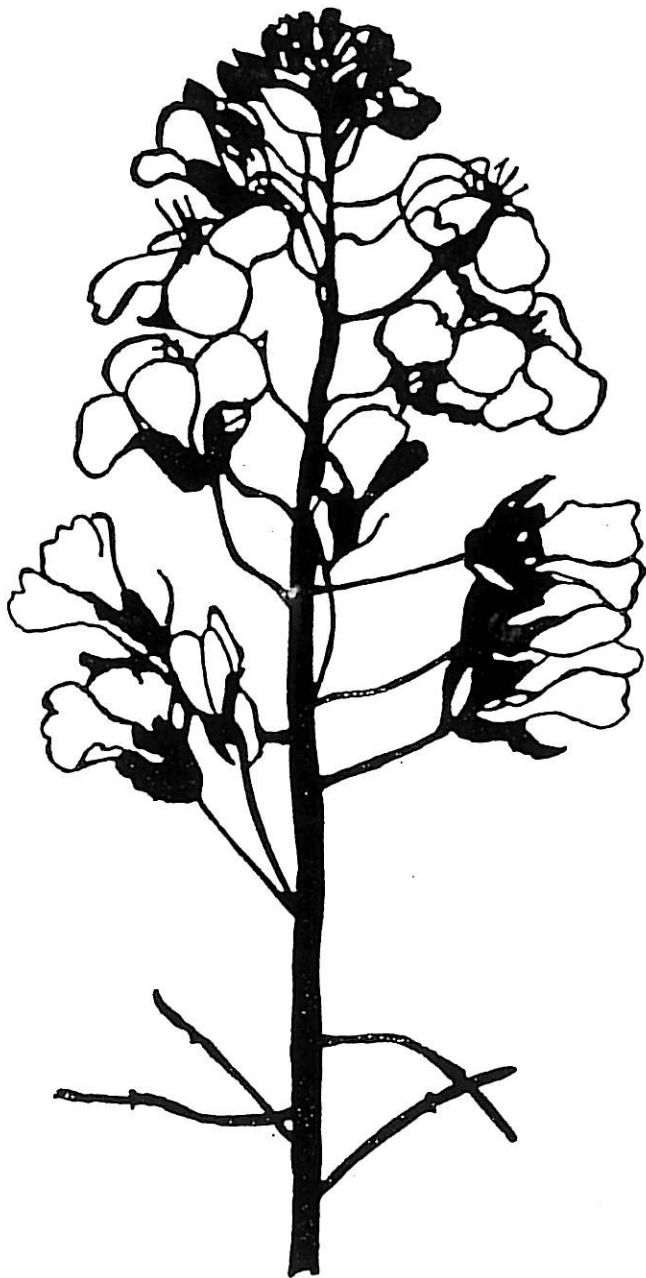


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Preliminary studies on the origin of the Chinese mustard (*Brassica juncea*)

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Introduction

Mustard, *Brassica juncea* (AABB, $2n=36$) is an amphidiploid species between *B. rapa* (AA, $2n=20$) and *B. nigra* (BB, $2n=16$). This species provides important oilseed and vegetable crops in China. The Chinese mustard (*B. juncea*) crops can be classified into five groups according to the usage types (Chen *et al.* 1997). Like *B. rapa*, Chinese mustard is a very old crop species exhibiting enormous morpho-diversity and occupying wide distribution throughout China. This fact together with the report on the occurrence of *B. nigra* in northwestern China leads to some Chinese scholars to put forward the hypothesis that the Chinese mustard species likely had independently originated in China. However, the occurrence of *B. nigra* in northwestern part of China (*i.e.* the Xinjiang Autonomous Region) was not confirmed in our previous studies (Chen *et al.* 1995). The Xinjiang wild *Brassicaceae* materials we had access to were revealed to be wild mustard (*Sinapis arvensis*, SS, $2n=18$) rather than *B. nigra*. Nevertheless, Wu *et al.* (1996) speculated that the Chinese mustard might have originated in China, from the spontaneous interspecific hybridization between *B. rapa* and the Xinjiang wild mustard followed by the loss of one pair of chromosomes of the wild mustard. If the speculation by Wu *et al.* (1996) held true, the Chinese mustard would be more similar with the Xinjiang wild mustard than with *B. nigra*. The current study has been undertaken to address this issue by performing RAPD marker analysis.

Materials and methods

The materials of this study comprised the following species: the Chinese mustard (*B. juncea*), black mustard (*B. nigra*), the Xinjiang wild mustard (*S. arvensis*), yellow mustard (*S. alba*) and *B. rapa*. For the Chinese mustard and *B. rapa*, both oilseed and vegetable accessions were included. For the other each species, only one accession was available. The methods for DNA isolation, RAPD assay and data analysis for calculating genetic distances were according to An *et al.* (1999).

Results and discussion

Both oilseed and vegetable accessions were included for the Chinese mustard and *B. rapa* species in this study. Regardless of the usage types, the different accessions of the same species were clustered together based on the genetic distances. Thus, the RAPD marker analysis revealed that the genetic differentiation within species was smaller than that among the species.

In Table 1 is shown the genetic distances among the Chinese mustard (*B. juncea*), black mustard (*B. nigra*), the Xinjiang wild mustard (*S. arvensis*), yellow mustard (*S. alba*) and *B. rapa*.

Table 1. The genetic distances among the Chinese mustard, black mustard, the Xinjiang wild mustard, yellow mustard and *B. rapa*

	1	2	3	4
Chinese mustard (1)				
Black mustard (2)	0.483			
Wild mustard (3)	0.702	0.575		
Yellow mustard (4)	0.636	0.514	0.540	
<i>B. rapa</i> (5)	0.743	0.698	0.743	0.636

The Chinese mustard has the smallest genetic distance with black mustard, followed by yellow mustard, the Xinjiang wild mustard and *B. rapa*. In this study, the black mustard accession is from Europe whereas the wild mustard is from the northwestern part of China (the Xinjiang Autonomous Region). In spite of this, the Chinese mustard was more similar with black mustard than with the Xinjiang wild mustard. Therefore, the present results do not support the hypothesis that the Chinese mustard had formed between *B. rapa* and the Xinjiang wild mustard inside of China (Wu *et al.* 1996). It is not possible to conclude China as an independent origin centre for the Chinese mustard unless distribution of *B. nigra* is at least confirmed in northwestern or other parts of China. From Table 1, it can also be seen that the three monogenomic mustard species (*B. nigra*, *S. alba* and *S. arvensis*) are more similar with one another than any one of them with *B. rapa*, implying that these three mustard species probably share the same evolutionary lineage.

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APPLICATION OF NEAR INFRARED REFLECTANCE SPECTROSCOPY TO ANALYSIS OF ACID DETERGENT FIBER IN *BRASSICA* SPECIES

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INTRODUCTION

Fiber is of considerable interest because it is not readily digested and can devalue seed and meal as a component of animal feeds. Reduction of the fiber content would improve the digestible and metabolizable energy values and improve the digestibility or availability of protein and amino acids. The analytical methods for obtaining the fiber content in plant material are tedious because of its slowness. Near infrared reflectance spectroscopy (NIRS) has been shown to be a reliable technique to determine oil, protein and moisture concentrations of grains and oilseeds, and is widely used for analysing the chemical composition of forages (Shenk et al., 1979; Marten et al., 1985). In this study we assess the capacity of NIRS to predict acid detergent fiber (ADF) content in the amphidiploids Brassica species *B. napus*, *B. carinata* and *B. juncea*. To conduct this study a multispecies calibration equation was developed based on the three species mentioned previously.

MATERIAL AND METHOD

To perform NIRS analysis, 104 intact seed samples belong to the three species described above were scanned in a NIRS monochromator (NIR Systems mod. 6500, NIRSystems, Inc., Silversprings, MD, USA), and analysed for ADF content (AOAC method 973.18). These samples were selected taking into account their variability for chemical composition and geographical origin.

The wet chemistry values and the absorbance values for each sample were used to develop the calibration equation for ADF using the ISI program CALIBRATE. Four mathematical treatments (0,0,1,1 (derivative, gap, first smooth, second smooth); 1,4,4,1; 1,10,10,1 and 2,5,5,1) were tested on the calibration set. The standard error of calibration (SEC), coefficient of determination (RSQ), standard deviation to standard error of cross validation-ratio (SD/SECV) and 1-VR (1 minus the ratio of unexplained variance to total variance) statistics were used to characterise the different equations obtained and to determine the degree of robustness to predict ADF content on the species studied.

RESULTS AND DISCUSSION

Table 1 shows the ADF content for the set used to conduct this study. The *B. juncea* (n= 49), *B. carinata* (n= 36) and *B. napus* (n= 19) ADF contents ranged from 6.98 to 15.19; 5.33 to 15.47 and 10.05 to 13.45 %, respectively. The concentrations found by us for *B. juncea* are similar to those given by other authors (Newkirk *et al.*, 1997) for this species, who reported values of 12.79% ADF in low glucosinolate meal. The same author gave concentrations of 20.6% ADF for *B. napus*, which is higher than those presented by us in this work. *B. carinata* ranged from 5.33 to 15.47 %, which is the widest range of all. The lack of published studies about the ADF content of this species does not let us to have a reference of the values found by other authors.

Table 1. ADF statistics of the accessions used in the calibration equation (% DM basis)

	n	range	mean	SD
B. juncea	49	6.98-15.19	10.64	2.05
B. carinata	36	5.33-15.47	10.26	2.31
B. napus	19	10.05-13.45	11.81	0.79
total	104	5.33-15.47	10.76	2.07

Table 2 shows the calibration and cross validation statistics. The squared coefficient of multiple determination (RSQ) of ADF was 0.90 (Fig 1), being this value lower than that obtained by Marten, who reported a RSQ of 0.98 for several species of small grain forages (these results were based on ground meal) (Marten et al., 1983). The standard error of calibration (SEC) for Brassica species was 0.67 %, near to that obtained by the same author in his study. These differences are probably due to use intact seeds for NIRS analysis, which introduces additional noise in the spectra. Although Winch (Winch et al., 1981) and Jones (Jones et al., 1987) emphasised the importance of fine grinding for these kind of analysis, it is more interesting for us the use of whole seed due to the preservation of the seed and because it takes a substantial reduction in the NIRS analysis time. The results obtained by us showed that NIRS can be used to predict ADF content in Brassica species with enough accuracy for breeding purposes.

Table 2. Calibration and cross validation statistics for ADF (% DM basis).

N	104
SEC	0.67
RSQ	0.90
SECV	0.96
SD/SECV	2.15
1-VR	0.78

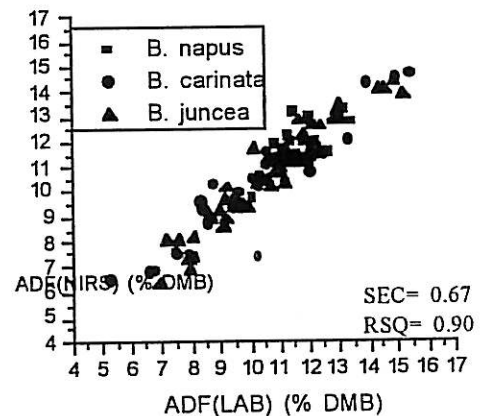


Fig. 1. Calibration plot for ADF.

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occurrence of Phenolic Compounds in *Erucastrum* and *Brassica* (*Brassicinae*)

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Introduction

Within the *Cruciferae*, the subtribe *Brassicinae* includes the closely related *Erucastrum* Presl, and *Brassica* L., two genera whose taxonomic relationships, based on morphology, suggest that they constitute an evolutionary trend when the taxa of *Brassica* represent the more evolved features. About 20 species constitute the genus *Erucastrum*, all of them are wild taxa mainly distributed in the western Mediterranean region, on the contrary, genus *Brassica* include near to 40 taxa, most of which are wild species but also include some important crop (e.g. *B. oleracea* and its varieties).

The phenolic compounds are secondary metabolites widely distributed in plant kingdom and appears to be a useful tool to the analysis of relationships among plant species at several levels. The present work involved comparative analysis of flavonoids of 15 wild taxa, that are listed in Table 1.

Material and Methods

Plant material investigated was obtained from seeds that had been collected from their natural habitats and stored under long-term preservation conditions (Gómez-Campo, 1990) at the germplasm bank of the Departamento de Biología Vegetal, Escuela Técnica Superior de Ingenieros Agrónomos de Madrid (Table 1). In all cases, the leaves of each sample were collected from ten adult plants, at the flowering stage, which had been cultivated in the greenhouse over the three successive spring-summer periods.

Botanical nomenclature is based on Greuter *et al.* (1986).

Table 1. Plant material

<i>E. littoreum</i> subsp. <i>littoreum</i>
<i>E. littoreum</i> subsp. <i>glabrum</i>
<i>E. littoreum</i> subsp. <i>brachycarpum</i>
<i>E. elatum</i>
<i>E. varium</i>
<i>E. virgatum</i>
<i>E. strigosum</i>
<i>B. oxyrrhina</i>
<i>B. tournefortii</i>
<i>B. barrelieri</i>
<i>B. maurorum</i>
<i>B. spinescens</i>
<i>B. fruticulosa</i> subsp. <i>fruticulosa</i>
<i>B. fruticulosa</i> subsp. <i>mauritanica</i>
<i>B. fruticulosa</i> subsp. <i>cossoniana</i>

Flavonoid isolation and identification

Leaves, previously dried in a drying chamber, were ground to powder. Phenolic compounds were extracted following Sánchez-Yélamo (1994). 200 µl of extracts were applied and chromatographed in a two-dimensional paper chromatography (2D PC) on Whatman 3MM sheets developed in BuOH-HOAc-H₂O (6:1:2) and 2% HOAc, to compare flavonoid glycosidic patterns. Twenty replicates were made in each case.

Individual compounds were isolated from the combined paper chromatograms, eluted with hot 80 % ethyl alcohol (EtOH) and the concentrated extracts subjected to acid hydrolysis with 2N HCl for controlled periods. Enzymatic hydrolysis were carried out using β-glucosidase (E.C. 3.2.1.21). In each case, flavonoid aglycones were removed from the resulting sugar-flavonoid mixtures by extraction with EtOAc and were identified using standard procedures by one-dimensional paper chromatography (1D PC) and thin layer chromatography (TLC) in comparison with authentic markers (Markham, 1982, Harborne, 1988, 1989). The identification of sugars was carried out by TLC on pretreated Silica gel plates following Hansen (1975). Individual glycosides were purified by running two consecutive descending 1D PC, first with 15 % Acetic acid (HOAc) and then with 50% HOAc; and identified by UV spectrography and by several chromatographic techniques (PC, TLC and HPTLC).

Results and Discussion

A total of twenty-three flavonoids were isolated from the foliar extracts of the taxa surveyed. The compounds were glycosides derivatives of kaempferol, quercetin and/or isorhamnetin. Table 2 indicate the identified flavonoids with their chromatographic characteristics and UV spectral data.

Each taxon shows a characteristic chromatographic pattern. All samples of *Brassica* showed the presence of kaempferol, quercetin and isorhamnetin mono-, di-, and tri-glycosides except for *B. spinescens* where only di- and triglycosides were detected. The absence of isorhamnetin glycosides appears to be a peculiar character of *Erucastrum* taxa (Table 2), and could be indicative of a generic limit at chemical level.

Table 2. Flavonoids identified in studied taxa

Spot No	Flavonoid	Rf x 100				Colour UV/+NH ₃	Taxa	
		BAW (1)	OHAc (1)	EFAW (2)	WEAA (3)		<i>Erucastrum</i>	<i>Brassica</i>
1	K 7-Gal	64	4	-	-	Y/Y	++	+++
2	Q 7-Gal	47	3	-	-	Y/Y	++	+++
3	K 7-Glc	45	3	-	-	Y/Y	+++++	++++
4	Q 7-Glc	26	2	-	-	Y/Y	+++++	++++
5	Q 3-Glc	44	20	52	28	D/Y	+++++	+++
6	K 3-Glc	53	31	65	17	D/Y	+	++
7	I 3-Glc	47	34	68	28	D/Y	-	++
8	I 3-Digal	35	54	37	61	D/Y	-	+++++
9	Q 3-Digal	32	68	21	71	D/Y	++++	+++++
10	K Trigly	15	75	90	97	D/Y	++	++++
11	K 3-Digal	32	67	36	63	D/Y	+	+++++
12	K 3-Gal-7-Rha	53	49	65	-	D/Y	+++++	++
13	I 3-Gal-7-Rha	53	50	65	-	D/Y	-	+
14	I Trigly (Rha+Gal)	33	77	22	82	D/Y	-	+++++
15	K Trigly (Rha+Gal)	25	77	24	91	D/Y	+++++	++++
16	Q Trigly (Rha+Gal)	21	70	16	77	D/Y	++	++++
17	K 7-Gal-3-Digal	27	60	-	-	D/Y	++	++
18	Q 3-Glc-7-Rha	40	55	63	37	D/Y	+++++	+
19	Q 3-Diglc	34	25	51	54	D/Y	++++	+
20	K (3,7)(Rha+Gal)	43	45	46	57	D/Y	++	++
21	Q (3,7)(Rha+Gal)	41	45	40	45	D/Y	+++	++
22	K Trigly (Rha+Glc)	37	67	-	83	D/Y	+++	+
23	Q Trigly (Rha+Glc)	31	63	-	81	D/Y	++	+

K= kaempferol; Q= quercetin; I= isorhamnetin; Glc= glucose; Gal= galactose; Rha= rhamnose; -gly: glycoside; BAW= n-butanol: Acetic acid: Water (6:1:2); OHAc= 2% Acetic acid; EFAW= Ethyl acetate: Formic acid: Acetic acid: Water (100:11:11:27); WEAA= Water: Ethanol: Acetyl acetone (4:2:1). (1)= PC; (2)= TLC; (3)= HPTLC; D= dark; Y= yellow (+) represent n^o of taxa into each genus with presence of compound.

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MEIOTIC STUDIES ON INTERGENERIC HYBRIDS BETWEEN *BRASSICA NAPUS* AND *ORYCHOPHRAGMUS VIOLACEUS*

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Wild crucifers have proven valuable gene sources for improving *Brassica* crops. The species *Orychophragmus violaceus* (genome OO, $2n=24$) is valued as an ornamental plant in China for its purple flowers. The species also possesses agronomically valuable traits such seed oil with a high percent of palmitic and linoleic acid (14.3% and 53.2%, respectively), low linolenic acid (4.7%), and zero erucic acid (0.9%), disease resistance, and high seed yield (Luo *et al.* 1994). Using embryo rescue, Luo *et al.* (1994) produced hybrids in a cross between *B. napus* (AACC, $2n=38$) Canadian cv. Oro (♀) and *O. violaceus* (♂). We have produced progeny in this cross without embryo rescue and characterized the plants using cytogenetic and molecular techniques. The present paper reports on the meiotic behaviour of 19 progeny plants.

Materials and methods

Seed of *B. napus* cv. Oro and *O. violaceus* was provided by the AAFC-Saskatoon Research Centre, Canada, and by the Huazhong Agricultural University, China, respectively. Plants were raised in a growth chamber programmed for 18°C/15°C day/night (18 h day). Flowers were emasculated prior to pollination and pollinated pistils were bagged until seed had set. Seed was harvested at maturity. Meiotic preparations were made according to Cheng *et al.* (1994).

Results and discussion

Fifty-three seeds were obtained out of 298 pollinations. Twenty-two plants were raised from the 53 seeds and classified into three types on the basis of morphology. Type I comprised five plants with pubescent leaves and yellow flowers, traits that are characteristic of *O. violaceus* and *B. napus*, respectively. Type II comprised three plants showing traits not observed in the parents. One of the plants had shrunken and waxy leaves and possessed axillary flowers, that is flowers that developed in the axil of the normal flowers. The petals of the remaining plants had a pronounced central tissue ridge. Type III included the remaining 14 plants, which were morphologically similar to *B. napus*.

Meiotic pairing was studied in the pollen mother cells (PMCs) of 19 plants. Hybridization between *B. napus* and *O. violaceus* was expected to produce trigenomic hybrids with $2n=31$ chromosomes (10A + 9C + 12O). Type I plants. All plants had $2n=29$ chromosomes (Table 1). Three plants had up to three chromosome fragments. The majority of the PMCs (62.1%) had the pairing configuration 1 III + 9 II + 8 I, while the remaining PMCs had 10 II + 9 I. All bivalents appeared to be homomorphic, i.e. they consisted of chromosomes similar in size and centromeric position. At anaphase I (AI), the chromosomes of each bivalent separated and moved to opposite poles, while univalents lagged behind. The meiotic studies suggested that 12 chromosomes had been eliminated in these plants and that the genome now consisted of 10 duplicated chromosomes and nine unduplicated chromosomes. RAPD analysis (to be detailed elsewhere) revealed that the plants had all of the bands specific to the *B. napus* parent, but only 11.7% of the bands specific to the *O. violaceus* parent. It is therefore possible that most of the eliminated chromosomes belonged to the *O. violaceus*

genome.

Type II plants. The plant producing axillary flowers had $2n=35$ chromosomes that paired to form $17 \text{ II} + 1 \text{ I}$ in most of the PMCs (Table 1). This plant also contained chromosome fragments. The two plants with the petal tissue ridge had $2n=36$ and $2n=37$ chromosomes. The $2n=37$ chromosome plant contained chromosome fragments. None of the three plants possessed *O. violaceus*-specific RAPD bands.

Type III plants. Plants of this group included (Table 1): one plant with $2n=19$ chromosomes; one plant with $2n=37$ chromosomes; seven plants with $2n=38$ chromosomes (19 II) and one plant with $2n=38$ chromosomes ($17 \text{ II} + 4 \text{ I}$); and one plant with $2n=39$ chromosomes. The majority of the plants had one to three chromosome fragments. None of the plants possessed *O. violaceus*-specific RAPD bands. The absence of RAPD bands and morphological traits specific to *O. violaceus* suggested that the plants mainly contained *B. napus* chromosomes.

The present studies suggested that spontaneous chromosome elimination and duplication occurred in the intergeneric hybrids. The plant with $2n=19$ chromosomes could be a haploid of *B. napus*, the result of elimination of the chromosomes of *O. violaceus* following hybridization. Plants with $2n=36, 37, 38,$ and 39 chromosomes could be of hybrid origin followed by chromosome elimination and duplication. In interspecific hybrids of *Hordeum lechleri* x *H. vulgare*, Linde-Laursen and von Bothmer (1998) provided evidence that chromosome elimination and duplication were affected by the position of the chromosomes in the genome. The fact that Type I plants contained the same chromosome number and lacked 88.3% of the *O. violaceus*-specific RAPD bands suggested preferential elimination of certain chromosomes of *O. violaceus* in the intergeneric hybrids of *B. napus* x *O. violaceus*.

Table 1. Meiotic pairing of progeny derived from the cross *B. napus* (♀) x *O. violaceus* (♂).

Progeny	Number of plants	Chromosome number	Meiotic configuration	Number of PMCs
Type I	5	29	$10 \text{ II} + 9 \text{ I}$	28
			$1 \text{ III} + 9 \text{ II} + 8 \text{ I}$	59
Type II	1	35	$17 \text{ II} + 1 \text{ I}, 16 \text{ II} + 3 \text{ I}$	22, 2
	1	36	$17 \text{ II} + 2 \text{ I}$	23
	1	37	$18 \text{ II} + 1 \text{ I}$	17
Type III	1	19	not determined	
	1	37	$18 \text{ II} + 1 \text{ I}$	18
	7	38	19 II	37
	1	38	$17 \text{ II} + 4 \text{ I}$	8
	1	39	$18 \text{ II} + 3 \text{ I}$	11

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Interspecific hybridization between *Brassica campestris* and *B. spinescens* and the cytogenetic analysis of their progenies

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Wild species are often variable source of important genes in crop improvement. In many intergeneric and interspecific hybridizations, the high bivalent chromosome pairings of the F₁ hybrids appeared in first meiotic association (Harberd and McArthur, 1980; Inomata 1997). The possibility of gene transfer from wild relatives to crop brassicas had been reported (Inomata, 1992, 1993, 1994a, 1996; Vyas *et al.*, 1995). Present paper deals with the production of interspecific hybrid between *Brassica campestris* and *Brassica spinescens*. *B. spinescens* possesses some desirable characteristics (Takahata and Tsunoda, 1981; Gupta *et al.*, 1995). The production of the F₁ hybrid, and the cytogenetics and crossability of the progenies were examined.

The materials used in the experiment were *Brassica campestris* subsp. *chinensis* cv. Seppaku-taina ($2n=20$) and *Brassica spinescens* ($2n=16$). When emasculated flowers of *B. campestris* bloomed, the conventional cross with *B. spinescens* was made. The *B. campestris* used in the backcross was the same as the use in the production of the F₁ hybrids. Ovary culture was carried out according to the previous paper (Inomata, 1990). Somatic chromosomes and chromosome associations in the PMCs were examined by using the method of Inomata (1994b).

Sixty-nine ovaries were cultured in 4 days after pollination. Three seeds were obtained and one hybrid grew up. Other one hybrid was obtained by leaving the pollinated flowers as a control. The medium used in the experiment was Nitsch and Nitsch (1969) with White's amino acids and 300 mg/l of casein hydrolysate. All hybrids showed 18 somatic chromosomes, which were dihaploid. The leaves of the F₁ hybrids were intermediate in morphology between the parents. Table 1 showed the results for the pollen fertility and chromosome associations in the first meiotic division of the hybrids. The mode in first meiotic division was 18₁ and the maximum number of bivalent formation was four. Table 2 showed the result for different types of division in microspore mother cells of the F₁ hybrids. The mode was tetrad, however, many microspores showed dyad. Seventy-five flowers in the F₁ hybrids were backcrossed with *B. campestris* but no seeds were obtained. In the present experiment, partial homology between A genome and genome of *B. spinescens* were existed, but the direct gene transfer might be difficult from *B. spinescens* to *B. campestris*.

