

Cytology of interspecific hybrid between *Brassica napus* and *B. oleracea*

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Abstract

It is important to widen the germplasm of *Brassica napus* by transferring genetic components from its progenitor *B. oleracea*. Herein, 29 genotypes of *B. oleracea* from four cultivated and seven wild types were used as male parents to cross with one elite *B. napus* cultivar, Zhongshuang 9. A low crossability was detected in all the crosses. Various chromosome pairings were observed in pollen mother cells at diakinesis of F₁ and the configuration with 9II + 10I occurred predominantly, and the fertility of F₂ and F₃ generation recovered fast. Our data suggest a relatively high possibility of euploid in the low generations of interspecific hybridization between *B. napus* and *B. oleracea*. The clues to overcome the low crossability between *B. napus* and *B. oleracea* were discussed.

Introduction

Brassica napus (rapeseed, AACC, 2n = 38), originated from spontaneous hybridization between *B. rapa* L. (AA, 2n = 20) and *B. oleracea* (CC, 2n = 18). Compared its parental species, modern rapeseed has a relatively narrow genetic basis due to the limited geographic distribution and extensive breeding, which limit the potential of genetic improvement of rapeseed (Becker et al. 1995; Seyis et al. 2003). Fortunately, diversity parental species crossing with rapeseed is an important strategy to enlarge its genetic resources. Genetic components of *B. rapa* have been transferred into rapeseed due to high crossability between rapeseed and *B. rapa* and high frequency of euploids gametes in the low generations of interspecific hybridization between *B. napus* and *B. rapa*. The objectives of this study are to investigate the crossability between *B. napus* and variant forms of *B. oleracea*, and to observe cytological behaviors of the progenies deviated from *B. napus* and *B. oleracea*.

Materials and methods

The crossability between an elite rapeseed cultivar, Zhongshuang 9, and 29 genotypes of *B. oleracea*, including four cultivated forms (*B. oleracea* L. var. *acephala*, *B. oleracea* L. var. *alboglabra*, *B. oleracea* L. var. *gemmifera* DC, *B. oleracea* L. var. *gongylodes* L.) and seven wild forms (*B. cretica* Lam., *B. incana* Ten., *B. montana* Pourr., *B. oleracea* L.ssp. *oleracea*, *B. rupestris* Rafin., *B. insularis* Moris. and *B. villosa* Biv.) was evaluated according to the seed set of pollinated pods. And one interspecific hybrid (H-1) derived from the cross between Zhongshuang 9 and K154 (*B. oleracea* L. var. *acephala*), was developed via embryos rescue and tissue culture for cytological analysis. The fitness of pattern of chromosome segregation was calculated with the ratio of actual frequency of chromosome segregation pattern in F₁ to theoretical value according to the methods of Lu et al. (2001).

The F₂ and the rapeseed-like individuals of F₃ with chromosome number 38 were evaluated for reproductive capability such as pollen fertility and seed-set.

Results

Although the pollen grains of *B. oleracea* could grow well on the stigma of *B. napus* ten hours after pollination, a low crossability was found in all of interspecific hybrids between rapeseed and various types of *B. oleracea*. When 1,762 flowers of Zhongshuang 9 were pollinated by 29 *B. oleracea* lines, only 428 seeds were harvested with low seed-set in pollinated pods ranging from 0 to 1.49 seeds per pod.

The young buds of H-1 were collected to characterize its chromosome behaviors. The hybrid had the expected chromosome numbers in its somatic cells (2n = 28, ACC). And the average chromosome associations were 9.06I + 9.12II + 0.10III + 0.08IV + 0.01V. Two pairing patterns, 9II + 10I (60.17%) and 10II + 8I (21.65%) appeared more frequently than the others. A high fitness (16.4) for the pattern of chromosome segregations with 9-19 was detected in the hybrid.

In general, the hybrid exhibited low reproductive capability in pollen fertility and seed-set relative to parental lines. However, the fertility of its progenies recovered fast. Although F₁ hybrids had low pollen

fertility (~10%) and poor seed-set (less than one seed per pod), The F₂ progenies had 55.5% pollen fertility on average, ranging from 10.8% to 95.6%, while the rapeseed-like individuals of F₃ exhibited normal pollen fertility (more than 95%). The average seed-set was 12.26 seeds per pod for F₂, and 24.02 for F₃.

Discussion

B. napus is an allopolyploid species, containing A and C subgenomes with high collinearity. In this study, a low crossability was observed between *B. napus* and both of the cultivated types and wild types of *B. oleracea*. However, relatively high crossability in intraspecific hybridization in *B. oleracea* and in interspecific hybridization between *B. napus* and *B. rapa* was documented. Given that reproductive isolation is controlled by a deleterious interaction between functionally diverged genes from the hybridizing species or evolutionary divergence of genomic positions, and that such evolutionary divergence of genomic positions exhibits neutral effects in their native genetic backgrounds, whereas negative effects occur in the heterozygotes (Ouyang et al. 2010). It seems to provide two clues to overcome the low crossability between *B. napus* and *B. oleracea*. First, the neutral alleles controlling crossability in intraspecific hybridization in *B. oleracea* could be introgressed into *B. napus*. The other clue is to breed *B. oleracea* with the compatible alleles from *B. rapa* or *B. napus* by backcrossing.

Reference

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