG as a plant virus vector, for gene expression and epitope presentation.

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