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Table 1. Pollen fertility and chromosome associations in the first meiotic division of the F₁ hybrids of *Brassica campestris* x *B. spinescens*

Plant number	Chromosome number in root tip	Pollen fertility (%) ^a	Number of PMCs observed	Mean chromosome associations per cell at metaphase I (range in parenthesis)		
				III	II	I
1	18	0	50	0.02 (0-1)	0.8 (0-2)	16.1 (13-18)
2	18	0	51	0.02 (0-1)	1 (0-4)	15.9 (7-18)
Total or range		0	101	0.02 (0-1)	0.9 (0-4)	16 (7-18)

^a: 500 pollen grains were counted.

Table 2. Different types of division in microspore mother cells of the F₁ hybrids in *Brassica campestris* x *B. spinescens*

Plant number	Different types of division in microspore mother cell				
	Dyad	Triad	Tetrad	Pentad	Total
1	61	36	437	5	539
2	143	5	358	0	506
Total	204	41	795	5	1045

A CREATED SPECIES : *BRASSICA CAUDATUS*

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Brassica caudatus is an amphidiploid, derived out of two species *B. tournefortii* and *R. caudatus* (Kumar, 1995 & 1995) . It bears non-dehiscent siliqua.

The plant is bushy with pedicillate, green and pubescent leaves. Petals are white in colour. Pods are bicarpellary with long caudate beak. Seeds are round in shape and blackish brown in colour with 1000 seed weight as 8.5gm. Number of seeds per pod is 2 to 8, though only few pods bear seed.

It is crossed normally and reciprocally with *B. napus*. It showed incompatible F_1 's after crossing with *B. napus* and in their reciprocal cross it expressed male-sterility. But male-sterile plant developed fertile plant after crossing with *B. caudatus*. This indicates that plant appears as a source of male-sterility as well as their restoration.

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Studies on Chromosome behavior in Intergeneric Hybrids of Some Cruciferous Plants

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Abstract

Cytogenetical studies were made on chromosome behavior in intergeneric hybrids of some cruciferous plants. In leaf cells of intergeneric hybrids (F_1) of *Brassica alboglabra* × *Orychophragmus violaceus*, morphological differences were observed between chromosomes from *B. alboglabra* and those from *O. violaceus*. At prometaphase 9 chromosomes from the genome of *B. alboglabra* were lightly stained, each with only a small portion darkly stained around the centromere. In intergeneric hybrids of *Brassica campestris* × *Raphanus sativus* var. *raphanistroides*, meiotic studies showed that in some cells 19 univalent chromosomes were observed at metaphase I, and in a few cells the genome separation was observed at early anaphase I, where 10 univalent chromosomes from the genome of *B. campestris* moved to one pole, and 9 univalent chromosomes from the genome of *R. sativus* var. *raphanistroides* moved to the other.

Key words: chromosome behavior, intergeneric hybrid, cruciferous plant

Since Li et al. (1993) found genome separation in intergeneric hybrids of *B. napus* × *O. violaceus*, many investigators observed genome separation and similar chromosome behavior in intergeneric hybrids between *Brassica* species and *O. violaceus*. (Li et al., 1998; Wu et al., 1996). In recent years the authors firstly observed similar chromosome behavior in intergeneric hybrids of *B. alboglabra* × *O. violaceus* and of *B. campestris* × *R. sativus* var. *raphanistroides*. The experiment results are as follows.

Materials and methods

The following crucifers were used in the experiment, i. e. mustard (*Brassica alboglabra* cv Jielan, $2n=18$), purple-flowered rapeseed (*Orychophragmus violaceus*, $2n=24$), Chinese mustard cv chuanyu 8 (*Brassica campestris*, $2n=20$) and Chinese oil radish (*Raphanus sativus* var. *raphanistroides*, $2n=18$). In order to obtain intergeneric hybrids hand pollination and embryo rescue were used in the experiment. The basic medium for embryo culture was according to Murashige and Skoog (1962). In the preparation of chromosome slides Carnoy's fluid (ethyl alcohol 3 and acetic acid 1) was used for fixation and carbol fuchsin solution for staining.

Results

1. The obtaining of intergeneric hybrids of *B. alboglabra* × *O. violaceus* has been reported. (Yin et al., 1998). In this paper, the authors firstly observed morphological differences between chromosomes of the two parental genomes of the intergeneric hybrids. At prometaphase of the mitotic division of the hybrid leaf cells, 9 chromosomes (apparently from the genome of *B. alboglabra*) were lightly stained, each with only a small portion darkly stained around the centromere, while 12 chromosomes (apparently from the genome of *O. violaceus*) were darkly stained.
2. In our experiment the intergeneric hybrids of *B. campestris* × *R. sativus* var. *raphanistroides* were intermediate between the two parents, with some matroclinous characters, such as the green color and the oval form of their leaves etc., and with some patroclinous ones, such as white flowers and

short siliques etc. The chromosome number of most cells at metaphase I was 19 (10+9, univalents). The hybrid plants (F₁) were partially sterile. Therefore, the above hybrids were true intergeneric hybrids of *B.campestris* × *R.sativus* var. *raphanistroides*.

During meiosis, 19 univalent chromosomes were observed at metaphase I in many cells. At anaphase I we observed that in a few cells 10 chromosomes (apparently from the genome of *B. campestris*) moved to one pole, and 9 chromosomes (apparently from the genome of *R. sativus* var. *raphanistroides*) moved to the other, i.e. genome separation occurred in a few cells,

Discussion

1. In our experiment in leaf cells of intergeneric hybrids of *B.alboglabra* × *O.violaceus*, morphological differences between chromosomes of the two parental genomes were observed. At mitotic prometaphase 9 chromosomes from the genome of *B.alboglabra* were lightly stained, each with only a small portion darkly stained around the centromere, while 12 chromosomes from the genome of *O. violaceus* were darkly stained. It is suggested that the morphological differences between chromosomes of the two parental genomes may be resulted from different condensation of chromosomes and may lead to genome separation.

2. In our experiment of the intergeneric hybrids of *B.campestris* × *R.sativus* var. *raphanistroides*, 19 univalent chromosomes were observed at metaphase I. In a few cells 10 chromosomes from the genome of *B. campestris* moved to one pole, and 9 chromosomes from the genome of *R.sativus* var. *raphanistroides* moved to the other at anaphase I. Thus the genome separation occurred.

3. Genome separation and similar chromosome behavior were found not only in cruciferous plants, but also in some other plants. For example, Thompson (1962) observed genome segregation in *Rubus*. Schwarzacher et al. (1992) observed genome separation in intergeneric hybrids of *Hordeum vulgare* × *H.bulbosum*. Therefore, genome separation and similar chromosome behavior are considerably common, and have some importance in both plant cytogenetical studies and plant breeding.

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The study on the F₁ hybrids between *Brassica chinensis* and *Raphanus sativus* L. var. *raphanistroides* Makino

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Abstract Through embryo rescue, we successfully obtained the hybrid between *B. chinensis* L. and *Raphanus sativus* L. var. *raphanistroides* Makino that has two kinds of flowers; one larger, the other smaller. The larger-flowered has better fertility than the smaller-flowered. The observation on their meiosis showed that the smaller-flowered was chromosome undoubled. At MI, there were 19 unpaired chromosomes, which were assembled in the center of the cells or scattered in the cells. At AI, the unpaired chromosomes divided randomly to two poles. In this process there occurred the "chromosome fractionation", of which very rare were "chromosome set fractionation" of 10-9. The larger-flowered was the chromosome-doubled hybrid. At MI, 19 bivalents were arranged on the equatorial plate. At AI, 19 univalents were divided into two poles of the cell. At AII, there occurred asymmetrical tetrads and polyads. In PMCs of the larger-flowered, it also occurred "chromosome fractionation" and "chromosome set fractionation". The larger-flowered can produce some normal gametes of $n=19$, $n=9$, $n=10$.

Key words: *Brassica chinensis* L., *Raphanus sativus* L. var. *raphanistroides* Makino, Intergeneric hybrid, Chromosome fractionation, Chromosome set fractionation.

Introduction

R. sativus L. var. *raphanistroides* Makino ($2n=18$), belonging to *Cruciferae*, is a valuable oilseed plant. It is characterized not only by resistant to drought, coldness and *Sclerotinia sclerotiorum*, but also by more branches, grand seed and earlier mature (Zhang et al, 1991). It is good oilseed resource. Wide hybridization is a potentially useful method for transferring valuable traits between crops of commercial interest (Roy, 1984, Wu and Luo, 1995). So, intergeneric hybridization between *Brassica* cultivars and *R. sativus* L. var. *raphanistroides* Makino may be useful for the introduction of good traits into rapeseed. Through embryo rescue, we firstly obtained the hybrid between *B. chinensis* and *R. sativus* L. var. *raphanistroides* Makino. This hybrid had a few fertility rates. When selfed, they can produce offspring (F₂). The meiotic analyses of PMCs are reported to explore the fertility of the hybrid between *B. chinensis* L. and *R. sativus* L. var. *raphanistroides* Makino.

Materials and Methods

B. chinensis c.v "Chuan you 8" ($2n=20$) as female parent and *R. sativus* L. var. *raphanistroides* Makino as male parents, the cross was performed in the field by hand emasculation and pollination. We rescued embryos of heart-shaped stages and cultured them in MS medium supplemented with sucrose (3% W/V), 6-benzyl aminopurine (BAP, 2 mg/l), naphthalene acetic acid (NAA, 0.1 mg/l). The pH was adjusted to 5.8. When buds appeared on young embryos. The buds were cut off and transferred to the medium. These buds developed into plantlets, and clusters of buds appeared on the calli formed at the bottom of the buds. By the successive culture of buds, many buds were obtained for application in wide hybridization. After the buds were

cultured on the rooting medium (MS supplemented with sucrose (3% W/V), agar (0.8% W/V), indolebutyric acid (IBA, 0.5 mg/l)) for 15 days, the plants were transplanted into field.

The fresh leaves and young flower buds were used to determine the chromosome numbers of the hybrids. After immersed in ice-water (0°C) for 12 hours, the young leaves were fixed in Carnoy's solution. Before squashed, they were hydrolyzed in 1 mol/l hydrochloric acid 60°C for 12 min. and stained with modified carbol fuchsin solution. The young flower buds were also fixed in Carnoy's solution, transferred into 70% ethanol in 24 hours and then stored at 4°C. The anthers were also stained with modified carbol fuchsin and squashed.

Results and Discussion

On mother plants of *B. chinensis* c.v "Chuan you 8", 26 flowers were pollinated with pollen of *R. sativus* L. var. *raphanistroides* Makino. We rescued young embryos (after pollinated 18 days) on the medium mentioned above, and one hybrid was identified. So, the seed set percentage was 0.1923%.

The chromosome numbers of the hybrids (F₁) were expected number of $2n=19$ (Fig 1, D) and $2n=38$. The regenerated plants had many characters of *R. sativus* L. var. *raphanistroides* Makino. The basal leaves of *B. chinensis* had complete oval leaves, and that of *R. sativus* L. var. *raphanistroides* Makino had lanceolate dehiscent leaves, and the regenerated plants intermediate between them (Fig 1, A, B). Leaf's color of *R. sativus* L. var. *raphanistroides* Makino was grass-green, that of *B. chinensis* and the regenerated plants were green. Silique of *B. chinensis* was longer than that of *R. sativus* L. var. *raphanistroides* Makino and the regenerated plants (They are called radish silique). The flavor of *R. sativus* L. var. *raphanistroides* Makino and the regenerated plants were sarcastic. The flower colors of *R. sativus* L. var. *raphanistroides* Makino and the regenerated plants were white and that of *B. chinensis* was yellow. Flowering season of *R. sativus* L. var. *raphanistroides* Makino and the regenerated plants were delayed. These were all shown that the regenerated plants were the hybrid between *B. chinensis* and *R. sativus* L. var. *raphanistroides* Makino.

The hybrid plants had two kinds of flowers, one larger, the other smaller (Fig 1, C). They were respectively called the larger-flowered and the smaller-flowered. The larger-flowered had good fertility, and the smaller-flowered had a few fertility rates. When selfed, they can produce offspring (F₂). The hybrid offspring (F₂) had polymorphism and at least, there were 4 kinds of plants in the population of the hybrids.

(F₂):(1)matroclinous ones;(2)the hybrids;(3)One with yellow flowers;(4)one with white and yellow flowers. The meiotic observation on the larger-flowered and smaller-flowered were made to study their fertility.

The smaller-flowered was undoubled (2n=19). At MI, 19 unpaired chromosomes exist in PMCs. They were assembled in the center of the cells or scattered in the cells At AI, the unpaired chromosomes were divided randomly to two poles cells (chromosome fractionation), chromosome set fractionation of 10-9 existed in few cells (Fig 1,E and F; Fig 2,A). The gametes of n=10 and n=9 were due to chromosome set fractionation. Those gametes had fertility and the smaller-flowered can produce few offsprings.

The larger-flowered was doubled or partly doubled. At MI, 19 bivalents were arranged on the equatorial plate in most cells (Fig 2,C). At AI, 19 univalents were divided into two poles of the cell (Fig2,D). At AII, there occurred asymmetrical triads, tetrads and polyads besiad ordinary tetrads(Fig 2,E,F,H). Asymmetrical triads (10-10-9)(Fig 2,F) were due to chromosome set fractionation of dyads (19-10) at the second meiosis. Therefore, the gametes of n=19, n=9, n=10 were produced. The larger-flowered had good fertility. Asymmetrical tetrads and polyads were due to chromosome fractionation.

Above all, 3 kinds of functional gametes of n=19, n=10, n=9 were produced by the F₁ hybrid between *B. chinensis* and *R. sativus* L var *raphanistroides* Makino. They can be formed many kinds of 2n=38, 2n=29, 2n=28, 2n=20, 2n=19 and 2n=18. The material of 2n=38 was different from *B. napus* (2n=38). Origin, evolution and relationship among species would be disclosed through observation on the results of hybridization between *B napus* and this material, and a good breeding material for oilseed would be provided. The materials of 2n=29 and 2n=28 were sesquidiplods that can be very useful in chromosome function research and chromosome engineering. The material of 2n=18 was nuclear-cytoplasmic hybrid with nucleus of *R. sativus* L var *raphanistroides* Makino and cytoplasm of *B. chinensis* which would be good material to study the relationship between nuclear and cytoplasm and nuclear-cytoplasmic hybrid vigor. In addition, there were great difference among matured pollen grains (Fig 2,I) and the hybrid offspring (F₂) had many kinds of plants. Those results were also shown that the hybrid between *B. chinensis* and *R. sativus* L. var. *raphanistroides* was due to chromosome set fractionation.

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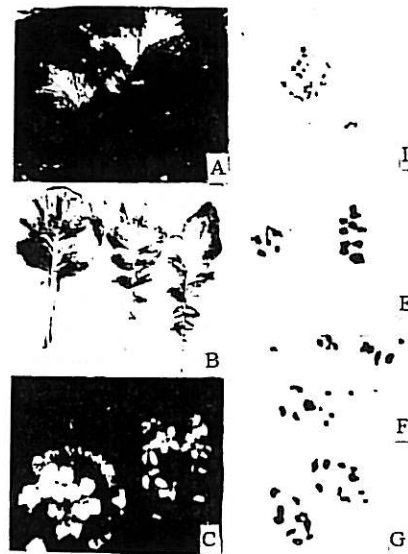


Fig.1

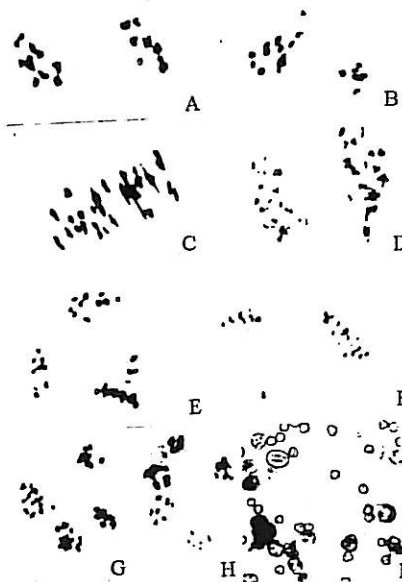


Fig.2

Fig1. A. The regenerated plant of self-fertile hybrid between *B. chinensis* and *R. sativus* L.var.*raphanistroides* Makino. B. The leaves of the hybrid and parents (left: *B. chinensis*; right: *R. sativus* L. var. *raphanistroides* Makino). C. The larger and smaller flower of the hybrids. D. The somatic cell chromosome of the hybrid (2n=19). E-F. The chromosome set fractionation of the smaller-flowered plant of the hybrid (10-9). G. The meiotic behavior of *B. chinensis* at AI. Fig2. A. The chromosome set fractionation of the smaller-flowered plant of the hybrid. B. The chromosome fractionation of the smaller-flowered plant of the hybrid. C-I. Meiotic behavior of the larger-flowered hybrid between *B. chinensis* and *R. sativus* L. var. *raphanistroides* Makino. C. The cell of 2n=38=19II at MI. D.19 univalents moving toward the opposite pole of the cell at AI E-F Asymmetrical triads. G. Asymmetrical tetrads (13-13-6-6) H. Polyad I. The heteropollen grains of the F₁hybrid.

An Efficient Plant Regeneration Protocol from Seedling Explants of *Brassica juncea* RH-781, a Freeze Tolerant Cultivar

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Introduction

Brassica juncea is a major oilseed crop in India. There is a heavy loss of yield because of frost during flowering season. Two of the possible strategies considered to overcome this are transfer of genes responsible for frost tolerance and selection of variant cells or *in vitro* raised plants using low temperature as selection pressure. Both of these require efficient regeneration protocols specific for each genotype. Here we present shoot regeneration protocol direct from seedling explants, which is being used in this laboratory.

Material and Methods

Seeds of *Brassica juncea* RH-781, a var. with lesser susceptibility to frost, were washed under running tap water, surface sterilized by dipping in 0.2% HgCl₂ for 8 minutes and finally washed 3-4 times with sterilized water to remove excess of HgCl₂. These seeds were inoculated on modified MS (Murashige and Skoog, 1962) media (MS +0.8% agar +3% sucrose) and allowed to germinate at 25±1° C under light intensity of 4000 lux and 16 h photoperiod. Cotyledons and hypocotyls from 4-5 days old seedlings were excised and inoculated on different regeneration media listed in the Table 1. Observations on percent shoot formation and number of shoots formed were taken at 10-day intervals.

Results and Discussion

The frequency of shoot formation from cotyledon explants was higher in the media that were tried, as compared to hypocotyl explants. When cotyledons were used as explants, all the media produce shoots and R₄ medium showed highest (nearly 100%) regeneration followed by R₃. Hypocotyl showed much less regeneration capacity. R₂ medium was found to be the best for regeneration from hypocotyls followed by R₄. Statistically there was no significant difference between response of hypocotyls to R₂ and R₄ medium. Therefore, R₄ was concluded to be the best medium as maximum shoot formation was promoted in this combination from both cotyledon and hypocotyl explants.

From the above study it is clear that cotyledons are the best explant for shoot regeneration in *Brassica* in the media listed in Table 1. Similar observations have been made by Yadav *et al* (1991) ; Shrama *et al* (1991) ; Arora *et al* (1996) in contrast to the observations of Lazzeri and Dunwell (1986) and Dash *et al* (1995) who concluded that hypocotyl were the most regenerative explant . This might be due to differences in culture conditions, genotype, age of the explant and different media combinations.

Table 1. Response of seedling explants of *Brassica juncea* var. RH-781 for shoot and root formation in different media

Medium (ppm)	Cotyledons		Hypocotyls	
	%Shoot formation	No of Shoots	%Shoot formation	No of Shoots
R ₁ MS+BAP(1.0)+IAA(0.2)	70 ± 5.2	M	20 ± 4.7	2 – 5
R ₂ MS+BAP(2.0)+IAA(0.2)	65 ± 7.8	2 – 10	50 ± 8.3	2 – 8
R ₃ MS+BAP(1.0)+IAA(1.0)	90 ± 6.2	M	20 ± 5.9	2 – 4
R ₄ MS+BAP(2.0)+IAA(0.5)	97 ± 2.7	M	45 ± 7.9	2 – 5
R ₅ MS+BAP(1.0)+IAA(0.2) +Kn(1.0)	70 ± 5.8	M	20 ± 2.1	1 – 3

M= Multiple shoots; BAP= banzyl amino purine;
IAA= Indole-3-acetic acid

In view of percent shoot regeneration and number of shoots/explant, cotyledons as explant and MS+BAP(2.0)+IAA(0.5) as the media for raising *in vitro* plants are in use for further freeze stress experiments.

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Induction of progressive organogenesis in three varieties of genus *Brassica*

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Introduction

In vitro organogenesis has been reported by several workers in the genus *Brassica* (Zee and Johnson 1984; Narasimhulu and Chopra 1988). Different explants have been used to achieve a organogenic response in *Brassica* through an intermediate callus phase (Bagga *et al.* 1985; Delpierre and Boccon-Gibod 1992). When detached cotyledons are cultured *in vitro* on a medium containing growth regulators, their normal developmental pathway is altered. This altered pathway may lead to callus induction, and subsequently to root, shoot or somatic embryo formation (Murata and Orton 1987; Narasimhulu and Chopra 1988).

In our investigation, we have used cotyledonary explants of three varieties of *Brassica* to induce progressive organogenic responses (rhizogenesis and caulogenesis). These studies to establish different developmental stages in process of organogenesis and embryogenesis somatic, are essential for thereafter to analyse biochemical and molecular changes during those stages (Leshem and Sussex 1990), which permit us to know better the processes of morphogenesis.

Material and Methods

Seeds from three commercial varieties of genus *Brassica* were used for this study: a cultivar of cauliflower [*Brassica oleracea* L. var. *botrytis* L. subvar. *cauliflora* (Gars.) DC., "cauliflower large Christmas"] (CC; 2n=18); and two cultivars of rapeseed [*Brassica napus* L. var. *oleifera* DC.] (AACC; 2n=38), a Canadian cultivar of spring (cv. Oro), and a French cultivar of winter (cv. Jet Neuf).

Cotyledonary explants for callus induction were obtained from 10 days-old seedlings growing *in vitro*. Callus inducing medium (CIM) consisted of MS medium (Murashige and Skoog 1962) supplemented with 2,4-D (2 mg l⁻¹), kinetin (0.1 mg l⁻¹), sucrose (3.5%) as carbon source and Difco Bacto agar (0.8%). After four weeks of culture, the induced calli were used for the 'root inducing treatment' (RIT) or the 'shoot inducing treatment' (SIT). Induction and progressive development of adventitious roots or shoots were obtained from three or two consecutive steps, respectively.

RIT = (1) calli designated as R1 were obtained after two weeks of subculture of induced calli on medium MS + 0.1 mg l⁻¹ 2,4-D + 0.1 mg l⁻¹ kinetin; (2) calli designated as R2 were obtained after another two weeks of subculture on the same medium; and (3) after another four weeks of subculture on the same medium were obtained the R3 calli.

SIT = (1) calli designated as C1 were obtained after two periods of three weeks of subculture of induced calli on medium MS + 4 mg l⁻¹ kinetin; and (2) calli designated as C2 were obtained after three weeks of subculture on MS medium.

The cultures were maintained in a culture room under a 16 h photoperiod, an irradiance of 40 μmol m⁻² s⁻¹, and a temperature of 25 ± 1 °C. The experiments were repeated thrice, and each replicate consisted of 50 callus tissues. The organogenic response was analysed for each callus type previously defined (R1, R2 and R3 for rhizogenesis; C1 and C2 for caulogenesis).

Results and Discussion

The cotyledonary explants of the three cultivars of *Brassica* dedifferentiated and formed calli on CIM. Histological studies have confirmed the progressive development of adventitious roots and shoots. The frequency of rhizogenic calli was increased from R1 to R3 calli in all cultivars (Fig. 1a). This increase was higher in R3 calli of *B. oleracea* and *B. napus* cv. Jet Neuf than in *B. napus* cv. Oro. The caulogenic response was less intense than the rhizogenic. The frequency of caulogenic calli was significantly higher in C2 than C1 calli for

the three cultivars (Fig. 1b). Similarly to rhizogenic treatment, the caulogenesis also was lower in C2 calli of *B. napus* cv. Oro than in *B. oleracea* and *B. napus* cv. Jet Neuf. In general, *B. napus* cv. Oro was least responsive to organogenesis. This marked difference in rhizogenic and caulogenic responses at interspecific and intraspecific levels may be due to the genotypic differences. Ono *et al.* (1994) have also reported a strong influence of genotype on shoot regeneration in different cultivars of *B. napus*.

