

Abstract

Cytoplasmic male sterility (CMS) carries immense significance in realizing hybrid vigor in order to enhance the productivity of a crop species. Rearrangement in the mitochondrial genome is generally attributed as the causal mechanism in conferring CMS trait in plants. The wild abortive (WA) rice cytoplasm has been extensively utilized in rice hybrid seed production all over tropical rice growing countries. However, no information is available to explain what makes the WA to be a powerful CMS system.

To understand the mechanism operative in the WA system of CMS rice at the molecular level, the role of mitochondrial *orf155* gene as a possible factor responsible for causing male sterility in rice with WA cytoplasm has been investigated in the present study. Comparison of RFLPs and transcript profile has shown the presence of polymorphism in *orf155* gene among the male-fertile and male-sterile rice lines. Southern analysis has shown that a single copy of a variant form of *orf155* gene is present in the mitochondrial genome of the CMS-WA rice line, whereas a single copy of the wild form of *orf155* is present in both male-fertile maintainer and restorer lines. The northern and RT-PCR analyses has revealed that the variant *orf155* gene in male-sterile line generates two kinds of transcripts- one edited (~0.7 kb) like the wild form and the other unedited (~1.1 kb), whereas the *orf155* gene of maintainer and restorer lines produce only one type of edited transcript (~0.7 kb). Differential transcription profile and RNA editing (demonstrated by RT-PCR analyses) of the *orf155* gene-specific transcripts between WA-sterile and male-fertile rice lines point towards the implication of *orf155* gene as the likely candidate for causing CMS syndrome in the WA system.

CMS in plants is a classical example of genomic conflict, where opposing maternally inherited cytoplasmic genes induce male sterility and nuclear genes counteract the effect by restoring male fertility. Northern blot analyses of various maintained CMS and fertile restored hybrids have revealed that the transcript profile of *orf155* gene remains unaltered by the presence of nuclear *Rf* gene(s). However, RNA editing of the *orf155* gene-specific transcripts is affected by the presence of

dominant nuclear *Rf* gene(s), as revealed by RT-PCR analyses of the coding DNA region of the *orf155* gene in maintained CMS and restored hybrid plants. The larger ~1.1 kb *orf155* transcript, which is unedited in sterile line, undergoes editing like the major ~0.7 kb transcript in the presence of *Rf* gene(s). The mechanism of fertility restoration is sporophytic in case of CMS-WA rice system, as evident from pollen testing of fertile restored hybrids. Additionally, the classification of genes belonging to the *Rf-1* locus in various rice lines has revealed that editing of the transcript of the variant *orf155* gene is dependent upon the function of *Rf-1A* and/or *Rf-1D* genes belonging to the *Rf-1* locus on chromosome 10 of the rice genome.

Keywords: Cytoplasmic male sterility, WA (Wild Abortive) cytoplasm, *orf155* gene, Mt-genome, RFLP, RNA editing, Nuclear restorer of fertility (*Rf*) gene.