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STUDIES ON INTER-SPECIFIC HYBRIDIZATION IN OILSEED CROP *BRASSICAS*

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ABSTRACT

Twelve intra- and inter-specific crosses using *Brassica juncea* and *Brassica napus* as parents were made to study the potentials of inter-specific hybridization in the improvement *B juncea*. The results suggested that the F1 cross compatibility of inter-specific crosses was similar to that of intra-specific crosses. Getting adequate number of plants in the F2 of inter-specific crosses for higher productivity would not be a limitation. The individual F2 selections were ranked on the basis of stepwise multiple regression index using seed yield as dependent variable. Suggestions were made to go for inter-specific hybridization in the improvement of *B juncea*.

Key Words: Brassica; Cross compatibility; Inter-specific hybridization; multiple regression index.

INTRODUCTION

Unlike cereal crops, the increase in productivity in the oilseed *Brassicas* through breeding has not been striking. One reason is that the improvement of *Brassica* has mostly been confined to the exploitation of the naturally occurring genetic variability in the cultivated species (Rai, 1989). *Brassica juncea* commonly cultivated in the Indian sub-continent has limited variability left for direct selection for higher yield. Genetic variability being limited, breeders need to resort to wide hybridization which can be a viable method for incorporation of desirable characters including yield. Improvement of *Brassica* through inter-specific hybridization by introgression of genes which are otherwise not available has been suggested by many workers (Namai *et al*, 1980; Roy, 1980a; Bajaj et al, 1986).

Little work has been done on the improvement of oilseed *Brassica* through inter-specific crosses. The main limitation has been inter-specific cross incompatibility and F1 plant sterility. As reported elsewhere (Roy, 1977 and 1980b) it is now possible to overcome the sterility barriers both in the F1 and subsequent generations by crossing specially selected compatible genotypes.

In the present paper we discuss some of the problems and potential prospects of inter-specific hybridization using *B. juncea* x *B. napus* inter-specific crosses.

MATERIALS AND METHODS

A number of strains/varieties from two species were chosen as parents for the crossing programme and crosses were made in a 'line x tester' mating design as below.

Varieties	Code	Species
Lines		
YN 3	YN	*B. juncea*
DIRA 313	DR	-do-
Pusa Barani	PR	-do-
Testers		
BDSM 7	BD	B. juncea
NC 57347	JN	-do-
BO 54	BO	B. napus
ISN 706	BN	-do-

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The F1 seeds obtained from crosses made during winter 1989-90 were used for advancing the generation in an off-season nursery (May to September, 1990) at Ranichauri, Hill campus of G.B. Pant University of Agriculture and Technology. F1 crosses were grown along with their parents in three replications. Each entry was sown in a single row of 3 m length and the spacing was 45 cm x 10 cm. At maturity seed from each F1 plant was harvested separately. For each cross there were about 90 plants over three replications.

Pollen sterility study

The flower buds collected from both field grown (off-season nursery) and pot grown plants were studied for pollen sterility/fertility using acetocarmine as stain under compound microscope. The number of fertile and sterile pollens were counted using five random microscopic fields.

The F2 generation of 12 crosses was raised on a plant to row basis at IARI (Indian Agricultural Research Institute)l New Delhi during winter, 1990-91. Parents were also grown alongside their F2. Apparently productive plants were selected and labelled at pod development stage. The F2 populations of inter-specific crosses were small consequently, the sample size was also small. While, the populations of intra-specific crosses were large enough to select adequately large number of plants. A stepwise multiple regression was employed following Draper and Smith (1966) to construct an index for ranking the F2 plants. Seed yield was used as the dependent variable and the rest as independent variables.

The expected value based on the regression equation was calculated for each F2 plant. The F2 plants were ranked in the ascending order based on those expected values. The distribution of F2 plants was divided into four equal portions and the frequency of F2 plants occurring in the top 25% (T1); 26 to 50% (T2); 51 to 75% (T3) and 76 to 100% (T4) were obtained in each set.

Productive recombinants were identified for seed yield and three other important traits namely, number of primary branches (PB), number of secondary branches (SB) and harvest index (HI). *'productive recombinants'* were defined as those which showed positive deviation from the mean of the F2 plants, taken as floor value. Productive segregants for one and more than one character were identified in each cross and in each stratum of ranked F2 distribution (RFD).