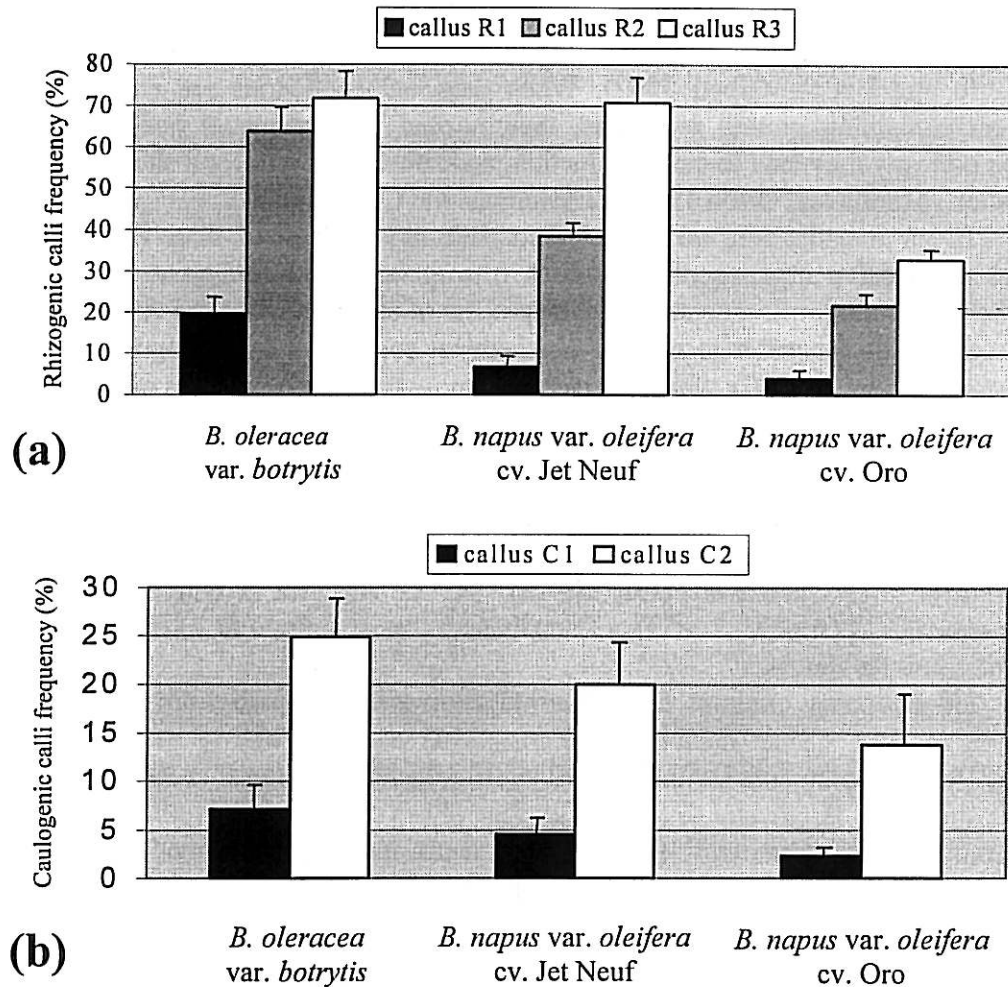


Fig. 1. Organogenic responses from cotyledon-derived calli of three *Brassica* cultivars. (a) Root inducing treatment (R1, R2 and R3 calli); (b) Shoot inducing treatment (C1 and C2 calli).

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ELECTROFUSION OF PROTOPLASTS IN BRASSICACEAE

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INTRODUCTION

The technique of protoplast fusion is used for somatic hybridization between oilseed rape (*Brassica napus*) and other genotypes from the Brassicaceae to modify seed oil compositions in oilseed rape.

In different groups several somatic hybrids have been produced in the family of Brassicaceae, but most *via* chemical fusion. One goal of our work is to establish an electrofusion method with *Brassica napus* in comparison to chemical fusion with polyethylene glycol (PEG). It is a well-known fact that techniques of electrofusion are superior to the fusion with PEG. Here first results are presented.

MATERIAL AND METHODS

Seeds from three lines of *Brassica napus* (WW 1089, Maplus, Drakkar), one line of *Brassica juncea* (CR 98/86), one line of *Barbarea verna* (BAR 3/78), one line of *Camelina sativa* (Volyn'skaja) and two lines of *Crambe hispanica* (CRA 13/81, CRA 15/84) were soaked in 70 % ethanol for two minutes, surface sterilised in a solution of 7.5 % sodium hypochlorite for 30 minutes and rinsed four times with sterile water. Then they were placed on hormone-free Murashige and Skoog medium (MS) with 2 % sucrose (Murashige and Skoog, 1964). Germination and plant growth took place at 22° C in a 16 h photoperiod under 2000-3000 lux with fluorescent and universal white light. Protoplasts were isolated from leaves of four-week-old *in vitro* plants by an enzyme treatment with 0.25 % cellulase and 0.05 % macerozyme overnight at 24° C with slow agitation. After isolation, protoplasts were washed with W5 (Menczel et al., 1981) and diluted to a final density of $4 \times 10^5 \text{ ml}^{-1}$. Both fusion partners were mixed 1:1, centrifuged and resuspended in a fusion solution containing mannitol and sucrose. For somatic hybridization, protoplasts were fused by use of the cell fusion system KRÜSS CFA 500 under different instrumental setups. After fusion protoplasts were cultured in liquid M1 medium (Li and Kohlenbach, 1982) with 0.5 mg l^{-1} NAA, BAP and 2,4-D each for five weeks. Regenerated microcalli were transferred onto different solid media for callus growth and shoot regeneration. These include medium N2 (Sacristan et al., 1989, modified), medium N3 (MS basal medium with 1.0 % sucrose, 2.0 % mannitol, 200 mg l^{-1} casein hydrolysate and 0.5 mg l^{-1} BAP, NAA and thidiazuron each) and MS medium with 2.0 % sucrose, 0.5 mg l^{-1} NAA and 2.0 mg l^{-1} BAP (Khehra and Mathias, 1992). Regenerated shoots were transferred to hormone-free MS medium for rooting. After the preselection of the fusion products with flow cytometry, microsatellites (Kresovich et al., 1995) were used to identify the regenerated plants as hybrids.

RESULTS AND DISCUSSION

In initial experiments the fusion parameters were varied to determine the optimum culture conditions. One problem was the fusion solution. The type of sugar used influences the viability of protoplasts before and during the fusion process. Mannitol or sorbitol alone had a negative effect. Also, fusions were not successful in solutions containing low concentrations of CaCl_2 .

Whereas the combinations of *B. napus* + *C. sativa* and *B. napus* + *B. verna* resulted only in callus regeneration the combination of *B. napus* + *B. juncea* led to shoot regeneration with the following electrical field parameters: ac (alternating current) field of 170 V/cm and 1.8 MHz and one dc (direct current) fusion pulse by 20 μ s of 1600 V/cm. From this experiment 52 tetraploid plants were obtained. Fourty-six of these plants could be identified as hybrids by use of microsatellite markers (Table 1). Shoot development was also obtained from fusion experiments between *B. napus* and *C. hispanica* (Table 2) with the parameters: ac field 180 V/cm and 1.8 MHz and one dc fusion pulse by 20 μ s of 1680 V/cm. These shoots are too small for identification yet.

In further experiments the parameters of the electrical field have to be optimised to increase the regeneration capacity.

Table 1: Combination *B. napus* (WW 1089) + *B. juncea* (CR 98/86)

Regenerated calli (n)	Calli with shoot regeneration (n)	Regenerated plants (n)	Tetraploid plants (n)	Hybrids (n)
24	6*	93	52	46

* on medium N3

Table 2: Combination *B. napus* + *C. hispanica*

Experiment	Regenerated calli (n)	Calli with shoot regeneration (n)
Maplus + CRA 13/81	89	11*
Maplus + CRA 15/84	28	2 [#]
Drakkar + CRA 15/84	45	2 ⁺

* on media N2, N3 and MS

[#] on medium N2

⁺ on media N2 and N3

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GENE ESCAPE STUDIES ON TRANSGENIC INDIAN MUSTARD(*Brassica juncea*)

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INTRODUCTION

Hybrids in Indian Mustard have become a reality as Proagro-PGS India Ltd. has been utilizing the Seedlink™ system(Mariani et al. 1990 and 1992) successfully for mustard hybridization programme. However, information regarding escape of transgenes to related crop species and safe isolation distance for growing transgenics is needed. Experiments were conducted from 1995 to 1997 to assess maximum distance and extent up to which transgene escape can take place in transgenic Indian Mustard under natural field conditions. Results of the experiments of 1995 have been published already.

MATERIALS AND METHODS

Trials consisting of transgenic mustard hybrids carrying *barnase*(male sterility inducer), *barstar*(fertility restorer) and *bar*(for resistance to herbicide glufosinate ammonium, active ingredient of Basta) genes alongwith checks were conducted in winters 1995-'96, 1996-'97 and 1997-'98 for yield and other agronomic characters. The trials were conducted in fields and locations permitted by Department of Biotechnology, Government of India. In 1995-'96 and 1996-'97, the testing location was Gurgaon. In 1997-'98, locations were four, i. e. Gurgaon, Bangalore, Bhinmal and Niwarsi. The trials were surrounded by ten non-transgenic mustard trapper rows sown at every five metre up to 50 metres in all sides of the field. Four hives of honeybees were kept at the four corners of the field to facilitate cross pollination. Eight seed samples were harvested from the corners and middle of each trapper row at the end of the season. The samples, 80 in total, were sown with both positive (Basta resistant) and negative (Basta susceptible) checks in two replications. The plot size was 5m x 0.6m, i. e two 5m rows at 30 cm row to row distance. Basta was sprayed on 20-25 days old seedlings at 2-4 true leaves stage to see extent of survival of plants after the spray. Plants surviving after the spray were rechecked for the presence of transgenes by a second spray of Basta and finally, screening by Polymerase Chain Reaction (P. C. R.). The percentage of plants surviving after Basta spray reveals the extent of gene escape from the transgenics to the plants of the trapper row from where the sample was drawn. These experiments were repeated for each location every year.

RESULTS AND DISCUSSION

The efficiency of the Basta spray and the Basta resistant *bar* gene was demonstrated in the experiments as casualty of plants in the positive checks and survival of plants in the negative checks were nil after the Basta sprays. Table number 1 shows the pooled means of survival percentage of plants after Basta spray of experiments of all the three years of testing, 1995-96 to 1997-'98. In 1995-'96, the transgenes escaped as far as 35 metres upto an extent of 0.007 percent and it escaped maximum at 5 metres at 0.161 percent (see table 1). In 1996-'97, the maximum distance up to which transgenes traveled was 30 metres and the extent was 0.01 percent. The maximum gene escape took place at 5 metres at 0.244 percent (see table 1). In 1997-'98, it was observed that transgenes escaped upto a maximum distance of 35m at 0.012 percent and the maximum extent was 0.241 percent at 5 metres (see table 1). The data remained consistent for years as well as for the different locations also. The role of

wind that it helps in the cross pollination of mustard, as reported earlier (GhoshDastidar and Varma, 1998) was noticed in the later years also.

Table 1: Data on Survival* of Plants after Basta Spray

Row	1995-'96 (Gurgaon)		1996-'97 (Gurgaon)		1997-'98 (Gurgaon, Bangalore, Niwarsi, Bhinmal)	
	Total No. of plants	Mean Survival (%)	Total No. of plants	Mean Survival (%)	Total No. of plants	Mean Survival (%)
50 m	10332	0.000	6374	0.000	7291	0.000
45 m	13160	0.000	8634	0.000	7646	0.000
40 m	16120	0.000	10977	0.000	8207	0.000
35 m	15524	0.007	8922	0.000	8210	0.012
30 m	16471	0.012	9960	0.010	7537	0.040
25 m	15133	0.007	9021	0.011	7373	0.014
20 m	15416	0.020	10304	0.019	7756	0.052
15 m	13718	0.044	7668	0.039	7621	0.066
10 m	13509	0.037	7914	0.051	7794	0.064
5 m	13052	0.161	7365	0.244	7891	0.241

* Survival = No. of plants living after spray/ No. of plants living before spray x 100

The maximum distances up to which flow of transgenes occurred were 30 and 35 metres in 1996-'97 and 1997-'98 respectively. This is in conformity with the result of 1995-'96 experiments when the maximum distance for transgene escape was found at 35 metres (GhoshDastidar and Varma, 1998). It is also noted that in all the three years, the maximum transgene escape took place within 5 metres and the extent was less than one percent.

CONCLUSION

It can be concluded that 35 metres is the safe distance for isolation in transgenic Indian mustard and maximum cross pollination can take place up to a distance of 5 metres only. It can also be concluded that under natural circumstances, cross pollination in transgenic Indian mustard is less than one percent.

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Development of YSMS-6, a genetic male sterile line of yellow sarson
(*Brassica campestris* var yellow sarson Prain)

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Scope of hybrid breeding to overcome yield plateauing is well known. The development of genetic male sterile line offers a feasible approach to hybrid breeding in yellow sarson (Ram Bhajan *et al.*, 1993). However, certain inherent deficiencies like lack of genetic marker to facilitate selective elimination of fertile sibs in vegetative stage in hybrid seed production plots using such recessive genetic male sterile lines limit their use in hybrid breeding programme. Search for more such sources could possibly lead to more efficient system. This paper reports the development of a new male sterile line in yellow sarson.

In one of the yellow sarson line, collected from farmers fields of eastern Uttar Pradesh (India), a few male sterile plants were identified during flowering. In the following season, plant to row progenies raised from open pollinated seeds of male sterile plants, showed segregation for male sterile and male fertile plants. Crossing of male sterile plants with fertile sibs and with other collection of yellow sarson, brown sarson and toria lines subsequently showed that this male sterility is recessive to fertility.

On the contrary, when this male sterile line (YSMS-6) was crossed with a local collection of yellow sarson (NDYS-37), male sterility was found to be completely maintained in F_1 during 1993-94. But this could not be reproduced faithfully in subsequent years. During 1994-95 and 1995-96, this repeated cross gave segregation for male sterile and male fertile plants in F_1 . When resorted to plant by plant crossing, assuming that NDYS-37 population could be a mixture of variable genotypes for the loci involved. This approach subsequently led to the isolation of three types of plants of NDYS-37-(i) one giving complete maintenance of male sterility, (ii) showing segregation for male sterile and fertile plants, and (iii) showing complete fertile expression in F_1 . Efforts are underway

to verify these observations and establish component lines of NDYS-37 which could give faithful expression in crosses with YSMS-6.

Quantitative measurement of floral parts showed that sepals, petals, stamens and gynoecium were comparatively smaller in size in freshly opened flowers of male sterile plants than in their fertile sibs (Table-1).

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Table-1 : Relative variation in floral parts of freshly opened flowers of male sterile and male fertile plants (in mm)

Floral parts	YSMS-6	YSMF-6
Sepal length	5.40	6.20
Petal length	8.95	10.25
Petal width	5.20	6.90
Stamen length	5.70	8.91
Filament length	3.80	5.85
Anther length	1.90	2.16
Gynoecium	6.80	7.40

EFFECT OF SALINITY ON CHLOROPHYLL AND FREE PROLINE OF MUSTARD (BRSSICA JUNCEA L.)

R.S.Parti, S.K. Gupta and M.L. Chhabra

Introduction

Salinity is one of the major limiting factors to the agricultural productivity. The detrimental effects of salinity are due to the influence of Ca^{++} , Mg^{++} , Na^+ , Cl^- and So_4^- ; the major ions, on the water activity of the external solution which affects the water status of the plant due to osmotic effect and also the direct effects of the ions on the physiological and biochemical functions of the cells. (Greenway and Munns, 1980). The present investigation was carried out to study the effect of Salinity on chlorophyll and free proline contents of leaf, seed and siliquae wall of mustard crop.

Materials and Methods

The experimnt was conducted in the green house. Mustard crop (Cv. RH-30) was raised in earthen pots, filled with 5kg of sandy soil (ECe 1.8 ds/m). The pots were lived with polyethylene bags. The desired salinity lelvels (ECe 4,8 and 12 ds/m) were obtained by adding Cl^- and So_4^- salts of Na^+ , Ca^{++} and Mg^{++} . The Cl^- and So_4^- in the ratio of 7:3 while Na^+ , Ca^{++} and Mg^{++} in the proportion of 4:1:3 were taken (Sharma and Manchanda, 1997). A basal dose of nitrogen (60ppm) phosphorus (30ppm) and micronutrients was also applied. After the emergence of seedlings, three uniforms plants/pot were retained. The leaf samples were taken at 35 days after Sowing and siliquae samples were taken at physiological maturity which were further separated into seeds and siliquae wall. These were immediately analysed for total chlorophyll, chlorophyll a, chlorophyll b (Hiscox and Israelstam, 1979) and free proline contents (Bates et al., 1973).

Results and Discussion

Table 1 indicate that chlorephyll a, chlorophyll b and total chlorophyll content generally decreased as salinity levels increased in leaf, seed and siliquae wall. At ECe 12 the reduction was quite marked. The concentration of chlorophyll was higher in leaf as compared to seed and siliquae wall. According to several workers, salinity decreases the chlorophyll content of plant (Garg and Lahiri, 1986). Free proline content was observed to increase consistently with increase in salinity levels in leaf, seed and siliquae wall (Table-1). The increase was more prenounce at higher level of salinity (ECe12). The proline concentration was maximum in siliquae wall followed by seed and leaf. It is suggested that proline accumulation does not initiate salinity adaptation but may itself be triggered to occur as a result of the initiation of other responses to salinity stress (Hasegawa et. al., 1986). Alia et. al. (1993) opine that proline accumulation has adaptive significance as it lowers the generation of free radicals and thus, reduces lipid peroxidation linked membrane deterioration under salt stress. The adaptive role of proline is related to survival rather than maintenance growth (Greenway and Munns, 1980).

Table:1 Effect of different levels of salinity on chlorophyll and free proline contents of leaf, seed and siliquae wall.

Salinity Levels (ECe dsm ⁻¹)	Chlorophyll a	Chlorophyll b mg/g. fr.wt.	Total chlorophyll	Free Proline (ug/g fr.wt.)
<u>Leaf*</u>				
Contrl	1.13 + 0.10	0.12 + 0.02	1.25 + 0.13	73.75 + 0.42
4.	1.04 + 0.05	0.12 + 0.01	1.16 + 0.04	75.22 + 0.32
8.	1.03 + 0.04	0.11 + 0.01	1.14 + 0.08	83.24 + 0.51
12	0.83 + 0.12	0.09 + 0.02	0.92 + 0.12	106.20 + 0.30
<u>Siliquae Wall**</u>				
Control	0.182 + 0.020	0.041 + 0.001	0.223 + 0.021	221.9 + 1.53
4	0.146 + 0.015	0.032 + 0.010	0.178 + 0.025	260.3 + 2.91
8	0.125 + 0.012	0.037 + 0.003	0.162 + 0.015	511.08 + 3.21
12	0.117 + 0.011	0.019 + 0.001	0.136 + 0.002	865.68 + 1.82
<u>Seed**</u>				
Control	0.127 + 0.021	0.021 + 0.001	0.148 + 0.021	189.5 + 2.80
4	0.119 + 0.030	0.020 + 0.002	0.139 + 0.042	205.1 + 3.27
8	0.117 + 0.015	0.010 + 0.001	0.127 + 0.016	238.2 + 1.73
12	0.112 + 0.012	0.009 + 0.001	0.121 + 0.011	291.16 + 2.11

* Leaf Samples were taken at 35 days after sowing.

** Siliquae wall and seed samples were taken at physiological maturity.

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Level of self-incompatibility in green sprouting broccoli (*Brassica oleracea* L var. *italica* Plenck)

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This is first ever conducted study about level of self incompatibility in green sprouting broccoli in India. This investigation deals with self-incompatibility aspects in four varieties of green sprouting broccoli of Indian origin.

The four cultivars used in the present study were Palam Samridhi, DBGB-12, DBGB-13 and DBGB-14. The crop was raised during 1996-97. The inflorescence at bud stage was partitioned into three parts. The first part was labelled and left to allow open pollination by insects. The second part was used for selfing by bud pollination and the third part was left for natural selfing by covering with muslin cloth bag. Twenty five siliqua from each part/plant were taken randomly and seeds were counted.

The compatibility status of the genotypes were assessed by following the methods advocated by Watts (1963) and Nieuwhof(1974) by determining fertility index. Mean seed number per pod in bud pollination expressed as percentage of mean seed number per pod under open pollination gave percentage self-compatibility (Watts, 1968). Bud pollination was done in support to the results obtained and further to see its feasibility for producing S_1 lines in case there is self-incompatibility.

The level of self-compatibility (Table 1) ranged from 2.87 to 6.96%. This indicated a high level of self-incompatibility at the varietal level. DPGB-13 had highest level of self-compatibility (6.96%). The seed setting under bud pollination was comparable to the seed setting under open pollination. Majority of plants studied in all the four varieties, were in the range of 0 to 10 % compatibility grade (Table-2), indicating the presence of high level of self-incompatibility in these genotypes. However, a small number of plants with fairly high self-compatibility were found in DPGB-13 and DPGB-14. Poor seed setting under bagged inflorescence, which is an indication of high level of self-incompatibility was further substantiated by the fact that normal seed setting was observed by bud pollination. The results obtained in this investigation suggested that a wide range of incompatibility from complete self-incompatible to self-compatible lines are present in this crop. However, outbreeding seems to be the main feature of breeding systems in these varieties. Consequently, while using these varieties for breeding, one must maintain acceptable level of heterozygosity in the production of synthetics/composites or increase heterozygosity for the production of F_1 hybrids. For the material exhibiting strong self-incompatibility reaction, mass selection and mass pedigree method of breeding could also be effective.

This investigation has demonstrated high level of self-incompatibility in four varieties of green sprouting broccoli along with success of bud pollination which offer a promise to exploit heterosis in this crop.

Summary

Four varieties of green sprouting broccoli were tested for level of self-incompatibility. These varieties exhibited high level of self-incompatibility along with success of bud pollination which offer a promise to exploit heterosis in this crop.

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Table-1 Mean seed set per siliqua from self-pollination by bagging, bud pollination and from open pollination.

S.No.Variety	Self pollination		Open Pollination	%Self-compatibility
	By Bagging	By Bud Pollination		
1. Palam Samridhi	0.38	10.56	13.20	2.87
2. DPGB-12	0.51	5.29	9.45	5.39
3. DPGB-13	0.39	3.55	5.60	6.96
4. DPGB-14	0.23	2.27	5.02	4.58

Table 2 Number of plants in different compatibility grades in four genotypes of green sprouting broccoli.

S.No.	Genotype	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100	Total
1.	Palam Samridhi	16	5	2	-	3	1	1	-	-	-	28
2.	DPGB-12	18	5	1	1	-	1	-	-	1	-	27
3.	DPGB-13	12	4	2	2	-	-	-	-	1	1	22
4.	DPGB-14	18	1	-	-	1	1	-	1	1	3	26

PARAQUAT INDUCED CHANGES OF CHLOROPHYLL CONTENT IN SHOOT CULTURE OF CABBAGE (*BRASSICA OLERACEA* VAR. *CAPITATA*)

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INTRODUCTION

Paraquat is a highly toxic broad-spectrum herbicide (Halliwell and Utteridge, 1989) widely used in agriculture. The mode of paraquat action in plants involves its reduction through the electron transport system in chloroplasts and reoxidation by molecular oxygen. Reoxidation creates superoxide radicals, which are actually lethal agents of the herbicide treatment (Rabinowitch and Fridovich, 1983). It is considered that the absence of reliable detoxication pathways is responsible not only for the herbicide action of paraquat but also for lethal poisonings of animals and humans. Therefore we investigated the effect of paraquat on the chlorophyll content in shoot cultures of some cabbage (*Brassica oleracea* var. *capitata*) genotypes.

MATERIAL AND METHODS

Shoot culture of cabbage was obtained from the segments of aseptically grown seedlings. Fully developed shoots were cultured on medium containing MS (Murashige and Skoog, 1962) inorganic salts, vitamins and cofactors, supplemented with 5.0 mg l⁻¹ IBA and paraquat at 0.5, 2.0, 10.0, 50.0 mg l⁻¹. After 24 hours plants were subcultured to hormone free medium. Chlorophyll content was measured spectrophotometrically in acetone extracts prepared from cultures 28 days after paraquat treatment. Measurements were replicated at least three times. Material used for experiments were four promising cabbage genotypes produced through our breeding programs.

RESULTS AND DISCUSSION

We expected that the increase of paraquat concentration in the medium will result in decrease of chlorophyll *a* and *b* content. However, in genotype P44 increased concentration of chlorophyll *a* was registered at low paraquat concentrations, 0.5 and 2.0 mg l⁻¹ (Fig. 1). This confirmed our previous findings that P44 plants manifested good growth and lateral shoot production in the medium supplemented with 0.5 and 2.0 mg l⁻¹ paraquat (Sretenovic Rajiicic et al., 1996). Also in genotype P194 paraquat at 10.0 mg l⁻¹ slightly increased chlorophyll *b* content (Fig. 2).

Do we have signs of possible tolerance towards paraquat in P44 genotype? This question remains to be investigated with more detail. At this stage we believe that there are ways by which plants attempt to overcome detrimental paraquat effects. Considering paraquat's mode of action, it is possible that small amount of the chemical can be stimulative for the plant growth. As it is said, "medicine or poison, depends on the concentration".

