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Using a candidate gene approach to identify the restorerof-fertility Rf4 gene for maize CMS-C

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Cytoplasmic male sterility (CMS) is a maternally inherited trait that prevents normal pollen development. Although CMS is a mitochondrial mutation, there are nuclear genes known as restorer-of-fertility genes (Rf) that restore normal pollen development. This makes CMS particularly useful in performing controlled crosses for plant breeding programs and seed production, where the desired female carries a CMS mutation and the male contains the nuclear Rf gene. In maize, CMS type C (CMS-C) can be restored to fertility by the Rf4 gene. Identification of Rf4 can provide insight into the cause of CMS-C pollen abortion, and expand our knowledge of nuclear-mitochondrial communication. Our approach for identifying Rf4 combines genetic information with the physical maps and emerging DNA sequence data. Previous studies have genetically mapped the Rf4 gene to the short arm of a chromosome 8 (Sisco, 1991). Our goal is to identify a candidate gene for Rf4 in the correct region of chromosome 8. Genes in this region were categorized based on the probability of the proteins to be targeted to the mitochondria using MitoPROT and Predotar. The genes were also analyzed for PPR motifs using TPRpred. We used the criteria of mitochondrial targeting and a probable PPR functional motif because most Rf genes identified to date are targeted to mitochondria and have a PPR motif in their sequence. Primers have been designed for candidate genes, and they will be amplified and sequenced from six different maize inbred lines: three with the fertility restoring allele Rf4 and the non-restoring allele rf4. The sequenced genes will be then analyzed for nucleotide polymorphisms that correlate with the Rf4 and rf4 alleles.