

## Genome re-sequencing of diverse sweet cherry (*Prunus avium*) individuals reveals a modifier gene mutation conferring pollen-part self-compatibility

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Self-incompatibility (SI) is an important reproductive mechanism to maintain a genetic diversity within a plant species. Concurrently, SI can be a limiting factor for efficient production and breeding of crops. Among the plant taxa with the S-RNase-based SI, *Prunus* in the Rosaceae exhibits *Prunus*-specific S-RNase-based SI system and its recognition mechanism is different from those of the others. It is known that specificity determination genes, *S-RNase* and *SFB*, for *Prunus* SI system are located on the *S* locus of the chromosome 6. However, the whole image of the SI reaction process remains to be clarified. To elucidate the *Prunus*-specific SI recognition mechanism, we focused on a self-compatible (SC) sweet cherry (*Prunus avium*) cultivar, ‘Cristobalina’, which is presumed to have a mutation of a pollen-part modifier gene located on the *M* locus of the chromosome 3. Illumina whole-genome sequencing and subsequent subsequence analyses were performed to identify a causal mutation in ‘Cristobalina’. Two ‘Cristobalina’ F<sub>1</sub> populations, both segregating for SC and SI individuals, as well as diverse SI sweet cherry cultivars were subjected to Illumina genome sequencing. Obtained reads were subdivided into 35-bp subsequences called k-mers. K-mers thus obtained were cataloged into SC and SI pools for the extraction of SC-specific k-mers. Then, the original reads containing the SC-specific k-mers were assembled into candidate contigs containing the *M* locus of ‘Cristobalina’. Next, SC-specificity of the contigs was confirmed thoroughly utilizing Illumina genomic reads from sweet cherry cultivars and ‘Cristobalina’ progenies. The SC-specific contigs were mapped to the sweet cherry whole genome sequence database released public in the last year to identify the candidate gene conferring SC in ‘Cristobalina’. As a result, the SC-specific transposon-like insertion in the *M* locus was identified as the most probable candidate. The insertion appeared to downregulate the expression of a glutathione S-transferase (GST)-like gene in pollen. This GST-like gene was named *MGST* (*M* locus encoded GST). Phylogenetic, evolutionary, and gene expression analyses revealed that *MGST* may have undergone lineage-specific evolution, and the encoded protein may function differently from the corresponding proteins encoded by *GST* orthologs in other species, including members of the subfamily Maloideae (Rosaceae). Thus, *MGST* may be important for *Prunus*-specific SI recognition mechanism. Functional characterization of the *MGST* will lead to further understandings of the *Prunus*-specific SI recognition mechanisms.