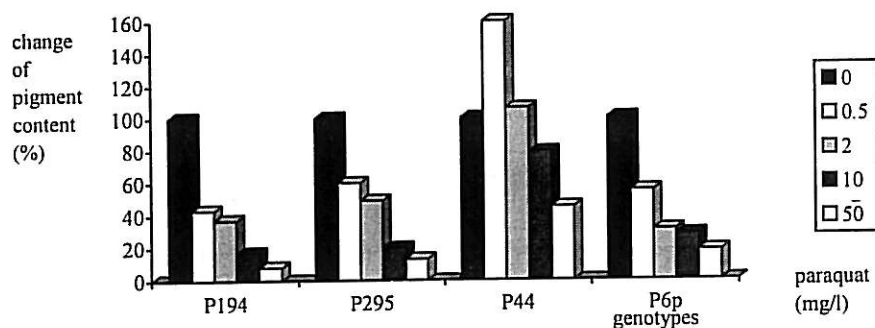


Fig. 1. Content of chlorophyll a

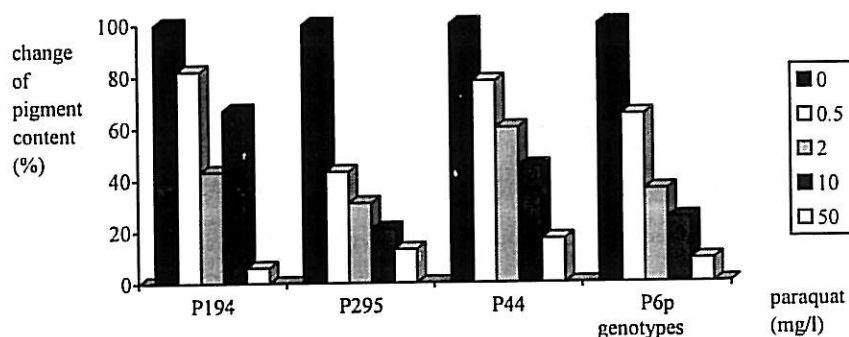


Fig. 2. Content of chlorophyll b

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Mass Selection for Seed Yield Improvement in Toria

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Toria (*Brassica campestris* var toria Duth & Full) is an important oiliferous Brassicaceae in northern India, which on account of its short duration fits in very well in multiple cropping system. Results of national yield trials over years have shown stagnating trends in yielding ability of new varieties which are in pipeline. The use of advance generation of promising crosses or blends of several lines/populations to form composites has been commonly practiced to evolve new varieties in this crop. Meagre efforts have, however, been made to improve toria populations by recurrent selection. This paper reports the results of two cycles of mass selection in three composite populations viz NDTC-9501, NDTC-9502 and NDTC-9513 of toria.

Mass selection was initiated in these three populations during 1996-97. For mass selection, populations were grown in separate isolations. Selection criteria was overall phenotypic superiority of plants together with uniformity for important traits like plant height and maturity with selection intensity of 10%. Three mass selected cycle-2 (MSC₂) populations alongwith their base populations (cycle-0) and a standard variety, PT-303 were evaluated during 1998-99 in RBD replicated thrice each with 6 rows of 3 m long. All recommended cultural practices were followed to raise a good crop. Observations on seed yield and maturity were recorded on plot basis and for ancillary characters on 5 competitive plants in each plot.

Analysis of variance showed significant differences among populations for all the characters. Visible improvement in seed yield was observed in all the three population (Table-1). Maximum *per se* improvement could be realised in NDTC-9502 MSC₂ (9.58%) over its C₀ population which was followed by 9.09% in NDTC-9513-MSC₂ and the lowest was in NDTC-9501-MSC₂ (5.10%). The per-cycle gain for seed yield was 4.79%, 4.55% and 2.55% for NDTC-9502, NDTC-

9513 and NDTC-9501, respectively. Overall, NDTC-9513- MSC_2 was the best population with 16.51% superiority over PT-303. Maturity duration of C_2 and C_0 populations was at par. Selection had positive effect on 1000-seed weight only in NDTC-9501 and NDTC-9502. Improvement in seed yield was accompanied by increase in plant height in all C_2 populations over corresponding C_0 populations. These results warrant for further scope of improvements in seed yield of these populations following mass selection.

Table 1. Performance of mass selected Cycle-2 populations for seed yield and other traits in toria

S. No.	Populations	Plant height (cm)	1000-seed weight (g)	Days to maturity	Seed yield	
					Kg/ha	% Superiority over C_0 population
1.	NDTC-9501 MSC_2	126.6	2.14	96	974.07	5.10
2.	NDTC-9501 C_0	118.1	1.87	97	888.88	--
3.	NDTC-9502 MSC_2	124.6	2.37	97	1018.51	9.58
4.	NDTC-9502 C_0	122.9	2.34	98	968.51	--
5.	NDTC-9513 MSC_2	126.7	2.53	97	1111.11	9.09
6.	NDTC-9513 C_0	125.9	2.47	97	1018.51	--
7.	PT-303 (SV)	111.2	2.47	99	953.70	--
	General mean	124.10	2.36	97.70	1031.48	--
	SEm \pm	6.01	--	0.44	48.15	--
	CD (5%)	18.34	--	1.32	135.19	--
	CV (%)	8.44	--	0.77	7.45	--

EFFECT OF FREEZING TREATMENT ON OIL CONTENT AND FATTY ACID COMPOSITION OF MUSTARD GENOTYPES (Brassica juncea L.)

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Introduction

Mustard is highly frost susceptible crop (Dhawan and Chhabra, 1983) and also exhibit marked biochemical changes when exposed to low temperature (Alberdi et. al., 1993). The present investigation was undertaken to study the effect of freezing treatment on oil content and fatty acid composition of mustard genotypes.

Materials and Methods

Mustard genotypes were grown in pots in screen house. Thirty days after anthesis, freezing treatment was given by keeping the plants in deep freezer at $-3.0^{\circ}\text{C} \pm 0.5^{\circ}$ for 2h (Dhawan and Chhabra, 1983). One set of mustard genotypes was kept untreated (control). After treatment plants were left to mature in screen house. At maturity seed samples were collected and were analysed for oil content by NMR technique, for fatty acid composition oil from each sample was extracted by petroleum ether, methyl esters were prepared and separated in a gas chromatograph (Luddy et al., 1968).

Results and Discussion

Oil content and fatty acid composition was affected considerably by freezing treatment (Table-I). On mean basis oil content was 42.20% in control while it was 38.43% in the treated samples. There was about 4% reduction in oil content with freezing treatment. Among the fatty acids, erucic acid which is the main fatty acid decreased while oleic and linoleic acids increased with freezing treatment in almost all the genotypes. Linolenic + eicosenoic acids, however remained unchanged. These results indicate a net increase in the unsaturation with freezing treatment. During freezing, fatty acid unsaturation was observed to increase in number of crop plants (Zuniga et al., 1990; Larson et. al., 1992).

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Table : 1 Effect of freezing treatment on oil Content and fatty acid composition of mustard genotypes.

Genotypes	Oil Content %		Fatty acids (%)											
			16:0		18:0		18:1		18:2		18:3+20:1		22:1	
	C	T	C	T	C	T	C	T	C	T	C	T	C	T
RH-819	42.2	38.7	2.9	3.2	0.8	1.0	10.4	13.7	11.6	13.8	20.0	20.1	51.8	48.1
RH-8114	41.8	39.8	2.7	3.0	0.7	0.9	12.5	13.1	13.2	13.9	22.8	21.8	48.7	48.0
RH-8570	41.7	39.1	2.8	2.5	0.9	0.8	11.7	12.3	13.4	15.1	21.4	21.7	49.3	46.8
RH-8519	42.5	37.3	2.4	2.6	1.1	1.0	11.8	14.8	12.7	15.6	20.4	20.7	50.3	46.2
RH-7387	40.7	37.1	2.5	2.3	0.9	0.8	10.6	13.1	12.5	16.3	22.1	21.2	51.2	47.5
RH-7859	42.1	40.7	2.2	2.4	1.0	1.1	11.3	12.1	12.6	14.8	21.8	21.0	50.8	48.6
RH-832	41.8	38.3	2.3	2.7	1.1	1.0	10.4	13.2	14.3	16.7	21.4	19.7	49.7	46.1
RH-8113	42.3	40.4	2.5	2.7	0.8	1.0	9.5	12.0	14.5	16.8	19.9	19.8	52.8	48.1
RH-7846	40.2	37.8	3.1	2.9	0.9	0.8	10.3	12.3	15.2	18.7	20.4	19.1	50.6	46.4
RH-8693	42.5	38.4	3.0	2.8	0.7	0.9	11.3	12.5	14.0	16.8	21.4	20.9	49.5	46.2
RH-8605	43.4	39.5	2.5	2.6	1.0	0.8	10.2	13.4	12.1	17.6	20.9	20.2	51.8	46.2
RH-8606	42.2	30.4	2.8	3.2	1.0	1.1	10.1	14.6	13.3	19.7	20.4	18.6	52.7	43.1
RH-8688	43.2	42.1	2.7	2.4	0.9	1.0	11.3	11.7	12.4	14.4	22.1	21.3	50.3	48.8
RH-8315	42.8	35.1	2.5	2.8	1.0	0.8	11.6	15.1	12.4	17.8	21.1	20.6	51.3	45.4
RH-781	43.5	41.7	2.7	2.2	1.2	0.9	9.8	10.7	14.4	16.7	21.4	20.7	50.3	48.7
Mean	42.20	38.43	2.74	2.65	0.96	0.92	10.85	12.72	13.31	16.08	21.18	20.56	50.70	46.95

C=Untreated (Control); T= Treated

STUDY OF HETEROSIS FOR SEED YIELD IN INDIAN MUSTARD [*Brassica juncea* (L.) Czern. and Coss.]

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INTRODUCTION

Mustard (*Brassica juncea*) is an important edible-oils yielding crop of India and neighbouring countries. The species being largely self-pollinated, offers scope for the production of commercial hybrids if cross combinations exhibit adequately high heterosis. Recently the availability of cytoplasmic male sterile lines (cms lines) in *B. juncea* (Prakash and Chopra 1990) and production of fertility restorer system (Kirti *et al.* 1997) has stimulated interest of plant breeders to exploit the heterosis breeding (Pradhan *et al.* 1993; and Agarwal and Badwal 1998). This report presents results pointing to positive heterosis (heterosis in desirable directions) for seed yield in combinations among 96 intervarietal crosses (F1) obtained by crossing 6 lines and 16 testers of *B. juncea* in LT mating design.

MATERIALS AND METHOD

Six varieties/strains of *Brassica juncea* were crossed to 16 germ plasm lines/selections of *B. juncea* in a line tester mating design. Ninety six crosses thus obtained were evaluated for seed yield, and key yield components (primary branches, pods on them and number of seeds in pods), by growing them in experimental field along with their parents and the best performing national variety (cv. Varuna) in replicated, randomised block design. Row and plant distances were kept 60 cm and 8 cm respectively. Two irrigations, 80 kg 'N', 60 kg 'P', 40 kg 'K' and 25 kg 'S' per hectare were applied. Plant protection and other agronomical practices were followed as per recommendations. Data on number of primary branches (PB) pods on 3 PB and number of seeds in 5 pods were recorded on 3 plants randomly drawn from each cross and parent when the crop attained physiological maturity. After harvesting the plot, seed yield was also recorded for both crosses and parents. Means for each character were worked out and +ve heterosis over better parent and the best national variety was calculated.

RESULTS AND DISCUSSION

Analysis of different sources of variation showed presence of significant differences among hybrids (crosses), parents (females and males) and parent versus hybrids. This indicated that there is good genetic diversity among parents to produce heterotic crosses. This was evidenced from both large proportion (66.6%) of crosses being heterotic and some crosses being highly heterotic (97.7% and 66.3 per cent heterosis over better parent and the best national var. respectively (Table 1).

Seven cross combinations (Table 1) exhibited more than 30% heterosis over both better parent and best national check. Another 11 crosses showed from 31.2 to 71.3% heterosis over better parent while six other crosses registered 32.5 to 52.4 per cent heterosis over the best national var. The results indicate that gain in yield is mainly due to increased number of pods and to some extent by the combination of branches and number of seeds per pod.

Among the 6 lines (female parents) the BIO.322-93 was the best performer in that it showed genetic superiority over both better parents and the best national check. It progenated 87.5% heterotic crosses. Among the male parents (testers), BIO-467-95 and RLM-198 were sires that contributed the most to heterosis for seed yield. The study indicates that there is adequate genetic divergence among Indian mustard (*B. juncea*) lines to generate a successful commercial hybrid programme. A judicious choice of material is critical to seeking gain in yield in this important crop.

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Table 1. Crosses showing over 30% heterosis for seed yield and key characters contributing towards yield

Cross	Seed yield		PB/Plant		Pods/PB		Seeds/Silq.	
	Het. over BP	Het. over NV	Het. over BP	Het. over NV	Het. over BP	Het. over NV	Het. over BP	Het. over NV
1. Crosses showing heterosis (%) over better parent and national var.								
PJK x RLM-198	97.7	39.8	-	43.3	29.5	48.5	-	0.8
PJK x CSR-499	73.4	44.0	-	11.7	4.9	18.4	-	16.4
BIO-322-93 x BIO-467-95	43.1	63.3	-	20.0	29.4	60.3	-	3.2
BIO-322-93 x JMG-401	42.6	62.7	16.2	43.3	10.2	34.9	-	-
BIO-322-93 x BIO-341-92	40.5	60.4	18.2	30.0	4.5	27.9	-	-
BIO-322-93 x CSR-1240	33.5	52.4	31.5	60.0	6.6	30.5	10.3	20.0
BIO-322-93 x BIO-YSR	31.5	66.3	-	33.3	19.8	46.7	-	12.8
2. Crosses showing heterosis (%) over better parent								
PJK x JMG-401	71.3	21.1	-	13.3	-	15.4	5.3	12.0
PJK x JMG-417	64.3	22.1	15.6	23.3	7.4	6.3	8.3	15.2
PJK x CSR-1246	34.6	5.0	-	1.7	1.7	13.2	-	15.2
BIO-772 x RLM-198	64.7	7.0	-	38.3	8.3	24.3	-	-
BIO-772 x BIO-53-93	38.5	28.9	23.6	46.7	16.5	22.1	-	20.8
BIO-772 x JMG-417	34.1	-	-	-	4.5	3.3	0.8	7.2
BIO-772 x CSR-499	33.6	11.0	-	33.3	1.6	14.7	-	17.6
BIO-200-94 x RLM-198	67.7	3.6	-	48.3	9.1	27.6	4.4	13.6
BIO-200-94 x BIO-467-95	32.3	22.5	-	-	4.5	29.5	8.8	18.4
BIO-200-94 x CSR-1246	31.3	2.0	-	13.3	11.9	24.6	-	11.2
BIO-344-94 x RLM-198	31.2	-	3.7	85.0	16.3	33.5	-	7.2
3. Crosses showing heterosis (%) over National var.								
BIO-322-93 x GM-1	23.5	52.4	4.5	15.0	18.0	44.5	3.0	11.2
BIO-322-93 x JMG-414	26.5	44.4	-	6.7	-	14.7	-	13.6
BIO-322-93 x BIO-53-93	25.7	43.4	21.2	33.3	3.6	26.8	-	18.4
BIO-322-93 x CSR-1246	23.0	40.4	21.3	51.7	-	14.3	-	13.6
BIO-322-93 x BIO-466-95	22.5	39.8	3.0	13.3	16.8	43.0	0.7	11.2
BIO-322-93 x CSR-258	16.1	32.5	16.7	40.0	-	31.3	-	13.6

PB/Plant = No. of primary branches per plant

Pods/PB = No. of pods per PB

Seeds/Silq.= No. of seeds per pod

BP = Better parent

NV = National variety

PJK = cv. Pusa Jai Kisan

Het over NV = Superiority of cross over best commercial National var.

Combining ability analysis using genetic male sterility in yellow sarson (*Brassica campestris* L.)

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An integral component of hybrid breeding in any crop is the assessment of general and specific combining ability. In the present study, three genic male sterile lines (YSMS-8163, YSMS-2 and YSMS-6) developed at this University were used as seed parent and 17 diverse strains as testers (males). These 20 parents alongwith their 51 hybrids were evaluated in RBD with 3 replications. Recommended cultural practices were followed for raising a good crop. Observations were taken on 10 metric traits including seed yield and oil content. Line x tester analysis as out lined by Kempthorne (1957) was followed.

The estimates of variances due to lines x testers were significant for all the 10 characters viz. days to maturity (DM), plant height (PH), length of main raceme (LMR), primary branches/plant (PB), siliquae on main raceme (SMR), siliquae/plant (S/P), seeds /siliquae (S/S), 1000-seed weight (TW), seed yield (SY), and oil content (OC) (Table-1). The results, thus indicated preponderance of non-additive genetic component for expression of these traits. Higher magnitude of degree of dominance ($\sqrt{\sigma^2_s/2\sigma^2_g} > 1$) for LMR, DM, PB, TW, SY and OC indicate predominance of non-additive gene action governing these characters. Low to moderate estimate of narrow sense heritability (h^2_n) (<30%) for these characters except SY was in confirmity with above observations.

Variance due to females was non-significant for two characters (PH, DM) while variance due to males was non-significant for three characters (PH, LMR, OC). Higher estimates of σ^2_A than σ^2_D coupled with lower estimates of degree of dominance (<1) and relatively higher magnitude of h^2_n were found for SMR, S/P and S/S. Predominant role of non-additive genes in controlling the expression of various quantitative characters in Colza has been reported by Verma *et al.* (1989), and Varshney and Rao (1997).

The presence of non-additive genetic variance in large quanta under lying the inheritance of important traits like DM, SY and OC favour the idea of maintenance of heterozygosity for desired expression of these traits. These observations, thus offer greater opportunities for boosting up the expression of ultimate end products i.e. seed yield and oil content through heterosis breeding.

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Table 1 : Analysis of variance for combining ability, variance components and heritability in narrow sense for 10 characters in yellow sarson

Characters Sources/ Components	Mean Squares/estimates											OC
	DM	PH	LMR	PB	SMR	S/P	S/S	TW	SY			
Lines	6.30	367.78**	323.94** +	13.56** +	733.69** ++	11516.62** ++	738.53** ++	0.94** ++	100.87** ++			9.10** ++
Testers	37.11** ++	303.53**	170.41**	7.84** +	159.27** +	10209.33** ++	151.89** ++	0.33** +	39.99** ++			2.25**
Lines X Testers	13.52**	357.02**	97.93**	3.74**	81.15**	1686.95**	45.87**	0.16**	14.87**			1.66**
Error	8.54	24.06	12.84	0.82	15.54	43.29	2.32	0.04	0.86			0.23
σ^2g (Males)	2.62	-5.94	8.05	0.46	8.68	946.93	11.78	0.02	2.79			0.07
σ^2g (Females)	-0.14	0.21	4.43	0.19	12.79	192.74	13.58	0.02	1.69			0.15
σ^2g (Pooled)	0.27	-0.71	4.97	0.23	12.18	305.87	13.31	0.015	1.85			0.13
σ^2s	1.66	110.99	28.36	0.97	21.87	547.89	14.51	0.04	4.67			0.47
$\sqrt{\sigma^2s/2\sigma^2g}$	1.74	--	1.69	1.45	0.95	0.95	0.74	1.15	1.12			1.32
σ^2A	0.55	-1.42	9.95	0.46	24.36	611.74	26.62	0.03	3.70			0.27
σ^2D	1.66	110.99	28.36	0.97	21.87	547.89	14.51	0.04	4.67			0.47
h^2n (%)	5.12	--	19.45	20.44	39.44	51.13	61.27	27.78	40.09			27.84

-- h^2n value not calculated due to negative estimate of σ^2g (pooled) i.e. $\sigma^2A = \text{zero}$.

* **, Significant at 5 and 1 per cent probability levels, respectively.

+ , ++, Significant at 5 and 1 per cent probability levels against lines x testers interaction, respectively.

INHERITANCE OF POD ANGLE IN *BRASSICA RAPA*

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In the forms of *Brassica rapa* L. commonly grown in Canada as an oilseed crop, the pods point outwards and upwards from the stem. An unusual introduction with distinctly downturned pods was obtained and crossed to the conventional form. The F_1 from a cross between the two types resembled the conventional parent, showing that the downturned pod character was recessive, and F_2 and BC_1 progeny segregation suggested a three gene situation.

Materials and methods

The parent with the downturned pods was a Sarson type introduction, which was obtained via the collection at Saskatoon (their identification R4105), obtained in turn from the University of California Davis (their identification 79-19). This downturned pod type is referred to as "parapluie". The conventional pod angled material used the cultivar AC Sunbeam.

All materials were grown in pots in a greenhouse. Crosses were made using bud pollination, and F_2 material was produced by interpollinating F_1 plants at random.

Pod angles were measured at the centre of the main raceme at maturity. A single pod per plant, judged to be representative of the centre of the raceme was used, and the arms of an adjustable protractor lined up with the main stem and the axis of the pod. Where the pod curved the straight line between the attachment of the peduncle and the base of the beak was used as the pod axis. In cases where the line of the peduncle differed from the line of the pod axis, the line of the pod axis was taken. Angles were recorded using pointing down parallel to the stem as 0° and fully erect as 180° .

Results and discussion

In the "parapluie" parent the upper pods were generally held at a lower angles than those further down the raceme. In addition the pods were fatter than those of the conventional material, making angle measurement more difficult.

The F_1 plants all resembled the conventional material (Fig 1), indicating that the downturned pod trait is recessive. Only a very small proportion of either the BC_1 (Fig 2) or F_2 (Fig 3) population exhibited the downturned pod character, and no plants showing the extreme angles were recovered. Using a cut-off of 65° the BC_1 segregated in a 85:6 ratio for normal to downturned, and the F_2 segregated in a 89:3 ratio. These values are not significantly different (χ^2) from those predicted for a 3 gene pair model (7:1 and 63:1 respectively). Kelly *et al.* (1995) reported that in *B. napus* the upright pod character was dominant to horizontal pods, and there were possibly two gene pairs involved. Thus in both species the more upright pod habit seems to be a dominant trait.

Kelly, A., Fray, M., Arthur, E.A., Lydiate, D.J., and Evans, E. 1995. The genetic control of petalless flowers and upright pods. Proc 9th Int. Rapeseed Congr. Cambridge, UK, July 4-7. p732-734

Fig 1, parent and F1 pod angles

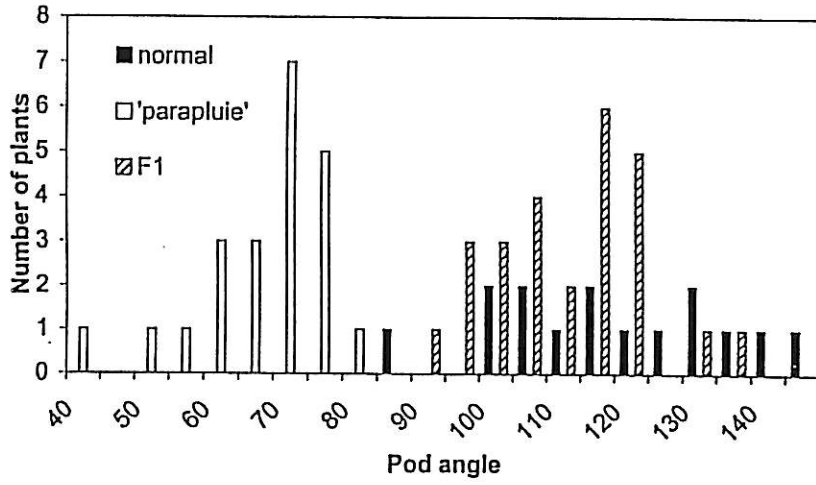


Fig 2, BC1 pod angles

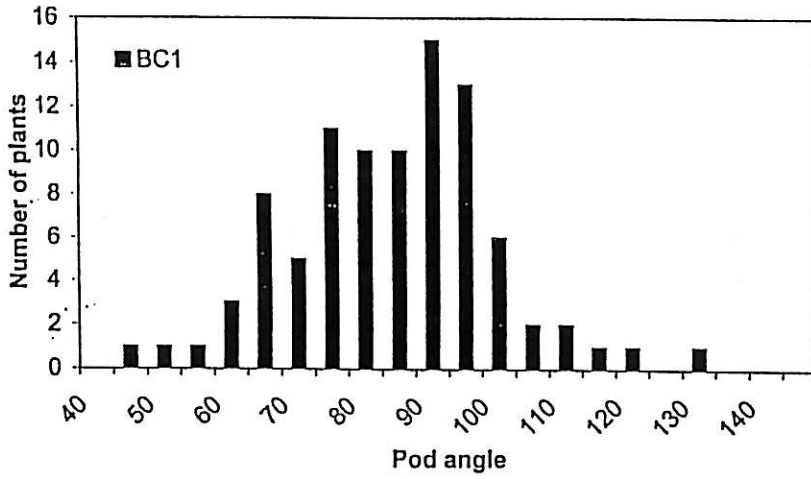
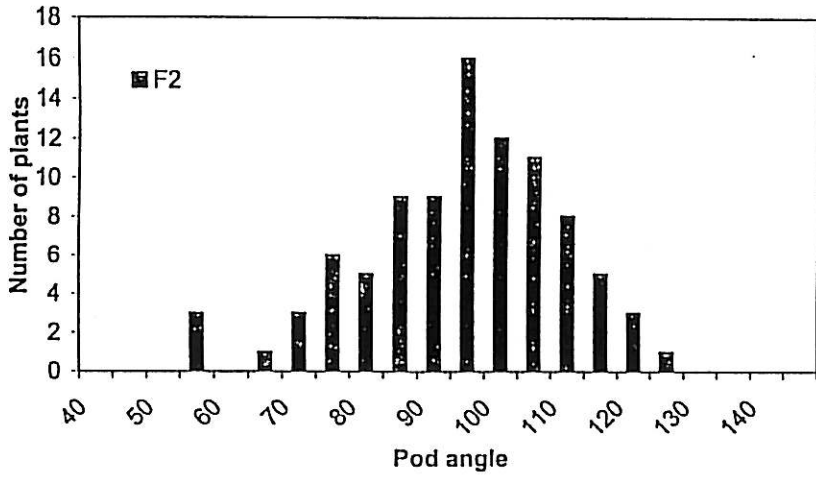


Fig 3, F2 pod angles



A 'parapluie' plant

RELATIONSHIP BETWEEN SEED SIZE AND OIL CONTENT IN *BRASSICA RAPA*

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During the course of the canola (*Brassica rapa* L., summer turnip rape) breeding program at Beaverlodge a number of populations were developed which had an increased seed size. This was considered potentially useful for several reasons. The decreased hull to embryo ratio in larger seeds would theoretically result in lower fibre levels in the residual meal. Larger seeds may also be easier to dehull, with a resultant improvement in the meal quality. Larger seeds would also produce larger cotyledons at emergence, resulting in less damage due to flea beetle predation (see Elliott, 1991).

