Targeted gene disruption reveals the involvement of a plastid-localized pentatricopeptide repeat protein in processing of plastid pre-mRNA

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A large gene family encoding proteins with pentatricopeptide repeat (PPR) motif consisting of 35 amino acids exists in land plants, from a moss *Physcomitrella patens* (1) to flowering plants, *Arabidopsis thaliana* and *Oryza sativa* (2), but does not in algae or cyanobacteria. Interestingly, most of PPR proteins are expected to localize in plastid or mitochondria, and several of these have been shown to be involved in RNA splicing, RNA cleavage, or RNA editing (3). However, it is mostly unknown how PPR proteins are involved in such different RNA processing events. We searched EST and genomic sequences deposited in public databases, and found 91 PPR protein genes in *P. patens*. Fifteen predicted proteins were expected to be plastid-localized, of which four were experimentally demonstrated to be plastid proteins. To investigate the function of plastid PPR proteins, we have generated and characterized the plastid-localized PPR protein gene disruptant of *P. patens*. PPR531-11 disruptant mosses displayed abnormal phenotypic characters such as the growth of protonema colonies, the number and shape of chloroplasts, and so on. To further investigate whether disruption of PPR531-11 gene impairs accumulation or processing of specific plastid RNAs, we performed northern blot analysis using specific probes of intron-containing plastid genes. This analysis revealed that PPR531-11 functions in the intergenic processing of plastid RNA between *clpP* and 5'-*rps12* and in the splicing of pre-*clpP* mRNA. These results indicate that PPR531-11 is important for plastid mRNA maturation and plastid biogenesis.

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