

1-25. **The coat protein of Turnip crinkle virus is required a full-length to maintain suppressing activity to RNA silencing but no relation with eliciting resistance by N-terminal region in Arabidopsis.**

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The coat protein (CP) of Turnip crinkle virus (TCV) is organized into 3 distinct domains, R domain (RNA-binding) connected by an arm, S domain and P domain. We have previously shown that the CP of TCV strongly suppresses RNA silencing, and have mapped N-terminal R domain of which is also the elicitor of resistance response in the Arabidopsis ecotype Di-17 carrying the HRT resistance gene. In order to map the region in the TCV CP that is responsible for silencing suppression, a series of CP mutants were constructed, transformed into *Agrobacterium*, coinfiltrated either with HC-Pro (the helper component proteinase of tobacco etch potyvirus) known as a suppressor of PTGS or GFP constructs into leaves of *Nicotiana benthamiana* expressing GFP transgenically. In the presence of HC-Pro, all CP mutants were well protected, accumulating mutant CP mRNAs and their proteins even 5 days post-infiltration (DPI). In the presence of GFP, some mutant constructs which showed the accumulation of CP mutants and GFP mRNAs at early stage but eventually degraded at 5 DPI. Only a mutant which carrying 4 amino acid deletion of R domain was tolerable to maintain suppressing activity, suggesting that the suppressing activity is not directly related with the eliciting activity. A transient assay also revealed that the mutants synthesized their proteins, suggesting that a full length of CP sequences and its intact structure are required to stabilize CP, which suppresses the RNA silencing.

1-26. **Complete nucleotide sequences of an *Rsv*-resistance overcoming isolate of soybean mosaic virus.**

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The complete nucleotide sequences of genomic RNA of an isolate of soybean mosaic virus (SMV-CN18), which has ability to overcome *Rsv* resistance of soybean, have been determined. A large open reading frame encodes a polyprotein of 3068 amino acids with a predicted Mr of 350 kDa. Based on comparison with the proposed cleavage site of other potyviral polyproteins, nine mature proteins are predicted as a following order, P1, HC-Pro, P3, CI, 6K, VPg, NIa, NIb and coat protein (CP). The mature proteins of the strain share various amino acid identity with known SMV-G2, -G7 and -N strain, with the greatest variability occurring in the P1 (91 %, 88 %, 96%) and the lowest variability in the CP (100 %, 99 %, 100 %). In addition, 5' untranslated region determined by 5' RACE is much more various than any coding regions. Difference in amino acid

sequences throughout the genome is discussed in relation to resistance and susceptibility of soybean cultivars to SMV-CN18.

1-27. Silencing of *CaCDPK4* (*Capsicum annuum* Calcium Dependent Protein Kinase) and Its Ortholog, *NbCDPK5* Induces Cell Death in *Nicotiana benthamiana*.

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We have isolated a full-length cDNA clone, *CaCDPK4* encoding a typical calcium-dependent protein kinase (CDPK) from hot pepper cDNA library. Genomic southern blot analysis showed that it belongs to a multigene family, but represents a single copy gene in hot pepper genome. RNA expression pattern of this gene revealed that it is induced by infiltration of *Xanthomonas axonopodis* pv. *glycines* 8ra into hot pepper leaves but not by water deficit stress. However, high salt treatment of NaCl (0.4 M) solution to hot pepper plants strongly induced *CaCDPK4* gene. In addition, this gene is weakly responsive to the exogenous application of salicylic acid or ethephon. Biochemical study of the GST-*CaCDPK4* recombinant protein showed that it autophosphorylates *in vitro* and the presence of EGTA, a calcium chelater, eliminates the kinase activity of the recombinant protein. As a way to identify the *in vivo* function of *CaCDPK4* in plants, VIGS (Virus-Induced Gene Silencing) was employed. *Agrobacterium*-mediated TRV silencing construct containing the kinase and calmodulin domain of *CaCDPK4* resulted in cell death of *Nicotiana benthamiana* plants. A highly homologous *N. benthamiana* CDPK gene, *NbCDPK5*, to *CaCDPK4* was cloned from *N. benthamiana* cDNA library. VIGS of *NbCDPK5* also resulted in cell death. The molecular characterization of this cell death phenotype is being under investigation.

1-28. Cloning And Characterization of Pathogen-Inducible EREBP-Like Transcription Factor (*CaNR19*) From Hot Pepper (*Capsicum annuum* L.)

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An EREBP/AP2-type transcription factor (*CaPF1*) was isolated by DDRT-PCR following inoculation of soybean pustule pathogen *Xanthomonas axonopodis* pv. *glycines* 8ra which induces HR on pepper leaves. Genomic Southern blot analysis revealed that the *CaPF1* gene is present as a single copy within the hot pepper genome. The deduced amino acid sequence of *CaPF1* has two potential nuclear localization signals, a possible acidic activation domain, and an EREBP/AP2 motif that could bind to a conserved *cis*- element present in promoter region of many stress-induced genes. The mRNA level of *CaPF1* was induced by both biotic and abiotic stresses. We observed higher-level transcripts in resistance-induced pepper tissues than diseased tissues. Expression of