It became apparent that increased seed size in many cases seemed to be associated with reduced oil content, which has been reported previously in other oilseed Brassicas, and in winter *B. rapa* (Olsson 1960). Available data sets from several years of trials were re-analysed to determine the correlation between seed size and oil content. Correlations were calculated within populations which were derived from a common origin, and where simultaneous selection for seed size and oil content had not been made since that population had been constituted.

Three classes of populations were examined; a) populations derived from conventional oilseed canola (low erucic acid, low glucosinolates) *B. rapa*, b) populations derived from crosses between rapeseed (high erucic acid, high glucosinolates) and canola in which selection for low erucic acid had been made, and c) populations derived from interspecific crosses. Data from these various populations are reported in Table 1.

Correlations between oil content and seed size varied considerably. The only statistically significant correlations were negative. In general the negative relationship between seed size and oil content was pronounced when the seed size was much larger than the conventional materials. The significant negative r values were obtained in populations derived from crosses to Sarson type, Raya type, and to *B. juncea*. From these data it is not possible to show whether the seed size effect is influencing oil content directly, or if there are linkage blocks associated with multiple loci associated with increased seed size which are causing the lowering of oil content, however in view of Olsson's (1960) work which did not use "exotic" crosses, a pleiotropic effect seems to be the most probable explanation.

Elliott, R.H. 1991. Influence of seed size on emergence, seedling vigour and tolerance of canola to flea beetles. Report to agronomy workshop July 7th at the 8th International Rapeseed Congress, Saskatoon, July 1991.

Olsson, G. 1960. Some relationships between number of seeds per pod, seed size, and oil content and the effects of selection for these characters in Brassica and Sinapis. Hereditas Genetiskt Arkiv. 46: 29-70

Table 1. Correlation between oil content and seed size for breeding populations of *B. rapa*

Population ¹	Number of lines ²	Average seed size (%) ³	r value ⁴
a1	20 (r)	102.9	0.215
a2	6 (r)	107.9	0.214
a3	14 (r)	104.6	0.074
b4	50	115.6	-0.177
b4a	32	131.5	-0.269
b5	37	103.9	0.132
b6	24	118.5	-0.644 ***
b6a	17	143.0	0.048
b7	17	115.9	-0.302
b7a	19	141.2	-0.341
b8	9(r)	107.7	-0.807 **
b9	12(r)	110.6	-0.540 *
c10	83	133.0	-0.194
c11	21(r)	110.8	-0.334
c12	21(r)	111.8	-0.548 **

¹ Populations are lettered as in the text. a1 and a2 were derived from crosses between breeding lines from the University of Alberta and lines from Svalof Weibull company. a3 was derived from crosses between cv. Tobin and lines from Svalof Weibull company. b4 derived from PI 175052 (an Indian Sarson type) x AC Sunshine, b5 from PI 347607 (an Indian type) x AC Sunshine, b6 from PI 352810 (an Indian Sarson type) x AC Sunshine, b7 from AC Sunshine x PI 352817 (an Indian Sarson type), b8 and b9 from AC Sunshine x PI 426240 (a Pakastani Raya type). b4a, b6a, and b7a are derivative populations from b4, b6, and b7 respectively. c10 was derived from an interspecific cross of AC Sunshine to *B. napus*. c11 and c12 were derived from an interspecific cross of AC Sunshine to a very large seeded *B. juncea* accession.

² The number of lines used for the correlation. (r) indicates data from replicated trials, in which case the correlation was calculated using overall line average. In non-replicated trials the check row was included many times.

³ Population average seed size expressed as a percentage of the seed size of check row cv. Parkland.

⁴ Pearson correlation coefficient between seed size and oil content. Significance at the 5%, 1%, and 0.1% levels indicated by *, **, and *** respectively.

A MINOR GENE CONTROLLING ERUCIC ACID LEVELS IN WHITE MUSTARD

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During the course of a breeding program aimed at modifying the erucic acid content of the oil of white mustard [*Sinapis alba* L. (*Brassica hirta* Moench)] genetic studies were undertaken on the erucic acid character. Previously Palafox (1975), and recently Drost *et al* (1999) reported partial dominance for erucic acid content. Our objective was to look for any minor gene action which would be of value in producing extreme levels of erucic acid.

Materials and methods

Three lines of *S. alba* were used as parents in the crossing program. High erucic parents were the cultivar Sabre, (approximately 56% erucic acid in the oil, designated A), and an unrelated line BHC7-1855, (approximately 54% erucic acid in the oil, designated B). The low erucic acid parent was line BHL3-926 (designated C, 2-3% erucic acid in the seed oil). Erucic acid measurement was by gas chromatography of the trans methylated fatty acids (essentially the method of Stringam and McGregor 1980).

From each parent line 31-33 seeds were examined for erucic acid content using the half seed analysis technique of Downey and Harvey (1963), and two to four half seeds with the highest and lowest erucic acid contents from each parent were grown. These are subsequently referred to as A-high, A-low etc. Crosses were made between the A and C lines, and between the B and C lines in all possible combinations of sub groups, i.e. A-high x C-high, A-high x C-low etc. Approximately 10 F₁ half seeds from each cross were examined for erucic acid content and the residual half seeds grown in the greenhouse. Because of the strong self incompatibility system in this species, F₂ seeds were produced by crossing F₁ plants from the same between-class cross but derived from different parental plants. Up to 25 individual F₂ seeds per cross were analysed for erucic acid content.

Results and discussion

Selection within the low parent (C) was highly significant in influencing the erucic acid level of the F₁ seeds in both the A x C and B x C crosses. Selection within either of the high parents A or B did not produce statistically significant differences in the erucic acid level of the F₁ seeds. Thus it appears that a minor gene for erucic acid content may have been detected in the low parent, but if present in the high parents such minor genes were not detectable in the F₁ due to environmental variation or analytical inaccuracies.

The most noticeable characteristic of the F₂ populations was the distribution at the low end of the erucic acid scale (Fig 1). In all four sub-populations derived from crosses involving the low C parent, the low end mode was the 0-2% class, whereas when the high C parent was involved, the low end mode was in a > 2% class. In fact only two observations in the 0-2% class were obtained from any F₂ in which the high C parent had been used. Any effect of selection within the A or B parents was not clearly expressed, and all data from the sub-classes within these parents was pooled. Thus the ability to select for minor gene differences within the low (C) parent can be demonstrated quite readily as an effect on the low end of the F₂ population, but the effects of selection within either of the high (A and B) parents was not detectable. These data, considered with the lack of differences between the F₁ progenies, indicate that the differences within the high parents are probably

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environmental rather than genetic.

Using 16% erucic acid as the dividing point for the total (A x C) F_2 population a ratio of 261:74 for (medium + high):low was obtained, a good fit (Chi square determination $P < 0.3$) to a 3:1 ratio. Using the same dividing point for the (B x C) F_2 population gave a ratio of 274:68, significantly different ($P < 0.05$) from a 3:1 ratio. In both populations there were fewer low erucic acid seeds than predicted.

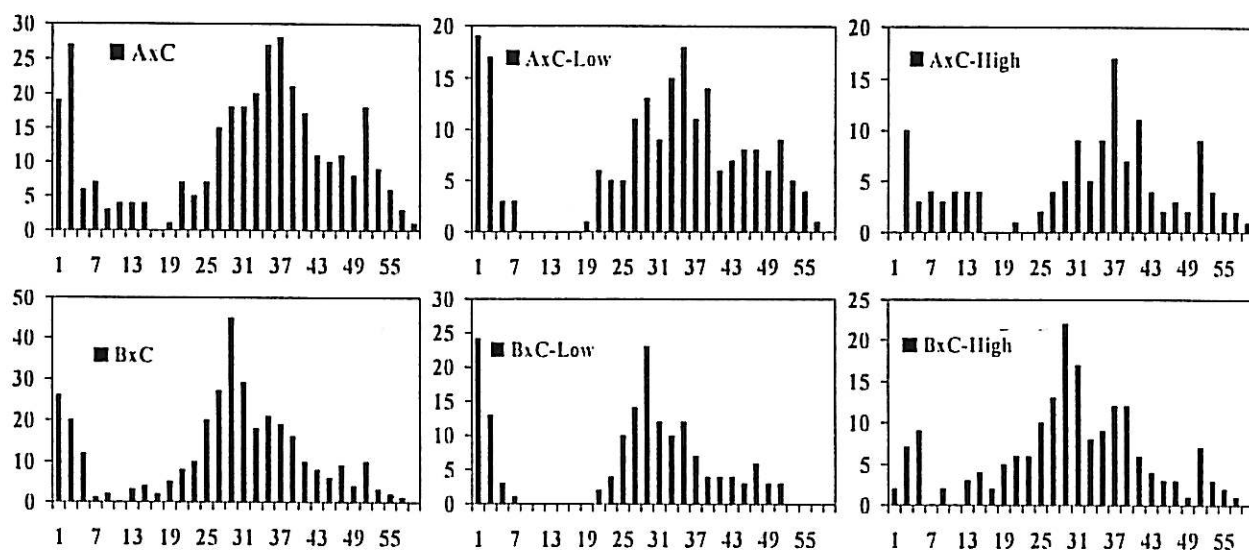


Figure 1. F_2 population distributions; x axis, percentage of erucic acid; y axis, number of seeds

The mode of the low sub-group of the (B x C) F_2 population was the 0-2% class, whereas for the (A x C) F_2 population it was the 2-4% class. In comparison with parent B the effect of parent A seems to be detectable in the F_2 as an increase in erucic acid content in the low end category, and an increase in the proportion of seeds at the extreme high end of the distribution. Overall these data support the presence of a minor gene, non allelic with the major gene contributing approximately 25% erucic acid. The data are consistent with this minor gene being present in parents A and C-high, while absent in parents B and C-low.

In a practical plant breeding program aimed at developing a high erucic acid line it would be desirable to have both major and minor genes combined. Since detection of minor effects in the presence of the major gene is difficult, the technique of crossing lines having levels of 2-3% erucic acid with high erucic acid lines would at least ensure that both genes were present in the breeding population.

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Genetic variation for multiple disease resistance in the families of interspecific cross of *Brassica juncea* x *Brassica carinata*

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Introduction: The three major foliar diseases viz., white rust (*Albugo candida*), *Alternaria* blight (*Alternaria brassicae*) and powdery mildew (*Erysiphe cruciferarum*) are very widespread and destructive on oilseed *Brassica* crops all over India (Saharan, 1997). In the present study, the possibilities to increase the level of multiple disease resistance (MDR) was evaluated in seven families of an interspecific cross, Varuna x PCC-2 of *B. juncea* x *B. carinata* generated from F₂ cross population.

Materials and Methods: The resistant and susceptible plants selected from F₂ population of a cross viz., Varuna x PCC-2 and their seven families viz., R x R, R x S, S x S, R self, S self, R open and S open were generated by intermating, selfing and open pollination. These seven families were screened under artificial inoculation conditions on 0-5 rating scale against white rust, *Alternaria* blight and powdery mildew to find out the genetic variation for disease resistance / susceptibility within and between families. The estimates of mean DSI, range and selection parameters such as genotypic coefficient of variance (GCV), phenotypic coefficient of variance (PCV), heritability (Broad sense) and genetic gain (per cent of mean) for MDR were worked out for each families progenies.

Results and Discussion: The perusal of Table 1 reveals that the within family variations for white rust in cross Varuna x PCC-2 on leaves were significant. The minimum DSI was recorded in progenies of R x R (0.96) which was statistically at par with progenies of R self (1.18) and significantly different from R open (1.34). The maximum DSI was observed in progenies of S open (3.51) which was statistically at par with progenies of S x S and S self families. The 18.64 per cent reduction in disease score was observed in progenies of R x R over R self. The minimum limit of disease score range was observed in R x R family, whereas, it was maximum in S self. The highest GCV (0.210), h² (75.31) and GG (55.21) were recorded in progenies of R x S, R x R and R self, respectively. Whereas, these were non-significant in the progenies of S x S. The progenies of all seven families, except, R x R and S x S showed the significant variation for white rust stagehead phase score. The between family variations for disease score were significant. The lowest DSI was recorded in progenies of S self while maximum in R x R. The maximum GCV (0.012) and GG (12.63) were recorded in R open, whereas, these were minimum in progenies of R x R and S x S families.

Within family variations for *Alternaria* blight infection on leaves were significant in all seven families. The minimum disease score was observed in the progenies of R x R (1.20) which was significantly different from other families. Whereas, the maximum disease score was recorded in progenies of S self (3.33) closely followed by progenies of S x S and S open. The 32.20 per cent reduction in disease score was observed in progenies of R x R over R self. The lowest limit of disease score range was recorded in progenies of R x R while it was maximum in S self. The maximum GCV (0.125), h² (84.43) and GG (37.86) were recorded in R self, whereas, the minimum GCV (0.014) was observed in progenies of R x R. The h² (65.22) and GG (10.91) were minimum in S open family. For *Alternaria* blight infection on siliquae within family variations were significant in all seven families. The lowest disease score was recorded in R x R family (1.30), which was significantly different from other families. Whereas, the maximum DSI was recorded in progenies of S x S (3.83), closely followed by progenies of S self and S open. The per cent reduction in disease score was recorded, 28.96 in R x R family over R self family. The lower range of disease score was observed in progenies of R x R, whereas, it was maximum in S x S family. The maximum GCV (0.096), h² (88.00) and GG (25.50) were recorded in progenies of S x S, S open and R x R, respectively. While minimum GCV (0.030), h² (59.72) and GG (10.35) were recorded in progenies of R open, S x S and S self, respectively.

Significant variations were observed for powdery mildew infection in progenies of each family. The minimum DSI was recorded in progenies of R x R (1.57) which was statistically different from other families. Whereas, maximum disease score was observed in S self family (3.90) which was statistically at par with progenies of S x S and S open. The 12.29 per cent reduction in disease score

was observed in progenies of R x R over R self family. The lower limit of disease score was recorded in R x R family while it was higher in S self family. The maximum GCV (0.217), h^2 (81.34) and GG (35.91) were recorded in progenies of R x S, whereas, these were lowest in progenies of R open.

The low multiple disease score was recorded in the progenies of R x R as compared to R self and R open plant progenies. The R x R cross progenies also recorded narrow variability with lower limits of disease score in all the families. In general, the plant progenies of R x S possessed greater variations with intermediate mean multiple disease score. Interestingly, the progenies of S x S family recorded less disease as compared to S self, indicating that even susceptible plants also possessed resistant gene (s) at different loci with minor effects. The highest h^2 and genetic gain for white rust (leaf phase) was observed in R x R plant progenies. The resistance and susceptibility traits inherited with very high degree from parents to offspring. However, the magnitude of heritability and genetic gain were higher where one or both the parents were resistant for white rust at leaf phase stage. No specific trend was observed for white rust stagehead infection. The genotypic coefficient of variance, phenotypic coefficient of variance, heritability and genetic gain were very low, indicating the narrow variation available for this trait coupled with the greater role of environmental factors in the expression of stagehead.

The disease score for *Alternaria* blight at leaf phase was also low in the progenies of R x R plants as compared to R self and R open families. Further, decrease in disease score of the crossed progenies of R x R indicated that intermating between resistant plants helped in increasing the level of resistance to *Alternaria* blight. In general, the intermediate disease score was observed in the progenies of R x S families. The numerical reduction in *Alternaria* blight (leaf phase) disease score was observed in the progenies of S x S plants cross families in comparison to S self plant progenies. The highest GCV, PCV, h^2 and GG for *Alternaria* blight at leaf phase were observed in R self.

The R x R plant progenies showed comparatively less disease score for powdery mildew infection as compared to resistant self and resistant open plant progenies. The level of susceptibility in progenies of S x S families was low as compared to S self families cross. The maximum GCV, PCV, h^2 and GG were observed in progenies of R x S. Based upon the evaluation of different plant progenies of all the seven families, it is concluded that the R x R families plant progenies would further improve the level of multiple disease resistance in oilseed *Brassica*. It may be attributed due to the accumulation of favourable allele(s) of resistance. Similar observations have been made by Lambert and White (1997), Singh, et al. (1986) and Singh and Singh (1989).

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Table 1 : Disease score and selection parameters for white rust, *Alternaria* blight and powdery mildew infections in the progenies of different families of cross Varuna x PCC-2

Families	Disease score			Selection parameter			
	Mean	Range	C.D.	GCV	PCV	h^2	G.G.
White rust (Leaf phase)							
R open	1.34	0.40-2.16	0.49	0.121	0.205	59.02	41.14
R x R	0.96	0.53-1.50	0.26	0.061	0.081	75.31	46.06
R self	1.18	0.27-1.84	0.39	0.138	0.191	72.25	55.21
R x S	2.16	1.36-2.98	0.51	0.210	0.283	74.20	37.70
S self	3.48	2.88-4.14	0.32	0.108	0.155	69.68	16.26
S x S	3.33	3.09-3.50	NS*	-	-	-	-
S open	3.51	2.83-4.05	0.29	0.084	0.114	73.68	14.62
Mean	2.28						
C.D.	0.33						

Contin.....

Families	Mean	Range	C.D.	GCV	PCV	h ²	G.G.
White rust (Stagehead)							
R open	0.54 (1.24)	0.12-0.92 (1.06-1.39)	0.19	0.012	0.025	48.00	12.63
R x R	0.70 (1.30)	0.56-0.81 (1.25-1.35)	NS*	-	-	-	-
R self	0.50 (1.22)	0.24-0.72 (1.11-1.31)	0.18	0.010	0.022	45.45	11.39
R x S	0.50 (1.22)	0.32-0.68 (1.15-1.30)	0.18	0.009	0.018	50.00	11.34
S self	0.36 (1.17)	0.16-0.72 (1.08-1.31)	0.16	0.006	0.015	40.00	8.64
S x S	0.46 (1.21)	0.28-0.68 (1.13-1.30)	NS*	-	-	-	-
S open	0.50 (1.22)	0.28-0.75 (1.13-1.32)	0.16	0.007	0.016	43.75	9.36
Mean	0.51 (1.23)						
C.D.	0.12						
Alternaria blight (leaf phase)							
R open	1.81	1.42-2.46	0.29	0.090	0.118	76.28	29.87
R x R	1.20	1.11-1.35	0.15	0.014	0.020	70.00	17.02
R self	1.77	1.33-2.40	0.26	0.125	0.148	84.43	37.86
R x S	2.40	2.00-2.77	0.39	0.101	0.143	70.77	23.00
S self	3.33	2.81-3.98	0.32	0.108	0.142	76.26	17.80
S x S	3.31	3.00-3.47	0.25	0.048	0.066	72.72	11.64
S open	3.24	3.00-3.57	0.26	0.045	0.069	65.22	10.91
Mean	2.44						
C.D.	0.47						
Alternaria blight (Siliquae phase)							
R open	1.82	1.50-2.00	0.16	0.030	0.039	76.92	17.22
R x R	1.30	1.18-1.47	0.24	0.037	0.053	69.81	25.50
R self	1.83	1.61-2.28	0.27	0.048	0.067	71.64	20.90
R x S	2.78	2.50-3.14	0.32	0.063	0.093	67.74	15.33
S self	3.79	3.55-4.07	0.18	0.045	0.056	80.35	10.35
S x S	3.83	3.33-4.23	0.48	0.096	0.160	59.72	12.87
S open	3.68	3.38-4.00	0.16	0.066	0.075	88.00	13.51
Mean	2.72						
C.D.	0.35						
Powdery mildew							
R open	1.89	1.62-2.16	0.32	0.018	0.053	33.29	8.36
R x R	1.57	1.16-1.92	0.33	0.066	0.096	68.72	27.98
R self	1.79	1.61-2.31	0.36	0.047	0.092	50.97	17.82
R x S	2.41	1.93-3.20	0.42	0.217	0.266	81.34	35.91
S self	3.90	3.52-4.35	0.28	0.084	0.110	76.28	13.38
S x S	3.85	3.37-4.26	0.38	0.088	0.128	68.32	13.09
S open	3.87	3.45-4.25	0.38	0.051	0.100	50.75	8.55
Mean	2.76						
C.D.	0.20						

R = Resistant ;

S = Susceptible;

GCV = Genotypic coefficient of variance;

PCV = Phenotypic coefficient of variance ;

h² = Heritability (broad sense);

GG = Genetic gain (per cent of mean);

Figures in parenthesis are transformed values; *NS, therefor no need to calculate selection parameters.



BIOLOGY OF MUSTARD APHID, *LIPAPHIS ERYSIMI* (KALT.) ON SELECTED *BRASSICA* GERMPLASM

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INTRODUCTION

Brassica oilseed crops are attacked by a number of insect-pests, of which mustard aphid, *Lipaphis erysimi* (Kalt.) is the most serious pest (Rai, 1976). The influence of genotypes of *Brassica* on biology of mustard aphid was earlier reported (Sachan and Bansal, 1975; Kalra *et al.*, 1987). The utilization of biological studies in relation to host-plant resistance to insect-pest is the most reliable technique and provides uniform and reproducible inferences (Rohilla *et al.*, 1993). The efforts were made to evaluate resistance to mustard aphid by observing biology on selected *Brassica* germplasm under laboratory condition at temperature (Max $18.0 \pm 1.5^{\circ}\text{C}$, Min $15.0 \pm 2.5^{\circ}\text{C}$) and relative humidity $69.0 \pm 4.5\%$.

MATERIALS AND METHODS

The biology of mustard aphid was studied on the leaves of 8 selected *Brassica* germplasm. Plants of these germplasm were raised in pots in glasshouse to get leaves at the time of requirement. The leaves of selected germplasm were placed in petridishes (20 cm diameter) containing moist filter paper at the bottom. Leaves were changed daily and aphid transferred to new leaves with the help of a fine camel hair brush. The newly emerged nymph was kept on each leaf and observed for its moulting at every 24 hours interval. Moultings were observed with the help of exuviae cast off by the nymphs with the help of Binocular. The adult longevity was determined on the basis of adult survival from emergence to the death of adult aphid. The natality of mustard aphid was recorded by counting the total number of nymphs emerged from a single adult mustard aphid in duration of reproductive period. The rate of nymphal emergence per day was calculated by dividing total number of emerged aphids to the days of nymphal emergence. The experiment was carried out in Completely Randomized Design in five replications.

RESULTS AND DISCUSSION

Mustard aphid completed four nymphal instars on all 8 selected germplasm. The nymphal development completed in shorter duration on susceptible check, BSH-1 than the other test germplasm. The nymphal period of mustard aphid varied from 7.7 to 9.9 days (Table 1) observing minimum (7.7 days) on BSH-1 and maximum (9.9 days) on RW-2-2, which were found significantly different. The maximum reproductive period of mustard aphid was found on the susceptible check, BSH-1 (18.2 days) which differed significantly from the reproductive period observed on promising germplasm. Significantly minimum (13.6 days) reproductive period was found on RW-2-2, which did not differ significantly from the reproductive period recorded on RLM-198, DIRA-337 and RH-7846. The post-reproductive period of mustard aphid also varied significantly. Significantly shorter adult longevity was recorded on *Brassica* germplasm RLM-198 and RW-2-2 and these two germplasm did not differ significantly from DIRA-337 and RH-7846 but differed significantly from BIO-902 and the susceptible BSH-1. Significantly longest total life span (28.1 days) was recorded on the susceptible BSH-1. Total life span of mustard aphid was significantly shorter on promising germplasm than the susceptible check, BSH-1.

The natality per female mustard aphid differed significantly among test germplasm recording maximum number (114.0) on the susceptible BSH-1, which differed significantly from all 7 promising germplasm. Significantly lowest (52.0) natality per female was observed on DIRA-337, which did not differ significantly from the natality per female recorded on RW-2-2 and RLM-198. Natality per female per day on test *Brassica* germplasm varied from 3.7 to 6.3 showing significant differences among test germplasm. Significantly highest (6.3) natality was observed on the susceptible BSH-1, which was found very near with the result of Agarwal *et al.* 1996.

