

Novel nonrestoring alleles for Owen-type cytoplasmic male sterility in sugar beet (*Beta vulgaris* L.)

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Introduction: Hybrid seed production in sugar beet relies on cytoplasmic male sterility (CMS). CMS is defined as male sterility caused by sterile factor (S) in mitochondria genome without any defects in the female reproduction and vegetative organs. Sterility caused by S is restored by the nuclear gene termed “restorer of fertility” (*Rf*). Owen-type CMS is the most utilized CMS for breeding in sugar beet. Two *Rf* genes have been identified in sugar beet and one of that is called *Rf1*.

CMS line must be maintained by maintainer line possessing normal cytoplasm and nonrestoring *rf* alleles. But *rf1* alleles in maintainer lines in Japan are only two types. It has a risk of genetic vulnerability. The reason why *rf1* alleles are only two types is sugar beet in Japan was introduced from other countries in 19th century and derived from only seven varieties. If there are other nonrestoring alleles in the world, we can find a lot of maintainer alleles from other varieties.

Aim: We attempt to identify novel *rf1* alleles from maintainer lines of United States Department of Agriculture (USDA).

Methods: Genotyping of *Rf1* alleles are conducted with *Rf1* linkage markers and southern blotting with probe designed in 3'UTR of *Rf1* gene. Crossing test conducted with TA-33BB-CMS, including Owen-type mitochondria, as a test parent. Individual of the progenies were carefully evaluated for anther color, dehiscence and pollen production during the flowering period.

Results and Discussion:

We found seven types of *rf1* alleles. Two of them showed restorer of fertility. Other five alleles were thought as maintainer alleles. We determined the DNA sequences of *rf1* gene and its surrounding region of four types which the *rf1* allele was thought as single copy. The sequence of *rf1-L(5L)* was same to that of *rf1-L(4L)* in coding region. Similarly, the sequence of *rf1-S(5S)* was same to that of *rf1-S(3S)* in code region. But it was different from *rf1-L* and homologous between them was 86 percent in nucleotide, so *rf1-S* was novel allele. There were substantially two single copy alleles and one multi copy allele. In conclusion, we found three types of nonrestoring alleles from maintainer lines developed in US and other countries.

Key Words: Cytoplasmic male sterility, genetic resource, plant breeding

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