RESULTS AND DISCUSSION

F1 cross compatibility

Twelve intra-(*jn* x *jn*) and inter-specific (*jn* x *np*) crosses using *B. juncea* as female were made in this study. Number of pollinations made and number of pods set in each cross were noted (Table 1).

Table 1. Percentage success in hybridization (100x number of pods set/number of pollinations made)

Tester / Line (*jn*)	*jn*			*np*		
	BD	JN	Mean	BO	BN	Mean
YN	51.7	83.1	67.4	100	96.6	98.3
DR	91.0	77.4	84.2	93.4	86.2	89.8
PR	58.0	97.7	77.8	68.4	92.7	80.5
Mean	66.9	86.1		87.3	91.8	

jn = B juncea; np = B napus.

Success in making inter-specific crosses (*jn* x *np*) was as high as in intra-specific (*jn* x *jn*) crosses. Roy (1980b) observed 73% seed set in a *B. juncea* x *B.Napus* cross. He also noted that there was marked difference in percent seed set (5.1%) in the reciprocal cross. Wahiduzzaman (1987) also reported that *B. Juncea* x *B. napus* crosses gave good hybrid seed set. We obtained as high as 89.5% success by using *B. napus* parents as male. It was slightly higher than the percent success when *B.juncea* was used as male (*jn* x *jn*). This could be attributed to the low success of hybridization with BDSM 7 (BD) as male.

F1 sterility and F2 population size

It is apparent that (Table 2) the F2 population size in each of the *B.juncea* x *B. napus* inter-specific crosses was considerably small compared to intra-specific crosses (*jn* x *jn*). This was because the

Table 2. F1 sterility and F2 population size in intra- and inter-specific crosses

Cross	PS	nF2	Cross*	nF2
YNBO	77.3	132	YNBD	1509
YNBN	91.8	132	YNJN	1840
DRBO	89.3	84	DRBD	1554
DRBN	78.0	135	DRJN	1246
PRBO	81.8	135	PRBD	1774
PRBN	68.8	114	PRJN	945

PS : Mean pollen sterility (%);
nF2 : F2 population size;
* : No pollen sterility

fertility in inter-specific cross F1's was very low. Roy (1980b) too observed low fertility (2 to 12 seeds per plant) in some *B. juncea* x *B. napus* crosses and moderate (20 to 30 seeds/plant) in some others. Consequently, the size of the F2 population of such crosses was proportionately small. Further, relatively high sterility did not affect F2 population size drastically. For example, the cross YNBN with 92% sterility has given 132 F2 plants which was slightly higher than (114 plants) PRBN with 69% sterility. Thus, it would seem possible to obtain reasonably large F2 populations even with relatively more sterile inter-specific crosses.

Roy (1980b) reported that in the F2 of a *B. juncea* x *B. napus* cross, plants with very high yield (41 to 58 g) were absent, but in the F3 there were 4% plants with very high yield. Similarly, there were 15% plants with high yield (8 to 40 g) in F2 and 64% in F3.

Taking the above results into proper perspective, it may not be an exaggeration to observe that so long as selecting a super productive recombinant in F2 is not the only goal, one can obtain adequate F2 plants in inter-specific crosses in general, for breeding for higher productivity.

Inter-specific hybridization

Intra-specific hybridization has been the essential pre-requisite and usually attempted by breeders to recombine desirable genes (attributes) from divergent parents of a species into a commercial variety.

Inter-specific hybridization on the other hand, is one of the most promising and at the same time frustrating tools available to the plant breeder (Stephens, 1961). Recombination in the progeny of inter-specific hybrids can generate staggering wealth of variability, but usually a majority of the segregants can be ill adapted and considerably inferior to the parental forms. These ill effects would be apparent whether they result from reshuffling of whole chromosomes or as a consequence of apparently normal crossing over. The advantage of increased crossing over may be more than off set by the disruption of genetic architecture of the species (Harland, 1936), where two differently organized complexes are reduced to random recombinations of their component parts.

Roy (1980b) observed that the F2 populations of inter-specific crosses were highly variable and gave transgressive segregation for fertility and plant type. In some crosses he recovered fertile or partially fertile F2 progeny. In one cross low fertility got improved by inter-crossing or by backcrossing to the compatible parent. In our studies we sought to inquire whether seed yield in inter-specific F2 is comparable to that in intra-specific F2 and whether such F2 could be used as the base material for selection for higher seed yield.