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Table 1. Nymphal period, reproductive period, adult longevity and natality of mustard aphid on selected *Brassica* germplasm

<i>Brassica</i> germplasm	Nymphal period (days)				Total nymphal period (days)	Pre-reproductive period (days)	Reproductive period (days)	Post-reproductive period (days)	Adult longevity (days)	Total life span (days)	Natality per female (No.)	Natality per female per day (No)
	I	II	III	IV								
BIO-902	2.1	2.2	2.0	2.0	8.3	1.0	16.8	0.6	18.4	26.7	73.4	4.4
DIRA-337	2.6	2.5	2.2	2.2	9.5	1.0	14.2	1.0	16.2	25.7	52.0	3.7
DLSC-2	2.2	2.4	2.1	2.2	8.9	1.0	15.2	1.0	17.2	26.1	62.0	4.1
RH-7846	2.5	2.6	2.2	2.1	9.4	1.0	14.4	1.0	16.4	25.8	60.6	4.2
RLM-198	2.5	2.6	2.3	2.3	9.7	1.0	14.0	1.0	16.0	25.7	55.4	4.0
RSK-84	2.3	2.2	2.0	2.1	8.6	1.0	15.6	1.0	17.6	26.2	73.6	4.7
RW-2-2	2.8	2.7	2.2	2.2	9.9	1.0	13.6	1.4	16.0	25.9	52.8	3.9
BSH-1 (S-Check)	2.0	2.0	2.0	1.7	7.7	1.0	18.2	1.2	20.4	28.1	114.0	6.3
CD at 5%	0.4	0.3	0.3	0.3	0.3	-	0.8	0.5	0.7	0.7	5.7	0.7

Isolate-specific resistance to clubroot in *Brassica napus* is expressed at high or partial level

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Introduction

Clubroot, caused by the obligate biotroph *Plasmodiophora brassicae*, is a damaging disease of *Brassica napus* in several areas of production. As cultural practices or chemical treatments are not efficient or too expensive, the development of resistant cultivars is considered as a need to control this disease for all *Brassica* species. Information about either host-genotype interaction or the genetic basis of the resistance remains however relatively limited in the subspecies *B. napus oleifera*, and comparison among published studies is difficult. Notably, the interpretation of results is often limited by non-homogeneous reactions owing to the use of field isolates of the pathogen (heterogeneous resting spore populations) in the tests, making difficult to define the race-specificity of the resistance genes. The use of homogeneous pathogen isolates (single spore derived isolates, SSI) can simplify and facilitate the understanding of the genetic mechanisms involved in this host-pathogen interaction. The objective of this study was to detect and to evaluate the resistance level of rapeseed genotypes using homogeneous *P. brassicae* isolates. This preliminary work is a part of a research aiming at the characterisation, identification and location in *B. napus* of loci controlling clubroot resistance that are expressed against different isolates of *P. brassicae*.

Material and methods

The rapeseed genotypes (*B. napus* var. *oleifera*) screened were: 'Darmor', 'Darmor-bzh', 'Samourai', 'Maxol', 'S006' (winter type) and 'Yudal', 'Stellar', 'Drakkar', 'Brutor' (spring type). These genotypes are used at INRA-Rennes as parents for the establishment of genetic maps in order to study QTLs associated with different agronomic traits.

Two field isolates and five SSIs of *P. brassicae* were used in the resistance tests (Table 1). The field isolates, K92 and BN98, come respectively from naturally infested cauliflower and rapeseed (cv. 'Navajo') crops in France. The SSIs Ms6, K92-16, Pb137-522 and SJ92-256 were derived from 4 field populations isolated from cauliflower clubs as described previously (Somé et al., 1996). The SSI eH was kindly provided by Dr. J. Siemens (University of Berlin). All isolates were maintained on the susceptible Chinese cabbage (*Brassica rapa* spp. *pekinensis* cv. Granaat); clubs were harvested, washed and stored at -20 °C. Isolates were characterised for pathogenicity on three differential *B. napus* cultivars as described previously (Somé et al., 1996).

Resistance tests were done in the glasshouse. Each line was tested in a randomised complete block design with 2 replicates and 20 plants per replicate. Inoculation was performed by applying 1 ml of a resting spore suspension (10^7 spores/ml) at the bottom of the stem base of each seedling at the 6-8 day stage. Eight weeks after inoculation, plants were rated for disease reaction based on a 0 to 3 severity scale and a disease index (DI) was calculated. The DI varied from 0 (no gall) to 100 (all plants at grade 3).

Results and discussion

The reaction of the nine rapeseed lines tested against the seven isolates of *P. brassicae* are summarised in Table 1. Tests were repeated twice at three months interval. As neither block nor experiment effect were significant ($p > 0.05$), data shown are means of the two replicates and the two experiments. In the ANOVA analysis, there were significant differences between lines, isolates and line*isolate interaction ($p < 0.0001$). The isolate BN98 was highly pathogenic to all lines tested; rapeseed genotypes were severely attacked, and the symptoms appeared quickly. The lines 'Yudal'

and 'Maxol' were very susceptible to all isolates. No line was resistant to the seven isolates and the resistance reaction to six isolates varied among lines. These results indicated that in the *B. napus*-*P. brassicae* model analysed, host-pathogen interaction was mainly differential. The relative effectiveness of the putative resistance genes carried out by 'Darmor', 'Darmor-bzh', 'S006', 'Stellar', 'Drakkar' and 'Brutor' may vary when the challenging population of the pathogen is changed. According to the isolate, the resistance expression varied from high to intermediate levels in a same host genotype. For example, 'Darmor-bzh' expressed a high level of resistance reaction to 3 isolates (DI < 25), a partial level of resistance to 2 isolates (DI 40-50) and a very susceptible reaction to the other 2 isolates. The finding of a partial resistance does not appear to fit easily into any of the schemes suggested by Crute et al. (1983) or Gustafsson and Fält (1986) for qualitative resistance in *B. napus*. Work is in progress to study the genetic basis of the resistance factors present in 'Darmor-bzh' and to analyse the relations between loci associated with partial and complete resistance.

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Table 1. Disease Index of the nine rapeseed genotypes inoculated with seven isolates of *Plasmodiophora brassicae*. The DI values correspond to the mean of two experiments.

Host Genotype	Pathogen isolate						
	Field isolate		Single spore isolate				
	K92 (P5) ^a	BN98 (P5)	Ms6 (P1)	eH (P1)	K92-16 (P4)	SJ92-256 (P4)	Pb137-522 (P7)
Darmor- <i>bzh</i>	20.9	100.0	92.5	40.0	50.0	22.5	13.0
Darmor	6.3	100.0	100.0	45.0	-	-	0.0
Samourai	100.0	100.0	100.0	50.0	100.0	-	100.0
Maxol	100.0	100.0	100.0	100.0	100.0	-	83.8
S006	21.6	100.0	74.4	33.8	23.0	-	13.4
Yudal	100.0	100.0	95.0	93.8	100.0	80.0	95.0
Stellar	22.5	100.0	48.8	100.0	18.8	-	13.8
Drakkar	2.5	100.0	100.0	95.0	25.3	-	1.3
Brutor	44.2	100.0	96.3	100.0	10.6	20.0	1.3
Control ^b	100.0	100.0	100.0	100.0	100.0	100.0	100.0

^a Pathotypes are indicated in parenthesis

^b Susceptible control: Chinese cabbage
- not tested

EFFECT OF GAMMA RAYS AND EMS ON INCIDENCE OF ALTERNARIA BLIGHT AND WHITE RUST IN M₂ GENERATION OF INDIAN MUSTARD (*Brassica juncea* L. CZERN & COSS.)

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Introduction

Indian mustard is the most important oilseed *Brassica* crop in India. The reduction in yield from 10 to 70% and 17 to 34% have been reported due to attack of Alternaria blight and white rust, respectively (AICORPO, 1981; Bains and Jhooty, 1979; Kolte, 1985). All the commercial varieties of Indian mustard are susceptible to these diseases. Transfer of resistance to Alternaria blight from *Camelina sativa* and *Capsella bursa-pastoris* has proved difficult. Mutation breeding has been initiated to create variation and search for resistant sources to these diseases.

Materials and Methods

Dry seeds of two varieties viz., Narendra rai and NDYR10 were treated with 25,50,75,100 and 125 kR of gamma rays and 0.25,0.50,0.75 and 1.0% concentration of Ethyl Methane Sulfonate and were grown to raise M₁ generation. The M₂ population was grown during 1997-98. The Incidence of Alternaria blight on leaves and siliqua and white rust on leaves was scored using 0-5 scale (0 = disease free, 1 = 1-10% leaf area affected by disease, 2 = 11-25% area affected, 3 = 26-50% area affected, 4 = 51-75% area affected, 5 = more than 75% area affected). Per cent disease intensity (PDI) was worked out.

Results and Discussion

All the plants in each treatment showed symptoms of Alternaria blight, however, disease intensity differed (Table 1). In Narendra rai, minimum disease intensity was observed at 75 kR on leaves and at 1% EMS on siliquae. At 50 and 125kR, the disease intensity on leaves was higher than control. Significant increase was observed at 0.25% EMS.

In NDYR 10, all the treatments showed higher disease intensity for alternaria blight as compared to control. However, significant increase was observed at 50kR and 0.50 and 0.75% EMS.

All treatments showed presence of susceptible reaction to white rust but the disease intensity differed. In Narendra rai, 0.25% EMS increased intensity of white rust incidence with highly significant difference as compared to control. Maximum variation was found at 75kR dose of gamma rays. 25kR dose also increased intensity of disease with significant differences. The 50 and 75kR doses of gamma rays and 0.50 and 1% concentration of EMS showed lower disease intensity.

In NDYR 10, incidence of white rust was found low in all the treatments as compared to control. Highly significant reduction in disease intensity was observed at 50 and 75kR of gamma rays (31.87 and 33.34% disease intensity, respectively) and at 1% EMS (32.55%) in comparison to control (36.25%). Significant disease reduction in NDYR 10 was observed at 25,100 and 125kR doses of gamma rays and at 0.25% EMS. Similar were the findings of Das and Rahman (1988), Verma and Rai (1980) and Rai (1983). It is concluded that mutagenesis was effective in creating variation for Alternaria blight and white rust. A number of mutants showing less incidence to Alternaria blight and white rust have been selected.

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Table 1. Range and mean for Alternaria blight (PDI) on leaves and siliquae in M₂ generation of Indian mustard

S.N.	Treatments	Alternaria blight (PDI)			
		On leaves		On siliquae	
		Range	Mean±SE	Range	Mean±SE
1	Narendra Rai (Control)	42.07- 48.60	45.86±0.86	16.21- 29.81	22.72±0.64
2	25 kR	42.07-49.52	45.34±0.74	12.75-26.95	19.04±0.72
3	50 kR	43.15-51.36	47.06±0.59	9.32-20.34	15.17**±0.69
4	75 kR	38.63-47.81	44.38±0.38	6.28-19.57	14.43**±0.58
5	100kR	41.10-49.61	45.36±0.74	11.35-21.35	17.13±0.61
6	125 kR	43.07-49.35	46.09±0.74	11.43-22.85	16.99*±0.92
7	0.25% EMS	45.35-53.75	49.09*±0.79	8.27-20.82	17.14±1.34
8	0.50% EMS	44.12-52.36	48.29±0.69	12.23-27.71	20.89±0.85
9	0.75% EMS	45.32-50.67	47.98±0.51	7.75-16.41	11.04**±0.60
10	1.00% EMS	42.74 -46.51	44.96±0.56	5.12-17.63	10.59**± 0.64
11	NDYR 10 (Control)	43.62-48.12	45.87±0.47	11.31-23.81	17.86±0.61
12	25kR	44.18-52.81	48.39±0.53	15.12-29.10	20.08±0.73
13	50kR	45.21-52.15	49.06*±0.52	14.26-26.12	19.77±0.52
14	75 kR	44.03-50.75	48.15±0.55	16.23-25.16	20.68±0.70
15	100kR	44.35-52.15	48.66±0.46	16.03-26.16	22.65±0.82
16	125kR	43.81-50.02	46.50±0.42	19.63-33.61	27.27**±0.81
17	0.25% EMS	42.17-50.82	47.63±0.73	22.93-30.62	27.85**±1.03
18	0.50% EMS	46.56-52.63	48.92±0.48	19.42-28.75	24.26*±0.82
19	0.75% EMS	45.73-52.81	49.39±0.65	18.81-33.21	24.84**±0.72
20	1.00% EMS	45.33-52.53	48.68±0.64	15.57-18.57	15.23±0.87
	CDat 5%		2.88		4.97
	CD at 1%		3.81		6.62

*, ** Significant at 1% and 5% level

Losses induced by *Alternaria* blight in yield and oil of rape and mustard in Pakistan

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Introduction:

Alternaria Blight (AB) caused by *Alternaria brassicae* (Brek.) Sacc. is a diminutive disease of rape and mustard in many parts of the world and can reduce production upto 70%. Under Pakistani conditions, AB is commonly observed on rape and mustard (Altaf *et al.*, 1988, NODP 1993 and Khan and Munir 1985) and is suspected to be a major factor responsible for significant losses of these crops in the country. The estimation of losses by AB were, however, never made. Therefore a study was initiated and results of the 1st year (1998-99) are summarised herewith. These results appear to be the 1st report on losses caused by AB in yield and oil content of rape and mustard in Pakistan.

Materials and Methods:

Eight genotypes of rape and mustard were used in this study. The experiment was conducted during rabi 1998-99 at NIFA in a Randomised Complete Block design with strip plot arrangement having three replication with plot size of 1.2 x 3m, row to row and plant to plant distances of 30 and 10 cm, respectively. Two irrigations and 60:30:30 kg of N: P: K were applied during the growing season. Normal cultural practices were carried out. There were two strips in each replication, one was protected by four sprays of the fungicide Mancozeb (@ 3g/litter) at 15 days interval starting from 15th of February while in the other, natural infection of the pathogen was allowed to develop blight. AB severity was recorded in unprotected plots in each replication on 10 bottom leaves in 5 randomly selected plants at seed formation stage according to 0-5 scale (Rai *et al.*, 1977). After harvesting the plots at maturity, yield and oil content were estimated for both protected and unprotected treatments in each replication. The mean yield and oil content were determined and percent loss for each genotype was calculated using the differences in yield and oil content of protected and unprotected treatments.

Results and discussion:

Results (Table-1) of yield and oil content under protected conditions were higher in all the genotypes of rape and mustard studied. Variation in genotype response towards AB attack and yield was observed. Within rapeseed, PR-7 showed highest yield under unprotected conditions with least AB severity, which resulted in the lowest losses in yield and oil content. This may probably be due to earliness and greater resistance to AB. Abasin-95 and Dunkled produced grain yield above 1500 and 1700 kg/ha respectively under protected conditions and were tolerant to AB. Despite being tolerant, mean yield and oil loss was 12 and 18% in Abasin-95 and Dunkled, respectively.

Among mustard genotypes, BM-1 was the highest yielder under both protected and unprotected conditions. Yield and oil losses were 24.29% and 17.50% respectively due to lesser tolerance exhibited by it against AB. Genotypes MMJ-1304 and MMJ-1277 have been reported (Shah *et al.*, 1999) susceptible/tolerant to AB. These genotypes exhibited maximum yield losses of 38.50% and 30.66% respectively while losses in oil content were less than 9% in both the genotypes.

This study clearly demonstrated that yield and oil content were negatively affected by the disease irrespective of the species and the genotype involved under Pakistani conditions. Therefore, keeping in

view the huge edible oil shortage in the country, serious measures are needed to minimise the quantitative and qualitative losses incurred to these important oilseed crops by *Alternaria* blight.

Table-1: Seed yield and oil losses in different genotypes of rape and mustard due to *Alternaria* Blight.

NO	Rape/Mustard	Seed Yield (Kg/ha)			Oil Content (%)		
		Unprotected	Protected	% loss	Unprotected	Protected	% loss
1	Rape Abasin-95	1389.0 (10.0)	1583.0	12.25	32.0	36.0	11.11
2	Siran	1361.0 (18.0)	1500.0	9.26	31.0	35.0	11.42
3	PR-7	1472.0 (5.0)	1611.0	8.62	36.0	38.0	5.26
4	Dunkled	1417.0 (12.0)	1722.0	17.71	32.0	39.0	17.94
1	Mustard BM-1	2250.0 (24.0)	2972.0	24.29	33.0	40.0	17.50
2	DLJ-3	528.0 (12.30)	611.0	13.58	30.0	34.0	11.76
3	MMJ-1277	1194.0 (16.0)	1722.0	30.66	31.0	34.0	8.82
4	MMJ-1304	444.0 (39.0)	722.0	38.50	32.0	35.0	8.57

Figures in parenthesis are the mean AB severities.

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Weather factors associated with *Sclerotinia* stem-rot of Indian mustard and development of a linear model for its prediction

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ABSTRACT

Stepwise multiple regression analysis (SMRA) equation between weather parameters as independent factors and severity of *Sclerotinia* stem-rot of Indian mustard as dependent factors were established on the basis of data obtained over 1996-99. *Sclerotinia* stem-rot progression in February had significant positive correlation with mean temperature ($YrX_3=0.6506$) and maximum relative humidity ($YrX_6=0.7407$). Best fitted predictive linear equation developed was $Y= 2.9245 + 0.0755X_3 + 0.0469X_6$, $R^2 = 0.74$

INTRODUCTION

The *Sclerotinia* stem-rot of *Brassica juncea* caused by *Sclerotinia sclerotiorum* (Lib.) De Bary is one of the major soil borne diseases characterized by water soaked lesion, which girdle the entire stem followed by cottony mycelial growth and sclerotia on Indian mustard (Singh and Tripathi, 1994). The basic information in respect of epidemiological factors associated with occurrence and severity of this disease has not been documented. Therefore, the present investigation were undertaken to correlate the weather components with severity of the disease and to develop a linear model for its prediction.

MATERIALS AND METHODS

Experiment was conducted to determine the disease progression in relation to environmental factors. Sowing was done on 15 October in triplicate in 2 x 2m plots. Plants were inoculated at collar region with sclerotia as well as mycelial discs on 20 January. Disease progression was measured regularly at the interval of 48 hr upto 30 days, after that plant expressed rotting symptom. Data on disease progression were recorded with the help of scale proposed by Lesvoi *et al* (1981) with some modification, where 0 = no lesion, 1 = 1-5 cm lesion, 2 = 5-15 cm lesion, 3 = 16-25 cm lesion, 4 = 26-35 cm and 5 = >35 cm lesion on stem.

Rotting index and disease intensities were computed as :

$$\text{Rotting Index} = \frac{\text{Sum of products (rating class x no of stem in that class)}}{\text{Total no of stem x maximum rating}}$$

$$\text{Disease Intensity (\%)} = \frac{\text{Sum of products of all numerical rating}}{\text{Total no of stem examined x maximum rating}} \times 100$$

Progress of *Sclerotinia* stem-rot was plotted on graph by taking time period on the X-axis and $\text{Log}_e (1/1-X)$ transformed value and percent disease intensity on Y-axis.

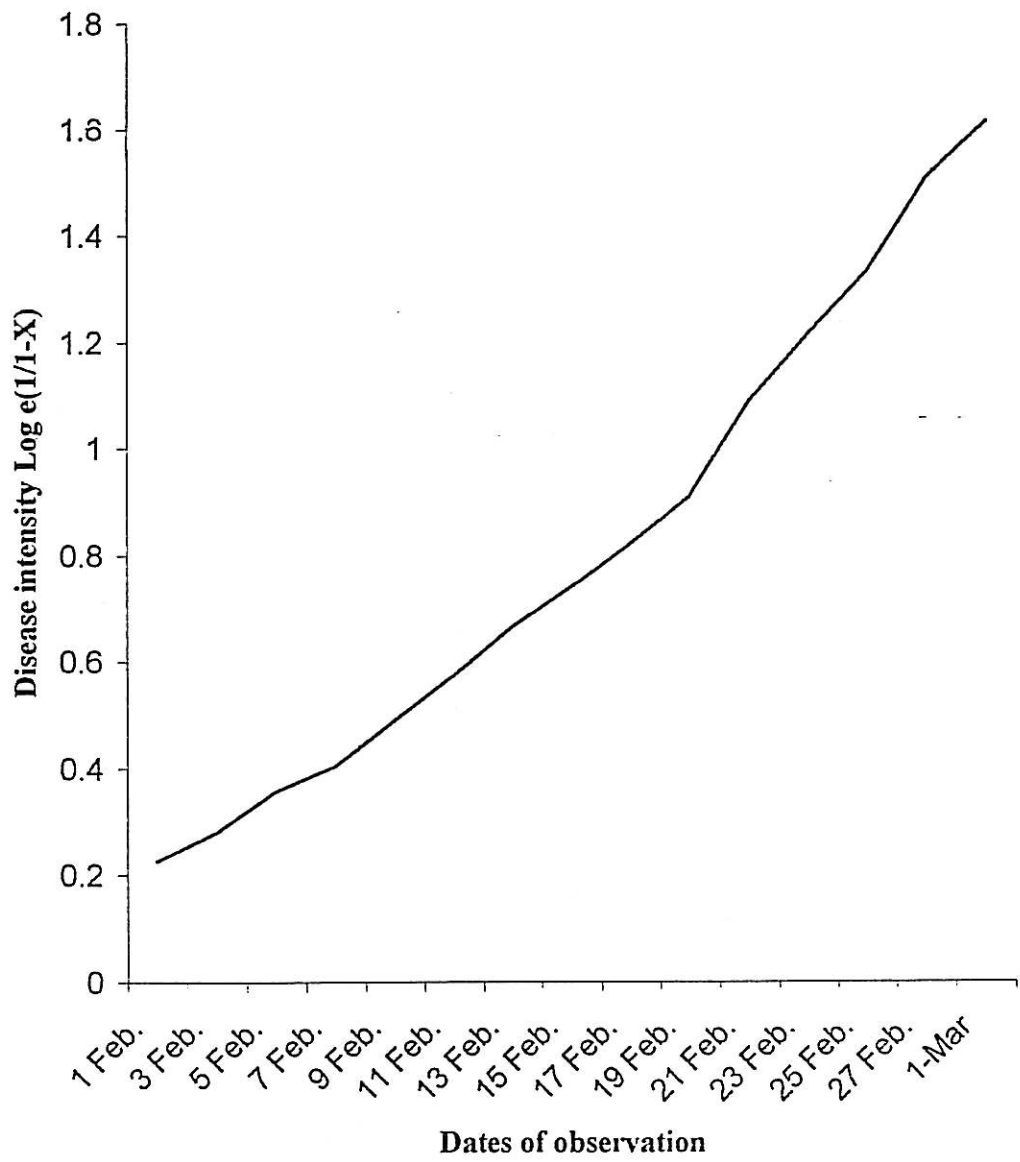


Figure 1. Sclerotinia stem-rot progression

INTERACTIVE EFFECT OF SULPHUR AND NITROGEN ON SOME PHYSIOLOGICAL PARAMETERS OF MUSTARD (*Brassica juncea* L. Czern. and Coss.)

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One of the most important constraints for crop growth is shortage of nutrients. Sulphur and nitrogen are closely related, synergistic and of vital importance to plants because sulphur is part of major constituents of aminoacids. Sulphur constitute the building block of proteins (Ceccotti, 1996). A strong interaction between sulphur and nitrogen has been reported by many workers (Zhao *et al.* 1993b., Mc Grath and Zhao, 1996). The present investigation is aimed to study the interaction between sulphur and nitrogen on some of the physiological parameters of mustard (*Brassica juncea* L. Czern. and Coss. Cv. Pusa Jai Kisan).

MATERIALS AND METHODS

A field experiment (Randomized block design) was conducted to study the interactive effect of sulphur and nitrogen on some of the physiological parameters of mustard (*Brassica juncea* L. Czern. and Coss. Cv. Pusa Jai Kisan). Mustard was grown on sandy loam soil at the experimental plots of Hamdard University, New Delhi, India. The treatments included two levels of sulphur (40 and 60 Kg ha⁻¹) and nitrogen (100 and 150 Kg ha⁻¹) in the following combinations viz. S₀+N₁₀₀ (T₁), S₄₀+N₁₀₀ (T₂), S₆₀+N₁₀₀ (T₃) and S₆₀+N₁₅₀ (T₄). Both sulphur and nitrogen were applied in two splits. The source of sulphur and nitrogen were gypsum and urea, respectively. Phosphorus and potassium were applied at the rate of 40 Kg ha⁻¹. Three replications were made of each treatment. The plot size was 16m² (4m×4m) with 10 rows and a row to row distance of 30 cm. Chlorophyll content of the leaves was determined by the method of Arnon (1949). Photosynthetic rate of the intact leaves was determined by using a portable photosynthetic system (Model Li 6200, Li-COR. Inc; USA). Soluble protein content of the leaves was determined by the method of Bradford (1976) using bovine serum albumin as standard. These parameters were measured on 45 days after sowing.

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RESULTS AND DISCUSSION

The development of yield components depend on the growth attributes of the plant and proper growth can be ensured through efficient manipulation of nutrients. Combined application of sulphur and nitrogen enhanced the parameters studied (Table 1). Chlorophyll content, rate of photosynthesis and soluble protein content increased by 168.8, 41.7 and 81.3% at T₃ (S₆₀+N₁₀₀) followed by 87.5, 17.4 and 37.5% increase at T₂ (S₄₀+N₁₀₀). Increase in the above parameters were also observed at T₄ (S₆₀+N₁₅₀) but not to the extent observed at T₃ and T₂. These results suggest that a proper balance of sulphur and nitrogen fertilizer is needed to ensure better growth of mustard.

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Table 1. Interactive effect of sulphur and nitrogen on some of the physiological parameters of mustard.

