

CPSF30 in Medicago

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CPSF30 is a component in 3' RNA processing that has been well characterized in the model species *Arabidopsis thaliana*. In this study the CPSF30 gene has been cloned out of *Medicago truncatula* and *Medicago sativa*. A large region within the gene exhibits significant conservation between *A. thaliana* and both *Medicago* species. This area of the gene was cloned out and 5' and 3' RACE were used to clone each full length gene. The *Medicago* genes were cloned into a protein expression vector. The vector was used to translate each gene into its corresponding protein and each protein was purified and dialyzed. CPSF30 in *A. thaliana* binds calmodulin and it was hypothesized that CPSF30 from both the *Medicago* species would also bind calmodulin based on the conservation of the amino acids between the model species and the *Medicago* species. CPSF30 from *A. thaliana* also has been shown to possess three CCCH type zinc fingers, with the first implicated in RNA binding and the third in RNA cleavage. Amino acid alignments showed the zinc finger regions were conserved and it was hypothesized that CPSF30 from both *Medicago* species would bind RNA and cleave RNA. Electromobility shift assays were used to determine that RNA binding did occur. RNA nuclease activity in *Medicago* is still being tested. CPSF30 knockout studies in *A. thaliana* have yielded some consistent phenotypes, such as a decrease in lateral roots and an elevated resistance to oxidative stress. CPSF30 *M. sativa* knockout/knockdown plants have been recovered, and the transgenic T0 plants exhibit aberrant root phenotypes. Additional analyses are ongoing.