The mean F2 seed yield in the top two strata (T1 and T2) of inter-specific crosses (*jn* x *np*) was as high as that in intra-specific crosses (Table 3). In the other strata, the mean seed yield of inter-specific crosses was relatively low. Nevertheless, it was comparable with that of intra-specific crosses. Roy (1980b) reported that there were 0% plants in F2 with very high yield (41 to 58 g); 15% with high yield (8 to 40 g); 44% with moderate yield (2 to 7 g); 41% with low yield (0.1 to 1.0 g). In this classification, we found nearly 25% with very high yield (Top stratum, T1); 50% with high yield (T2 and T3) and 25% with moderate yield (T4). The results pointed to the possibility for initiating breeding programmes to improve yield using inter-specific F2 particularly of *B.juncea* x *B. napus.*

Table 3. Mean seed yield (g) of intra- and inter-specific crosses in various strata of RFD

Strata	*jn* x *jn* (1)	*jn* x *np* (2)	se (1-2)
T 1	49.7 a	46.2 a	3.56
T 2	33.7 b	28.4 b	2.81
T 3	25.3	15.5	2.4
T 4	17.9	7.3	0.92
se (T 1- T 2)	0.87	4.44	
se (T 2-T3)	0.45	3.66	
se (T 3-T 4)	0.4	2.54	

Means with identical letters are not significant.

Roy (1984) selected partially fertile and intermediate plant types in F2 progenies from an inter-specific cross between *B.juncea* (black leg resistant) and *B. napus* (susceptible) grown in a disease nursery. In a preliminary yield trial conducted under normal disease situation, 96 single plant selections showed high yield potential at F7. Five selections gave 30-50% higher yield than a check. A number of selections were not only high yielding but also much earlier to mature compared to the check variety. In this context, it would be useful to examine whether response to selection for yield would be accompanied by a correlated response in traits like primary branches (PB), secondary branches (SB) and harvest index (HI) and whether selection responses from intra - and inter-specific crosses were comparable. Answers to these points were attempted by comparing the frequency of productive F2 segregants for seed yield (SY) and other traits.

In order to elicit meaningful information, the data on percentage productive recombinants were pooled over respective males of intra- and inter-specific crosses. It is apparent that (Table 4) the percentage productive recombinants was higher in intra-specific crosses particularly when four characters are considered together, while it was relatively high in inter-specific crosses when less number of characters were considered. However, the percentage productive recombinants in inter-specific crosses (*jn* x *np*) was in general, comparable to that in intra-specific crosses. Of the six inter-specific crosses (*jn* x *np*) only two (DRBN; PRBN) had productive recombinants for one and more than one character. While, the cross YNBO showed productive recombinants for two and more than two characters but not for one character.

Table 4. Percentage of productive recombinants (PR) in the F2 of intra- and inter-specific crosses

Female	Number of characters	*np*			*jn*		
(*jn*)		BO	BN	BO + BN	BD	JN	BD + JN
YN	4	16.7	0	16.7	9.2	15.3	24.5
	3	25.0	16.7	41.7	19.9	14.8	34.7
	2	41.7	50	91.7	20.7	25.7	46.4
	1	0	16.7	16.7	28.3	24.9	53.2
DR	4	0	16.7	16.7	15.7	17.2	32.9
	3	16.7	16.7	33.4	11.9	14.7	26.6
	2	50	16.7	66.7	20.9	25.1	46
	1	33.3	16.7	50	16.1	22.1	38.2
PR	4	0	7.7	7.7	15.2	15.1	30.3
	3	0	15.4	15.4	16.2	17.3	33.5
	2	25	7.7	32.7	30.5	26.6	57.1
	1	25	23.1	48.1	19	23	42

Suggestions

* Hybridization efforts should be intense to get adequate F1 seeds. Our studies do show that sterility is not a serious problem to tackle and that mere increase in the frequency of emasculation - pollination would overcome the limitations in seed set.

* Depending on the F1 heterogeneity (which inturn, is a function of parental heterozygosity), F2 should be obtained by intermating. Such a process will sustain usable genetic variability, produce desirable linkages and restore fertility.

* What is still more important is to continue to advance the material to F3 and further generations by mass intermating without conscious elimination of low genotypes (Dutta *et al*, 1986). In every generation, selection for desirable recombinants should be made particularly in the top stratum, where it has been shown that F3 families derived from F2 plants of T1 (top stratum) were found to provide higher frequency of selections for yield (Bandyopadhyay *et al*, 1985). Intermating has been advocated for promoting recombination with favourable potential physiological complementation (Wallace *et al*, 1972).

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