Treatment	Chlorophyll (mg g ⁻¹ F.W)	Soluble protein (mg g ⁻¹ F.W)	Rate of photosynthesis (μ mol CO ₂ m ⁻² s ⁻¹)
T ₁	1.12	3.2	20.6
T ₂	2.10	4.4	24.2
T ₃	3.01	5.8	29.2
T ₄	1.65	3.8	22.8

F.W -Fresh weight

MUTANT HETEROSIS IN OILSEED RAPE

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Introduction:

Many researchers have demonstrated expression of heterosis in F₁ generation of mutant crosses in a number of plant species. The heterosis can appear in crosses between mutants from the same initial parent variety. Heterosis for many characteristics (e.g. growth, quality) may be important in crop development and use, but heterosis in yield and yield components is the most important. This paper describes the use of mutants with the same genetic background as cross-components for significant mutant yield heterosis in oilseed rape (*Brassica napus* L.).

Materials and Methods:

Three stable mutants (M₉ generation) viz. RM-182, RM-159-2 and RM-152-2 and their initial variety Tower were used to investigate heterosis in F₁ of inter-mutant crosses in oilseed rape (*Brassica napus* L.). In Rabi 1997-98, hand-pollinated crosses were made and matured F₀ seeds were harvested cross-wise. In Rabi 1998-99, seeds were planted in cross to row as F₁ generation. Agronomic data regarding the number of primary branches, number of primary roots, number of pods, number of seeds per pod, total oil content and yield were recorded individually per plant at maturity.

Results and Discussion:

F₁ generation expressed a significant heterosis in about all the cross combinations regarding number of primary branches, number and length of primary roots, number and length of pods, seeds / pod, yield / plant and total oil content / plant. The maximum significant increase of 45.0 and 47.6 % for yield per plant was recorded in cross RM-152-

2 x RM-182 over the mutant parent and initial variety respectively (Table). All crosses attained more height than the parent plants and initial variety. Two crosses matured significantly earlier (15-18 days) than the parents. A significant manifestation of heterosis in the number of primary branches, number of roots was observed in all the crosses (Table). Cross RM-152-2 x RM-182 significantly outclassed the better mutant parent with 54.4 % and initial variety with 64.2 % increased pods / plant (Table). In respect of seed / pod, F₁ mutant crosses expressed significant heterotic performance with an increase in seeds / pod ranging from 4.0 - 17.9 % and in seed weight from 2.5 - 14.6 %. In case of total oil content only the cross RM-152-2 x RM-182 expressed significantly F₁ heterotic performance (Table). This initial breeding material will be a rich source of valuable basic gene pool for the future genetic improvement programme of oilseed brassica at NIFA.

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Table - Heterosis manifestations (% increase over the better parent (a) or original variety (b) in F₁ of selected Tower mutant crosses (*B. napus*).

Crosses	Branches per plant	Roots per plant	Pods per plant	Seeds per pod	Oil %	Yield g per plant
RM-152-2 X RM-182	a- 38.5* b- 46.2*	a- 53.2* b- 53.2*	a- 54.4* b- 64.2*	a- 7.7* b- 11.5*	a- 1.5* b- 6.4*	a- 45.0* b- 47.6*
RM-182 X RM-152-2	a- 20.0* b- 20.0*	a- 4.2* b- 8.3*	a- 28.0* b- 35.5*	a- 8.0* b- 4.0*	a- 1.0 b- 6.3	a- 18.4 b- 14.4
RM-152-2 X RM-159-2	a- 30.0* b- 30.0*	a- 21.7* b- 13.1*	a- 23.6 b- 37.5*	a- 17.9* b- 17.9*	a- 0.7 b- 7.8	a- 12.6 b- 14.4

* / p < 0.05

EFFECT OF FOLIAR APPLICATION OF AGRO-CHEMICALS ON SEED YIELD AND OIL QUALITY IN RAPESEED-MUSTARD

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INTRODUCTION

Earlier studies have shown that agro-chemicals like sulphuric acid caused physiological changes on the sprouting of wheat seedlings (Yu and Guo, 1993). Foliar sprays of thiourea at tillering and at flowering @ 1 kg/ha improved productivity in wheat (Sahu and Singh, 1995). In Indian mustard foliar sprays of sulphuric acid and thiourea influenced physiological traits and the yield (Khafi *et al.*, 1997). Keeping in view limited information and the beneficial effects of agrochemicals on these crops, this study was carried out to assess the effect of foliar application of sulphuric acid, thiourea and urea on yield, oil content and its quality in rapeseed-mustard.

MATERIAL AND METHODS

A field experiment was conducted by taking cv. GSL1 of *B.napus* and cv. PBR-91 of *B.juncea*, in RBD with three replications. There were five treatments i.e. control, water spray, thiourea (0.1%), sulphuric acid (0.1%) and urea (1%). The trial was conducted in the experimental area of Plant Breeding Department, Punjab Agricultural University, Ludhiana during 1998-99, following the recommended package of practices. Two foliar sprays of sulphuric acid and thiourea at 0.1% and urea at 1% concentration were done at 45 days and 60 days after sowing. Water sprays (800 litres/ha) were done in all control plots. Dried seed samples were analysed for oil content and fatty acid composition as reported earlier (Ahuja *et al.*, 1998).

RESULTS AND DISCUSSION

The highest seed yield was obtained with thiourea (0.1%) in both the species i.e. 1478 kg/ha in Indian mustard (*B.juncea*) and 1775 kg/ha in *gobhi sarson* (*B.napus*). These yields were higher by 5.1% and 5.8% over control respectively. The seed yield was also numerically higher with urea (1%) and sulphuric acid (0.1%) as compared to control. However, seed yield with various treatments were statistically non-significant in both the Brassica species (Table 1). A similar pattern was noticed by Khafi *et al.* (1997) who reported that two foliar sprays of sulphuric acid and thiourea at 0.1% concentration significantly increased yield parameters and yield of Indian mustard. Oil content was more or less similar with various treatments on both the species. However, sulphuric acid spray enhanced the oil content marginally. In general GSL-1 had 4% higher oil content than PBR-91. In GSL-1, thiourea and urea sprays decreased the erucic acid content by 2-4% as compared to control. Sulphuric acid treatment increased erucic acid content by 3-4%. In *B.juncea* cv. PBR 91 however, no definite trend in fatty acid make up with various sprays was observed. This may be due to differential response of different Brassica species to agro-chemicals.

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Table 1: Effect of foliar application of agro-chemicals on seed yield, oil content and fatty acid composition in rapeseed-mustard

Genotype	Treatment (spray)	Seed yield (kg/ha)	Oil content (%)	Fatty acid(%)						
				16:0	18:0	18:1	18:2	18:3	20:1	22:1
GSL-1	Control	1677	38.5	3.72	0.49	19.53	14.65	8.38	12.12	41.20
	Water spray	1682	38.7	3.59	0.58	20.40	14.26	8.76	10.54	41.80
	Thiourea(0.1%)	1775	38.4	3.44	0.66	20.88	14.30	8.42	13.20	39.12
	Sulphuric acid(0.1%)	1707	39.2	3.54	0.55	17.50	13.58	7.52	12.49	44.83
FBR 91	Urea (1%)	1728	38.7	3.56	0.65	22.64	15.87	8.28	11.38	37.62
	Control	1406	34.4	2.78	0.80	13.85	17.90	10.56	6.74	47.38
	Water Spray	1404	34.2	2.24	0.73	12.35	17.48	10.59	8.61	48.53
	Thiourea(0.1%)	1478	34.8	2.26	0.85	12.25	16.35	11.00	6.96	50.35
	Sulphuric acid(0.1%)	1417	35.0	2.53	0.66	12.75	17.04	11.12	7.62	48.28
	Urea(1%)	1450	34.7	2.53	0.71	12.24	16.60	10.10	6.39	51.43

RESPONSE OF ROCKET (*Eruca sativa* Mill.) TO NITROGEN LEVELS

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INTRODUCTION

Rocket is an ancient herbaceous crop primary grown in the Mediterranean region and western Asia, however, its production in the Northern and Eastern Europe is increasing as well. All plant parts over the soil surface are used for fresh consumption as salads or appetizers (Eşiyok, 1996). The interest in the green fresh rocket leaves is increasing and most of the Mediterranean countries have revised and established procedures of cultural practices (Baggio and Pimpini, 1994; Pimpini and Enzo, 1996). The current research has been done as a part of the work started by the authors for the improvement of cultural practices of rocket in Turkey (Eşiyok et al., 1998a, 1998b and 1999).

MATERIAL AND METHOD

Studies were conducted at the experimental farm of the Faculty of Agriculture, University of Ege to investigate the effects of different nitrogen levels on rocket growth. For each treatment a total of 4 g seeds per m² was sown in single rows in 3m² plots. A completely randomized block design with three replications was used to test four nitrogen levels (0, 6, 12, 18 and 24 kg/1000m²) applied in the form of ammonium sulphat prior to sowing. Constant rates of 18 kg K₂O and 12 kg P₂O₅ per decar (1000m²) was applied to the plots for phosphor and kalium needs of the plants. The soil was a clay - loam soil at 7.43 pH with % 2.10 organic matter, % 0.137 total nitrogen and % 0.07 total soluble solid content.

Total yield (g/m²) was determined by weighting all consumable fresh plant parts cut ca. 2 cm above the soil surface. A part of the plant material (ca. 500 g) was dried in the oven at 65 °C for 48 hours, from which dry matter (%) and biomass (g/m²) were worked out. Macro and micro element content (mg/100g), as well as NO₂-N and NO₃-N accumulation in plants were determined (ppm), and expressed as ratios in fresh samples.

The resulting data set was evaluated by using the linear models procedure of regression analysis on the SPSS statistical package program.

RESULTS AND DISCUSSION

Increasing nitrogen levels result in an increase in yield, biomass and NO₃-N and NO₂-N accumulation in rocket, and decreases in dry matter content (Table 1). N, K, Mg and Zn accumulation in rocket increased with increasing nitrogen application, the linear term was significant for all variables except Zn which had quadratic response. Na showed a linear decrease with increasing N applications. Variables which remained unaffected were Phosphor, Copper and Calcium.

Since the linear term was significant in the accumulation of toxicity, namely NO₃-N and NO₂-N, their further accumulation by increasing N levels can be forecast. NO₃-N accumulation raised from 288 ppm to 594 ppm while nitrogen application increases from 0 to 24 kg/da. Fresh yield increased from 2440 to 5100 kg/m² with increasing nitrogen levels. Since quadratic responses of yield to increasing N levels were as much significant as linear responses, little improvement might be expected by adding more nitrogen if high yield is the objective, and that at the expense of toxicity. This results are partly contradictory to some authors in this field, which report that nitrogen levels at 100 kg/ha are mostly sufficient for acceptable yield (Baggio and Pimpini, 1994; Pimpini and Enzo 1996). In our study this ratio would correspond to 12 kg N levels. However, it must be said that turkish farmers sow 4 g/m² seeds, which was 1.5 g/m² in the experiment Baggio and Pimpini (1994). Eşiyok et al., (1998a) proved this and reported yield increases up to 4 g/m², the ratio which is used in this study as well. Furthermore, it can be said that increases in yield above 12 kg / 1000 m² nitrogen application seems to be slowly.

Table 1. Changes in yield, yield components, NO₃-N, NO₂-N and macro and micro element contents in rocket as affected by nitrogen levels.

<i>Source of Variation</i>								
kg N/ 1000m ²	Yield (g/m ²)	DM (%)	Biomass (g/m ²)	NO ₃ -N (ppm)	NO ₂ -N (ppm)	N (mg/100g)	P (mg/100g)	K (mg/100g)
0	2442 d ²	7,90 a	192,95 d	287,7 c	0,0087 c	384,7 b	46,7	256 b
6	2642 d	7,30 d	192,85 d	349,3 bc	0,0223 bc	396,0 b	51,0	252 b
12	3358 c	7,70 b	258,63 c	395,0 bc	0,0360 bc	427,3 ab	50,3	261 ab
18	4408 b	7,50 c	330,59 b	452,7 b	0,0487 b	453,7 ab	52,7	266 ab
24	5102 a	7,46 c	380,54 a	594,3 a	0,1053 a	493,7 a	55,0	275 a
<i>Mean Squares</i>								
Linear	15066253 **	0,1387 **	78929 **	154083,3 **	0,0145 **	22798 **	100,83 ns	790,5 **
Quad.	373371 **	0,0579 **	2421 **	6339,4 ns	0,0015 ns	587 ns	0,21 ns	97,5 ns
Cubic	228813 *	0,2117 **	2318 **	3000,0 ns	0,0006 ns	12 ns	7,50 ns	26,1 ns
<i>Between</i>								
Groups	25493	0,0057	146	3197,1	0,0003	1867	19,60	51,9

Significance of P ≥ 0.05 (*), P ≥ 0.01 (**) and non significance (ns). ²: Duncan's multiple range test.

Table 1 continued

<i>Source of Variation</i>							
kg N/ 1000m ²	Ca (mg/100g)	Mg (mg/100g)	Na (mg/100g)	Fe (mg/100g)	Zn (mg/100g)	Mn (mg/100g)	Cu (mg/100g)
0	272	24,7 a	12,0 a	2,93 b	0,46 c	0,38 a	0,080
6	274	25,7 a	11,0 ab	3,34 ab	0,48 bc	0,33 b	0,090
12	260	24,3 ab	10,6 ab	4,90 a	0,50 bc	0,38 a	0,087
18	252	23,3 ab	9,5 b	3,65 ab	0,52 ab	0,38 a	0,070
24	246	20,7 b	9,0 b	3,47 ab	0,56 a	0,40 a	0,070
<i>Mean Squares</i>							
Linear	1642,80 ns	32,0 **	16,88 **	0,594 ns	0,000 ns	0,0024 *	0,0004 ns
Quad.	21,43 ns	10,5 ns	0,02 ns	3,417 ns	0,015 **	0,0017 *	0,0003 ns
Cubic	97,20 ns	0,1 ns	0,00 ns	0,002 ns	0,001 ns	0,0019 *	0,0002 ns
<i>Between</i>							
Groups	2355,80	4,1	1,19	0,786	0,001	0,0003	0,0001

Significance of P ≥ 0.05 (*), P ≥ 0.01 (**) and non significance (ns). ²: Duncan's multiple range test.

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DGS-1 --- a new high yielding variety of Gobi sarson.

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Brassica napus is an important oilseed crop and occupy second place in the economy of Indian agriculture. Amongst various species grown in India, Gobi sarson (Brassica napus) is a recent introduction and picking up very well in northern India for higher yield and superior oil quality. Brassica oils are primarily used for edible purposes and defatted meal is used as a supplement for animals. In Brassica breeding, considerable emphasis is being laid to develop high yielding varieties with low glucosinolate and erucic acid. In the present study, under short term breeding programme, large number of cultivars were imported from Sweden, U.K. and Canada to replace the existing cultivated B.napus varieties with double low and high yielding varieties.

Method and Materials

Selections were made from the exotic materials received from different countries of the world. DGS-1 was evaluated alongwith other varieties including check GSL-1 in a randomized block design with three replications during rabi 1995-96 to 1998-99 at Dryland Research Sub-station, Dhiansar Bari Brahamna, Jammu. The data was recorded on yield and its contributing traits. At the same time, seed samples were also used for the determination of biochemical profile.

Results and discussions

DGS-1 recorded the highest seed yield of 17.44, 17.02, 13.52 and 10.17 q/ha during the rabi 1998-99, 1997-98, 1995-96 and 1996-97, respectively. The same variety was also evaluated in the initial evaluation trials (IVT) under the AICRPO at four and six locations during rabi 1998-99 in Zone I and Zone II, respectively and recorded the higher seed yield of 13.88 and 19.29 q/ha in Zone I and Zone II than the check, GSL-1 (13.32 and 17.22 q/ha in Zone I and Zone II, respectively). This variety had the medium erucic acid (43.0%), low glucosinolate (47.0 μ moles/g defatted meal), oil content (36.253), free fatty acids (1.33), acid value (2.47), ether extractives (39.77), iodine value (100.55), saponification value (171.60), protein content (23.05%), palmitic acid (3.1%), stearic acid (0.9%), oleic acid (14.0%), linolic acid (16.1%) linolenic acid (13.7%) and Eicosenoic acid (9.2%). (Annual progress report, 99). It had shown the complete and moderate resistance to white rust and aphid, respectively. The results of mini-Kits of this variety had shown 30.80% increase over the check variety, GSL-1.

Acknowledgement

We are thankful to Nordic Gene Bank, Sweden for providing the different genetic stocks of Brassica napus.

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Table1 Seed yield and biochemical profile of new high yielding variety
DGS-1 and Check Variety GSL-1

Parameters	DGS-1	GSL-1
Seed yield (g/ha)		
at Exp. Res.Farm (av. of four years data) in AiCRPO trials	17.02	12.30
(av.of six locations in Zone-I)	19.19	17.72
Protein(1%)	23.05	23.55
Glucos-inolate (μ moles/g defatted meal)	47.00(TERI)	85.00 (TERI)
Oil Content	36.25	38.50
Ether extractives	39.77	38.97
Iodine value	100.55	103.75
Saponication value	171.60	171.75
Free fatty acids	1.33	2.10
Acid value	2.47	2.10
Fatty acid (%)		
Palmitic acid	3.1	3.7
Stearic acid	0.9	0.6
Oleic acid	14.0	18.4
Linolic acid	16.1	14.0
Linolenic acid	13.7	8.8
Eicosenoic acid	9.2	11.5
Erucic acid	43.0	43.0

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Impact of weather on growth and yield of mustard (*Brassica juncea* L.)

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Introduction

Weather affects the growth of plant in numerous ways, from emergence of seedlings to maturity, by influencing various physiological processes including photosynthesis and transpiration (Nehra *et.al.*1993). Its impact on morphology, development, biomass production, and the time to attain the phenological phases is well established. Further the full genetic potential of a particular cultivar is only obtained when an optimum climatic condition including other environmental factors is available.

Materials and Methods

The experiment was conducted during 1997-98 *rabi* (winter) at Dryland Research Station, Dhiansar (32°-39' N latitude, 74°-58' E longitude and 332 meters amsl). Two mustard cultivars Varuna and Pusa Bahar were sown on three different dates viz; October 9, October 24 and November 9 under rainfed condition with standard agronomic practices. The experiment was laid out in randomised block design (RBD) with four replications. Three phenophases viz; emergence to flower bud initiation (PS₁), flower bud initiation to pod formation (PS₂) and pod formation to maturity (PS₃) were identified. Daily weather data recorded at this station was used for this study.

Results and Discussion

Effect on yield:

The data so obtained from the experiment was analysed and presented in Table:-1

Table 1. Seed yield (q/ha) and total drymatter(q/ha) of mustard crop as affected by different treatments.

(a)	<u>Date of sowing</u>	<u>Seed yield</u>	<u>Total dry matter</u>
	D ₁ (09-10-97)	7.98	38.33
	D ₂ (24-10-97)	9.03	38.31
	D ₃ (08-11-97)	5.58	29.19
	CD at 5%	0.57	2.13
(b)	<u>Varieties</u>		
	V ₁ Varuna	7.75	36.63
	V ₂ Pusa Bahar	7.31	33.92
	CD at 5%	N.S	1.74

The results reveal that the treatment date of sowings have shown significant effect on both seed yield and total dry matter production. Second date of sowing (D₂) gave significantly higher seed yield than first(D₁) and third sowings(D₃). The night temperature experienced by the crop sown in D₂ during the period from flower bud initiation to maturity might be optimum for translocation of photosynthates. Whereas, in case of total dry matter the treatments first(D₁) and second date(D₂) were at par but significantly superior than third(D₃). Further, no significant difference was observed between varieties with regard to seed yield except total dry matter production

Relationship between grain yield and weather parameters:

Correlation and regression analysis between seed yield and average weather parameters including the cumulative heat units during the periods between emergence to flower bud initiation (PS₁), flower bud initiation to pod formation (PS₂) and pod formation to maturity were worked out and are presented in Table 2. The result indicates that the meteorological factors like relative humidity at afternoon (RH₂), evaporation (Ep) during the period PS₁, minimum temperature (MinT) during PS₂ and minimum temperature including vapour

Table:-2 Correlation coefficient between weather parameters during phenophases and seed yield of mustard

Phenophase	MaxT	MinT	GDD	RF	RH ₁	RH ₂	VP ₁	VP ₂	EP
PS ₁	0.771	0.519	0.742	0.487	-0.686	-0.875*	0.499	0.458	0.887*
PS ₂	0.433	0.815*	0.527	0.036	-0.303	-0.108	0.802*	0.766	0.549
PS ₃	-0.716	-0.905**	-0.658	-0.575	0.783	0.680	-0.840*	-0.869	-0.854

* Significant at 5% level

** Significant at 1% level

pressure both morning (VP₁) and afternoon (VP₂) hours, and evaporation (Ep) during the period PS₃ have significant effect on seed yield. A high significant negative association was observed between minimum temperature during PS₂ and seed yield which indicates that low minimum temperature during pod formation are favourable for seed yield. Low temperature during this period might be responsible for more accumulation of photosynthates in pods due to low temperature thereby, increased the yield. The regression equation developed after identifying the phase during which the weather parameters had significant correlation with seed yield. The regression model so obtained

$$y = 97.8 + 10.6 \text{ MinT (PS}_2\text{)} - 0.24 \text{ RH}_2\text{ (PS}_1\text{)} - 19.5 \text{ VP}_1\text{ (PS}_2\text{)} - 3.72 \text{ Ep (PS}_1\text{)} \quad R^2 = 0.80 \dots \text{eq.1}$$

$$y = 61.7 - 1.76 \text{ MinT (PS}_2\text{)} + 4.1 \text{ MinT (PS}_3\text{)} - 0.3 \text{ RH}_2\text{ (PS}_1\text{)} - 60.6 \text{ Ep (PS}_3\text{)} \quad R^2 = 0.91 \dots \text{eq.2}$$

The eq.1 can predict the seed yield 50 to 60 days in advance with lesser efficiency ($R^2=0.80$) whereas eq.2 predicts the seed yield at ripening stage with higher efficiency ($R^2=0.91$)

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Stability in *Brassica juncea* over North - South and East-West sowing directions acidic soil of high altitude

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ABSTRACT

The parent RW 873 and crosses PR 830 x RH 857, RW 29-6-3 x RH 851 and RH 843 x BR 40 were found to be stable for yield per plant, test weight and oil content along with other yield attributes.

Key words :- Indian mustard, South - North and East - West sowing, stability.

Introduction

During the last five decades, there has been a substantial increase in production of oilseeds in general and rapeseed-mustard in particular. In India, the major emphasis has been on increasing the seed yield and stability of lines to different agro-climatic conditions and, as such, the major breeding goals for these crops have been centered on yield, stability and wider adaptability. With these goals, the present investigation on *Brassica juncea* was undertaken to identify stable genotype over varying environmental condition and genotype x environment interactions for seed yield and other quantitative characters including the oil content.

Materials and Methods

The materials for the study generated during winter 1995-96 consisted of 45 genotypes (9 parents and their diallels excluding reciprocals). The above 45 genotypes were sown on three dates (E1 & E4 on 27th September, E2 & E5 on 4th October and E3 & E6 on 11th October 1997) at North-South as well as in East-West sowing directions with two replications on each date during winter season at the Birsa Agricultural University experimental area, Ranchi. The distance between the rows and plants was maintained at 30 and 10 cm, respectively. Cultural practices as recommended for the area were followed. The observations were recorded for eleven yield attributes (Table 1) from ten randomly selected competitive plants. Analysis was done following Eberhart and Russell (1966).

Results

Among the 45 genotypes, PR 830 x Vardan stable for 9 different characters *i.e.*, days to 50% flowering, plant height, number of siliquae per plant, number of seeds per siliqua, days to maturity, 1000-seed weight, seed yield per plant, and oil content (%) in seed yield and oil content. The genotypes per RW 873 x RH 851, Kranti x RW 29-6-3 and RH 843 x BR 40 were stable for eight yield and yield attributes. RW 873 x RH 851 and RH 843 x BR 40 for seed yield per plant and oil content in percent and other six attributes, where as Kranti x RW 29-6-3 stable for oil content and other seven characters excepting seed yield per plant. PR 830, Kranti x RH 851, RW 29-6-3 x RH 843, RW 29-6-3 x RH 851, RW 29-6-3 x Vardan, RW 29-6-3 x BR 40, PR 18 x RH 851, RH 843 x RH 851 and RH 851 were stable for seven yield attributes . Kranti and Kranti x RH 851 for yield per plant, RW 29-6-3 x RH 851, RW 29-6-3 x Vardan, RW 29-6-3 x BR 40, PR 18 x RH 851 and PR 18 x BR 40 for seed yield per plant and oil content and rest for other yield attributes were found stable. RW 873, PR 830 x RW 29-6-3, PR 830 x PR 18, Kranti x PR 18, Kranti x Vardan and PR 18 x RH 843 were found stable for six yield attributes.

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Table 1. Stability performance of crosses and parents for different traits.

