

IDENTIFICATION OF SELF-INCOMPATIBLE GENOTYPES IN NATURAL AND
RESYNTHESED RAPESEED LINES (B. NAPUS L.)

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INTRODUCTION

Self-incompatibility (SI) is a major outbreeding mechanism in the flowering plants. In the Brassicaceae SI is under sporophytic control of a single, multiallelic S-locus. The self-incompatibility system is utilized for the production of commercial hybrid varieties in B. oleracea and B. campestris (cabbage, broccoli, cauliflower, chinese cabbage).

Whereas B. oleracea and B. campestris are, with few exceptions, self-incompatible, their natural amphidiploid, B.napus, is self-compatible. Considering the feasible increase of seed yield and oil content, the development of hybrid cultivars of oilseed rape would be a promising approach. In oilseed rape the hybrid seed production on a commercial scale must depend on an efficient outbreeding system.

Two strategies are considered to be successful in establishing a self-incompatibility system in Brassica napus. Self-incompatible B. napus plants have been identified in natural populations of rapeseed (Olsson 1960). The other feasible way is the introgression of S-alleles from its diploid ancestors, B. oleracea or (and) B. campestris through interspecific hybridization (Mackay 1977).

Both strategies are subject of our investigation:

- selection of self-incompatible plants in winter rape populations
- resynthesis of B. napus from B. oleracea and B. campestris lines with defined S-alleles, followed by the introgression of the S-alleles in rapeseed cultivars.

The present paper reports preliminary results on self-incompatibility interactions in natural and resynthesized B. napus and its diploid ancestors.

MATERIALS AND METHODS

Lines of B. oleracea and B. campestris with defined S-alleles were provided by Dr. D. J. Ockendon (Wellesbourne) and Dr. T. Hodgkin (Invergowrie). Interspecific hybrids were obtained at high frequency through in-ovule embryo culture. Technical details on in vitro culture and origin, taxonomy and S-allele status of the genotypes are given in a separate paper (Plümper 1991). Amphihaploid hybrids were subjected to colchicine treatment. Several colchicine treatments were necessary to get diploid plants. Seed set was obtained by bud pollination.

Test pollination between the resynthesized lines and their diploid ancestors should ascertain their incompatibility status. The response of test pollination was studied by fluorescence microscopy. Pistils of open pollinated flowers were removed within 48 hours after pollination and examined with the aid of a UV- microscope according to Naether (1971).

Furthermore, thin layer isoelectric focusing of stigma samples is used to study differences between protein patterns and to identify S-allele specific glycoproteins.

Screening for SI in natural rapeseed populations was based on six winter rape crosses provided by German rape breeders. The material was grown in greenhouse, selfed for several generations and selected for reduced fertility (Beschorner and Odenbach 1989). Fertility was defined as the percentage of well developed seeds per silique in relation to the number of ovules. Fertility calculation based on pods of the main branch.

Test crosses between lines with low self-fertility should reveal information about the occurrence of similar S-alleles. Genotypes expected to carry S-alleles were subjected to haploidisation by anther culture to produce homozygous lines.

RESULTS & DISCUSSION

71 resynthesized B. napus-lines with different S-allele combinations were transferred to soil. To date 42 lines became diploid and set seed.

Results of first test crossings between resynthesized rapeseed and its parental lines are shown in table 1. First results indicate that S-alleles from both diploid ancestors are active in the amphidiploid hybrids. The combination of a self-incompatible B. oleracea-line with a compatible B. campestris-line (B. camp. 6) resulted in an amphidiploid,

which exhibited no incompatibility reaction with the B. campestris parent. Test-pollinations with natural rapeseed were done in respect to fertility control.

Introgression of the S-alleles into double-low winter rape cultivars by means of subsequent backcrossing is in progress.

Table 1. Results of test-pollinations between resynthesized B.napus and parental lines

Resynthetic line	pollinated with	Reaction
B.camp.5 x B.ol.8	B.camp.5 x B.ol.8	SI
	B.camp.5	SI
	B.ol.8	SI
	B.camp.5 x B.ol.9	SI
	Ceres	comp.
	Cobra	comp.
B.camp.5 x B.ol.9	B.camp.5 x B.ol.9	SI
	B.camp.5	SI
	B.ol.9	SI
B.ol.9 x B.camp.6	B.ol.9 x B.camp.6	SI
	B.camp.6	comp.
	B.ol.9	SI

Figure 1 illustrates the results of five test crossings between selected lines of natural rapeseed with low self-fertility, originating from a previous selection (Beschorner and Odenbach 1989). Whereas self-fertility of female and male plants was low, the fertility of the crossing branch increased. Because of the sporophytic nature of the Brassica SI-system, F_1 -hybrids between SI genotypes are expected to be self-sterile.

Results of crossing A provide evidence for the existence of S-alleles in the selected genotypes. The high levels of self-fertility of the other F_1 -hybrids (B-E) indicate, that low fertility in their parental lines is not necessarily related to SI. The occurrence of recessive S-alleles or fertility restoring elements is another possible explanation for these results. Recessive S-alleles can enable fertility restoration of the hybrid cultivar without the help of three-way crosses (Thompson 1978). Progenies of dihaploid plants derived from the selected lines are now under investigation to clarify the exact cause of low fertility.

Due to the restricted gene pool of *B. napus*, the number of theoretically available S-alleles in natural populations of rapeseed must be considered low.

Commercial hybrid rapeseed production will depend on a collection of strong S-alleles to ensure a high outcrossing rate under field conditions. Therefore the introgression of S-alleles from both diploid ancestors is the most promising strategy for hybrid rapeseed breeding based on self-incompatibility.

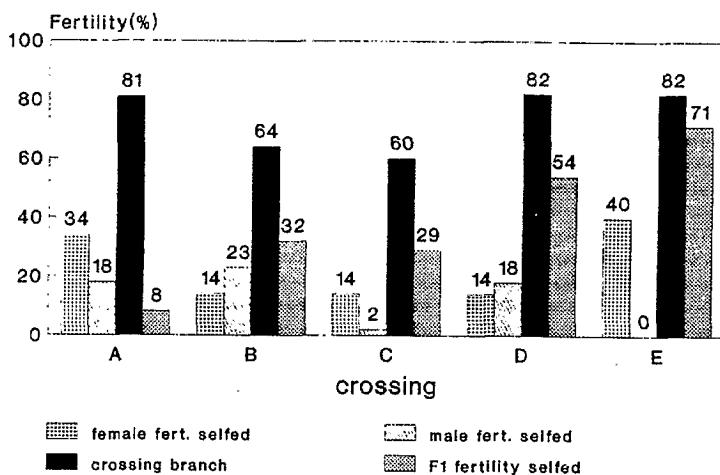


Fig 1. Results of test crossings between lines with low fertility

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