Genotypes	1	2	3	4	5	6	7	8	9	10	11	12
RW 873	*	*	*						*	*	*	6
RW 873 X PR 830		*					*					2
" X Kranti			*	*						*		3
" X RW 29-6-3				*				*			*	3
" X PR 18				*	*		*					3
" X RH 843			*	*		*						3
" X RH 851	*	*		*	*		*	*		*	*	8
" X Vardan	*		*		*		*	*		*		5
" X BR 40						*	*	*		*		4
PR 830	*		*	*	*	*	*		*			7
PR 830 X Kranti	*		*				*					3
" X RW 29-6-3	*		*	*	*	*				*		6
" X PR 18	*		*	*	*		*			*		6
" X RH 843	*			*			*		*			4
" X RH 851	*			*	*	*	*	*	*	*	*	9
" X Vardan		*		*		*		*		*		5
" X BR 40							*	*	*	*	*	1
Kranti	*	*			*		*	*	*	*		7
Kranti X RW 29-6-3	*	*		*		*	*	*	*	*	*	8
" X PR 18	*	*	*	*			*	*	*	*	*	6
" X RH 843		*	*	*			*		*			5
" X RH 851	*	*		*	*		*	*		*		7
" X Vardan	*			*	*		*	*		*		6
" X BR 40		*	*				*			*	*	4
RW 29-6-3	*			*			*		*	*		5
RW 29-6-3 X PR 18	*			*			*		*	*		3
" X RH 843	*	*		*	*		*	*	*	*	*	7
" X RH 851	*			*	*	*	*	*	*	*	*	7
" X Vardan		*		*	*		*	*	*	*	*	7
" X BR 40	*	*				*		*	*	*	*	7
PR 18	*			*	*		*		*	*		5
PR 18 X RH 843	*			*	*		*	*	*	*	*	6
" X RH 851	*	*	*	*	*		*		*	*	*	7
" X Vardan	*	*		*			*		*	*	*	3
" X BR 40	*		*	*		*		*	*	*	*	7
RH 843	*	*		*	*		*		*	*		5
RH 843 X RH 851	*	*		*	*	*	*		*	*	*	7
" X Vardan	*			*		*		*	*	*	*	5
" X BR 40		*	*	*	*	*		*	*	*	*	8
RH 851	*	*		*	*	*		*	*	*		7
RH 851 X Vardan			*		*						*	2
" X BR 40	*	*		*							*	4
Vardan	*										*	3
Vardan X BR 40			*	*		*			*	*	*	5
BR 40	*	*	*					*		*	*	5

1. Days to 50% flowering, 2. Primary branches per plant, 3. Secondary branches per plant, 4. Plant height, 5. Siliquae per plant, 6. Seeds per siliqua, 7. Days to maturity, 8. Harvest index, 9. 1000 – seed weight, 10. Seed yield per plant, 11. Oil content (%) and 12. Stable for characters.

Identification of high heterotic genotypes over environment in (*Brassica juncea*) in acidic soil of high altitude

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ABSTRACT

The Crosses PR 18 x BR 40 for seed yield per plant, oil content, harvest index and number of siliquae per plant, showed high heterotic effect over environments for heterosis over better and Mid parent.

Key words: Heterosis, mid-parent, better parent, mustard, *Brassica juncea*.

Introduction

Mustard is important oilseed crop. Its yield and yield attributes like other crop are more influenced by environment. Very scanty study was done heterotic variation over environments. In present study was undertaken to study the heterotic variation over different environments.

Materials and Methods

The materials for the study generated during winter 995-96 consisted of 45 genotypes (9 parents and their diallels excluding reciprocals). The above 45 genotypes were sown on three dates (E1 & E4 on 27th September, E2 & E5 on 4th October and E3 & E6 on 11th October 1997) at North-South as well as in East-West sowing directions with two replications on each date during winter season at the Birsa Agricultural University experimental area, Ranchi. The distance between the rows and plants was maintained at 30 and 10 cm, respectively. Cultural practices as recommended for the area were followed. The observations were recorded for eleven yield attributes (Table 1) from ten randomly selected competitive plants. Estimation of heterosis was done as per standard method.

RESULT

The crosses PR 18 x BR 40 showed high heterotic value of heterosis over better. Mid parent for the seed yield per plant, oil content, harvest index and number of siliquae per plant. Range of heterosis over mid and better was varied from -62.697 to 317.317 and -75.569 to 270.762, respectively, for different characters. RW 873 x RW 29-6-3 and RH 843 x BR 40 showed high heterotic effect for over both better and Mid - parent for days to maturity.

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Table 1. Range of heterosis and highly heterotic crosses for different characters.

<i>Characters</i>		<i>Range</i>	<i>Highly heterotic crosses</i>
Days to 50% flowering	MP	(-11.719)-(15.686)	RW 873 x RW 29-6-3, RH 843 x BR 40, Vardan x BR 40, RW 873 x PR 830
	BP	(-9.054)-(19.967)	RW 873 x RW 29-6-3, RH 843 x BR 40
Number of primary branches per plant	MP	(-29.832)-(25.529)	PR 830 x RH 851, Kranti x Vardan, RH 843 x Vardan, PR 830 x PR 18
	BP	(-42.035)-(20.000)	PR 830 x PR 18, PR 18 x BR40, PR 830 x RH 851, RW 873 x PR 830
Number of secondary branches per plant	MP	(-42.105)-(69.867)	Vardan x BR 40, RH 843 x RH 851, PR 830 x RH 851, RH 851 x BR 40
	BP	(-59.3)-(63.718)	PR 830 x RH 851, Vardan x BR 40, RW 873 x RH 851, RH 843 x RH 851
Plant height	MP	(-13.927)-(15.862)	RW 29-6-3 x BR 40, PR 830 x BR 40
	BP	(-7.781)-(32.479)	-
Number of siliquae per plant	MP	(-33.579)-(172.144)	PR 18 x BR 40, RH 843 x RH 851, Vardan x BR 40
	BP	(-46.961)- (138.505)	PR 18 x BR 40
Number of seeds per siliqua	MP	(-22.402)-(26.754)	RW 873 x RH 843, RH 843 x RH 851, PR 18 x RH 843, Kranti x RH 843
	BP	(-27.285)-(14.589)	RW 873 x RH 843, RH 843 x RH 851, RH 843 x Kranti, RW 29-6-3 x RH 843
Days to maturity	MP	(-10.249)-(9.152)	RW 873 x RW 29-6-3, RW 29-6-3 x RH 843
	BP	(-8.046)-(9.65)	RW 873 x RW 29-6-3
Harvest Index	MP	(-29.639)-(68.789)	PR 830 x RW 29-6-3, PR 830 x BR 40, RH 843 x BR 40, PR 18 x BR 40
	BP	(-30.601)-(46.409)	PR 830 x RW 29-6-3, PR 830 x BR 40, PR 18 x BR 40, RW 29-6-3 x PR 18
1000-seed weight	MP	(-19.801)-(35.63)	PR 830 x Vardan, RW 873 x PR 18, RW 29-6-3 x Vardan, RW 873 x RH 843
	BP	(-28.263)-(26.667)	RW 873 x PR 18, PR 830 x Vardan, RW 873 x RH 843, RW 29-6-3 x Vardan
Seed yield per plant	MP	(-62.697)-(317.317)	PR 18 x BR 40, RW 873 x PR 18, PR 830 x RH 851, RH 843 x Vardan
	BP	(-75.569)-(270-762)	PR 18 x BR 40, PR 830 x RH 851, RW 873 x PR 18, RH 843 x Vardan
Oil content (%)	MP	(-13.61)-(7.24)	RW 873 x PR 830, RW 29-6-3 x PR 18, PR 18 x BR 40, RH 843 x BR 40
	BP	(-13.87) - (4.52)	RW 873 x PR 830, RW 29-6-3 x PR 18, PR 18 x BR 40, RH 843 x BR 40

MP = Mid - Parent, BP = Better - Parent

Performance of *Brassica juncea* genotypes under different environments at high altitude

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ABSTRACT

Studies on 45 genotypes in six environments (3 North - South and 3 East - West sowing directions) showed significant superiority most of yield attributes in East - West sowing direction.

Introduction

Seed yield can be increased either through genetic manipulations or through appropriate management. If the genetic manipulation is coupled with proper management, seed yield can further be increased. The present work was initiated to study the performance of genetically manipulated lines of *Brassica juncea* L. in different environments.

Materials and Methods

The materials generated during winter 1995-96 consisted of 45 genotypes (9 parents and their diallels excluding reciprocals). These were sown on three dates (E1 & E4 on 27th September, E2 & E5 on 4th October and E3 & E6 on 11th October 1997) at North-South as well as in East-West row directions with two replications on each date at oilseed research farm of the University. The p^H of the soil being 5.9. The area is located between 23°17' latitude and 85°19'E longitude and altitude is 625 meters above the mean sea level. The distance between the rows and plants was maintained at 30 and 10 cm, respectively. Cultural practices as recommended for the area were followed. The observations were recorded for eleven yield attributes (Table 1) from ten randomly selected competitive plants.

Results

The performance of 45 genotypes of *Brassica juncea* L. over environments (three sowing dates and two row directions) is presented in Table 1. It is evident from the result that the mean performance for yield and yield attributes in general remained superior in East - west direction. It is further seen that late sown crop (E6) performed better with respect to various yield contributing parameters and which ultimately got reflected as seed yield per plant. There has, however, been loss in oil content at all the dates in East - West sown crop. It is due to negative relation with seed yield.

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Table 1. Mean performance of different attributes under different environments

Parameters	North - South Sowing				East - West Sowing			
	E1	E2	E3	Mean	E4	E5	E6	Mean
			<u>Days to 50 % flowering</u>					
Grand Mean	56.2	51.3	66.4	57.9	62.3	52.8	68.9	61.3
C.D. at 5%				10.8				17.7
			<u>Number of primary branches per plant</u>					
Grand Mean	3.8	4.0	3.7	3.9	3.8	4.4	4.7	4.3
C.D. at 5%				2.0				2.0
			<u>Number of Secondary Branches per plant</u>					
Grand Mean	5.7	6.9	5.2	5.9	5.1	4.8	5.1	5.0
C.D. at 5%				4.4				9.8
			<u>Plant height (cm)</u>					
Grand Mean	118.9	126.4	113.5	119.6	105.5	130.7	124.7	120.3
C.D. at 5%				21.1				22.8
			<u>Number of siliquae per plant</u>					
Grand Mean	136.9	168.9	114.8	150.6	117.2	95.7	288.1	185.1
C.D. at 5%				11.1				17.2
			<u>Number of seeds per siliqua</u>					
Grand Mean	10.3	11.1	10.3	10.7	10.9	11.8	10.3	11.0
C.D. at 5%				4.1				5.2
			<u>Days to maturity</u>					
Grand Mean	104.4	104.8	119.3	111.9	114.3	104.0	121.7	114.2
C.D. at 5%				18.7				8.6
			<u>1000-Seed weight (g)</u>					
Grand Mean	2.923	3.151	2.999	3.021	2.806	3.085	3.123	3.057
C.D. at 5%				0.870				0.739
			<u>Seed yield per plant (g)</u>					
Grand Mean	3.350	4.348	2.821	3.501	2.483	6.366	6.685	5.178
C.D. at 5%				2.604				4.905
			<u>Oil content (%)</u>					
Grand Mean	38.4	38.80	38.82	38.70	38.73	38.29	37.48	38.13
C.D. at 5%				2.35				3.04
			<u>Harvest Index</u>					
Grand Mean	0.171	0.209	0.179	0.188	0.169	0.192	0.165	0.175
C.D. at 5%				0.143				0.072

CHANGES IN LENGTH OF INNER STUMP OF WHITE CABBAGE (*BRASSICA OLERACEA* VAR. *CAPITATA* L.) CULTIVARS DURING STORAGE

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ABSTRACT

The purpose of the study is to trace the changes in the length of the inner stump that directly affect the results from the storage.

Three direct and eight heterotic cultivars of Bulgarian, Czech and Dutch origin were studied.

It was established that in cultivars with good storage properties, such as Erdeno F₁, Quisto F₁, Bislet F₁, Sagitta F₁, Menza F₁ and Ramco F₁ the prolongation of the inner stump is not significant compared to the cultivars with poor storage properties.

INTRODUCTION

During the storage of white cabbage produce there are complicated biochemical and physiological processes that affect the duration of the storage. They often cause morphological changes which registration can give valuable back information connected with the adaptability of the cultivars to long storage (Shirokov, E., 1974).

The purpose of the present study was to trace the changes in the length of the inner stump during storage that directly affect the results from the storage.

MATERIAL AND METHODS

The following cultivars have been studied: Erdeno F₁, Ramco F₁, Quisto F₁ and Bislet F₁ (origin – Sandoz Seeds), Menza F₁, Oscar F₁, Sagitta F₁ (Royal Sluis), Rodolfo F₁ (Bejo Zaden), Dobrowodske polopozdni and Trwanliwa D (SEMPRA, The Czech Republic). During storage temperature of 0 to 1 °C and relative humidity of 85-90 % was maintained. The records were taken at regular intervals (every 45 days average) from 50 cabbage heads from cultivar using the method of the fixed samples (Широков, 1964).

RESULTS AND DISCUSSION

One of the important characteristic features of cabbage cultivars with long storage ability is the long period of dormancy of the vegetative apex. The break of the apical vegetative bud dormancy is a result from complex biochemical processes connected with a new stage in the development of the cabbage plant. From that moment on there is a fast flow of biologically active substances from the leaves constructing the cabbage head to the apical bud. The inner stump prolongs which results in cabbage heads crack. Measuring the length of the inner stump we can make conclusions about the aptitude of the apical bud to premature wakening.

From the investigations carried out (Table 1) the largest prolongation of the inner stump is registered in Oscar F₁ and Dobrowodske polopozdni (+2.1 cm), followed by Rodolfo F₁ (+1.8 cm) and Trwanliwa D (+1.4 cm). The relatively shorter prolongation of the inner stump in the control should not mislead us, because it has been registered during

the second recording period. At the end of the storage of Balkan there were single cabbage heads that have been totally dead and there was no recording.

In the cultivars with good storage properties (Михов, Кр., 1997) the prolongation of the inner stump is not significant compared to the cultivars with poor storage abilities. The inner stump in Erdeno F₁, Quisto F₁, Bislet F₁, Sagitta F₁, Menza F₁ and Ramco F₁ is longer with 0.1 to 0.6 cm, respectively.

TABLE 1.

CHANGES IN THE LENGTH OF THE INNER STUMP DURING THE PERIOD OF STORAGE

CULTIVAR	LENGTH OF THE INNER STUMP (CM)			DURATION OF THE PERIOD OF STORAGE (DAYS)
	1	2	DIFFERENCE	
Balkan – the control	6.0	7.1	+1.1	94
Erdeno F ₁	6.1	6.3	+0.2	126
Menza F ₁	6.4	6.8	+0.4	126
Oscar F ₁	5.7	7.8	+2.1	126
Sagitta F ₁	6.0	6.4	+0.4	126
Rodolfo F ₁	5.9	7.7	+1.8	126
Ramco F ₁	5.5	6.1	+0.6	126
Quisto F ₁	7.0	7.2	+0.2	126
Bislet F ₁	7.0	7.1	+0.1	126
Dobrowodske polopozdni	8.0	10.1	+2.1	126
Trwanliwa D	6.2	7.6	+1.4	126

1 – at the time of putting under storage

2 – at the end of storage

CONCLUSIONS

There is a dependency between the storage abilities of the cultivars and the prolongation of the inner stump during storage. It was established that in cultivars with good storage abilities, such as Erdeno F₁, Quisto F₁, Bislet F₁, Sagitta F₁, Menza F₁ and Ramco F₁ the prolongation of the inner stump is not significant compared to the cultivars with poor storage properties.

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ASSESSMENT OF BROCCOLI (*BRASSICA OLERACEA* VAR. *ITALICA* PL.) HYBRIDS FOR LATE FIELD PRODUCTION

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ABSTRACT

The purpose of the study is to make a preliminary assessment of broccoli (*Brassica oleracea* var. *italica* Pl.) cultivars and hybrids for late field production under the conditions of Bulgaria. Ten broccoli hybrids were investigated with regard to the length of the vegetative period and some elements of their productivity.

The most appropriate broccoli hybrids for late field production are Cruiser F₁, Beaufort F₁ and Tribute F₁, which yield is 19400, 19000, and 15000 kg/ha, and length of the vegetative period – 72, 82 and 92 days, respectively.

INTRODUCTION

Broccoli (*Brassica oleracea* var. *italica* Pl.), one of the basic cabbage crops in the West European countries, is little known and rarely spread in Bulgaria. The preliminary assessment of cultivars and hybrids reveals the opportunity to choose the most appropriate ones for a given type of production (Dellacecca et al., 1988; Walton and Casada, 1986).

The purpose of our study was to test and assess some broccoli hybrids with regard to the length of the vegetative period and some elements of their productivity.

MATERIAL AND METHODS

During the period 1997-1998 ten broccoli hybrids were tested following a technology of late field production. The planting was grown at density of 28000 plants/ha with planting date 20 July. The experiment was conducted using the block method in 4 replications.

Registered were the length of the vegetative period (days from planting to harvesting of 50 % of the first flower heads), weight and head diameter and mean yield of the first flower heads from hectare. The results were analysed using Duncan's multiple range test.

RESULTS AND DISCUSSION

All the studied broccoli hybrids were with good adaptive capability towards the conditions and technology of growing. The length of the vegetative period is 72 – 92 days and the mean yield is 10500 to 19400 kg/ha (Table 1).

TABLE 1.

CHARACTERISTIC FEATURES OF BROCCOLI HYBRIDS

HYBRID	VEGETATIVE PERIOD (DAYS)	WEIGHT OF FLOWER HEAD (KG)	DIAMETER OF FLOWER HEAD (KG)	MEAN YIELD (KG/HA)
Cruiser F ₁	72 ^c	1.000 ^a	25.8 ^a	19400 ^a
Beaufort F ₁	82 ^b	1.100 ^a	23.5 ^{ab}	19000 ^a
Tribute F ₁	92 ^a	0.800 ^{ab}	23.2 ^{ab}	15000 ^{ab}
Emperor F ₁	72 ^c	0.650 ^b	20.9 ^{bc}	14000 ^b
Neptune F ₁	92 ^a	0.700 ^b	27.2 ^a	14000 ^b
Delicia F ₁	72 ^c	0.600 ^b	21.4 ^b	13500 ^b
Corvet F ₁	82 ^b	0.600 ^b	22.9 ^b	13500 ^b
Regilio F ₁	82 ^b	0.650 ^b	22.4 ^b	13500 ^b
Skiff F ₁	92 ^a	0.550 ^c	18.7 ^c	11000 ^{bc}
Sumosun F ₁	72 ^c	0.400 ^c	17.5 ^d	10500 ^c

The hybrids with the highest yield are Cruiser F₁ and Beaufort F₁ with mean yield of 19400 and 19000 kg/ha and length of the vegetative period 72 and 82 days, respectively. The flower heads are with weight of 1.000 and 1.100 kg and diameter of 25.8 and 29.5 cm, respectively.

In the hybrids with length of the vegetative period 92 days Tribute F₁ is with the highest productivity with mean yield of 15000 kg/ha, weight of the flower head of 0.800 kg, and diameter of 23.2 cm.

CONCLUSIONS

The most highly productive and the most appropriate for late field production broccoli hybrids are Cruiser F₁, Beaufort F₁ and Tribute F₁.

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ASSESSMENT OF CAULIFLOWER (*BRASSICA OLERACEA* VAR. *BOTRITIS* L.) CULTIVARS AND HYBRIDS FOR LATE FIELD PRODUCTION

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INTRODUCTION

The basic cauliflower (*Brassica oleracea* var. *botritis* L.) cultivars grown in Bulgaria are Erfurter and Suprimax. The choice of a new cultivar structure requires availability of highly productive and quality cultivars and hybrids adapted to the conditions of growing and production with maximum realisation of their biological potentialities (Larsen, J., 1987; Ruffio-Chable and Herve, 1987; Souza J. et al., 1987).

The purpose of the investigation was to study some cauliflower cultivars and hybrids for late field production with regard to some features of their morphological characteristics, the length of the vegetative period and their productivity.

MATERIAL AND METHODS

During the period 1996-1998 five cauliflower cultivars and hybrids were tested using the technology for late field production. Sowing was conducted on 10 June, and planting – on 15 July, with a planting scheme of 70/60 cm in 4 replications. Registered were length of stem, size of leaf rosette, diameter, length and compactness of curds, length of the vegetative period (days from planting to harvesting of 50 % of the plants), length of the harvesting period, curd weight, standard and non-standard produce. The results were analysed using Duncan's multiple range test.

RESULTS AND DISCUSSION

The length of the outer stem of the tested cauliflower cultivars and hybrids is 28 to 45 cm (table 1). In Celesta and Suprimax the length of the stem is 45 and 43 cm, respectively. The largest leaf rosette was formed by the plants of Celesta (100-110 cm) and Koket F₁ (95-99 cm). Koket F₁ had the shortest stem (28 cm).

TABLE 1.

MORPHOLOGICAL CHARACTERISTICS OF CAULIFLOWER CULTIVARS AND HYBRIDS

CULTIVAR (HYBRID)	LENGTH OF STEM (CM)	DIAMETER OF LEAF ROSETTE (CM)	ROSETTE LEAVES (NUMBER)	CURD DIAMETER (CM)	LENGTH OF CURD (CM)	COMPACTNESS OF THE CURD (RATING 1 TO 5)
Suprimax	43 ^a	90 – 93 ^{bc}	28 ^a	15.6 ^d	8.3 ^c	4.5 ^b
Celesta	45 ^a	100–110 ^a	22 ^b	17.4 ^c	8.7 ^{bc}	5 ^a
Turbo	29 ^{bc}	83 – 90 ^c	17 ^a	22.9 ^a	12.6 ^a	3.5 ^c
Parti F ₁	31 ^b	87 – 92 ^{bc}	24 ^b	20.0 ^b	11.7 ^a	4.5 ^b
Koket F ₁	28 ^c	95 – 99 ^b	21 ^{bc}	18.2 ^b	10.7 ^b	5 ^a

P = 0.05

The plants from the cultivar Suprimax and Parti F₁ are with very leafy rosettes, 28 and 24 leaves, respectively. The cultivar Turbo has the largest curds with diameter of 22.9 cm and length of 12.6 cm, but with the lowest compactness (rating 3.5). The curds formed by Celesta and Koket F₁ are with a very tight compactness (rating 5), which determines the high percentage of the standard produce obtained from them (Table 2).

TABLE 2.

CHARACTERISTIC FEATURES OF CAULIFLOWER CULTIVARS AND HYBRIDS

CULTIVAR (HYBRID)	LENGTH OF VEGETATIVE PERIOD (DAYS)	LENGTH OF HARVESTING PERIOD (DAYS)	WEIGHT OF CURD (KG)	TOTAL PRODUCTION			
				STANDARD		NON-STANDARD	
				KG/HA	%	KG/HA	%
Suprimax	90 ^a	20 ^b	0.805 ^c	13000 ^b	72.22	5000 ^c	27.78
Celesta	87 ^b	14 ^c	1.010 ^a	15000 ^a	88.23	2000 ^c	11.77
Turbo	78 ^c	21 ^a	0.933 ^b	10000 ^c	58.82	7000 ^b	41.18
Parti F ₁	74 ^d	21 ^a	1.320 ^a	13000 ^b	59.09	9000 ^a	40.91
Koket F ₁	93 ^a	14 ^c	0.850 ^{bc}	12000 ^{bc}	75.00	4000 ^d	25.00

P = 0.05

The highest standard yield was obtained from the cultivar Celesta (15000 kg/ha). Koket F₁ has the longest vegetative period and the shortest harvesting period (93 and 14 days, respectively). The cultivar Celesta was the most productive one, which is determined by the large curds that it forms. In the rest of the cultivars and hybrids with prolonging the period of harvesting the percentage of the non-standard produce is increased.

CONCLUSION

As a result of the conducted investigation the most appropriate for late field production cultivars and hybrids from the tested ones are Celesta and Koket F₁. They give plants with powerful habitus, very leafy rosette and relatively large curds with tight compactness.

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EFFECT OF SULPHUR AND BORON ON SEED YIELD AND QUALITY TRAITS IN *Brassica Campestris*

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Rapeseed and mustard are the major oilseed crops of India. Oilseeds belonging to *Brassica* family have higher sulphur requirement as their oil storage organs are rich in protein of which sulphur is a constituent (Subbiah and Singh, 1970). Boron requirement of this crop has also been well established (Gupta *et al.*, 1985). Very limited informations are available regarding the sulphur and boron nutrition of rapeseed particularly under subtropical temperate transitional zone of Jammu. Therefore, the present investigation was undertaken with a view to find out the effect of sulphur and boron on *B. campestris* var. Kos-1.

The experiment was conducted with rapeseed during winters of 1996-97 and 1997-98 at Regional Agricultural Research Station, SKUAST, Rajouri. Available boron was estimated to 0.27 mg.Kg⁻¹ (Parker and Gardner, 1981). Treatments comprised of four levels of sulphur (0, 10, 20 and 40 Kg .ha⁻¹) and four levels of borax (0, 2.5, 5.0 and 7.5 Kg. ha⁻¹) were evaluated in a randomized block design with four replications.

The results revealed that application of sulphur caused a significant increase in the seed yield of rapeseed (Table 1). The extent of increase were 25.8 and 20.9% above control with 20 and 40 Kg S. ha⁻¹ respectively. The oil content and protein content were increased significantly upto 43.3 and 22.1 % with 40 Kg S.ha⁻¹, respectively. Similar observation was also reported by Mohan and Sharma (1992).

Application of boron @ 5.0 Kg borax. ha⁻¹ caused significant increase in seed yield (22.9%) and protein content. This might be due to the fact that as the soil was boron deficient (critical limit 0.5 mg. Kg⁻¹), the crop, particularly rapeseed responded positively with applied boron, which is required for proper pollination, seedset and protein synthesis (Tisdale *et al.*, 1985). Therefore, application of boron as well as sulphur to rapeseed under intermediate zone of Jammu might be beneficial for optimum growth, seed yield and quality traits.

Table 1 : Effect of sulphur and boron on rapeseed (Average of 1996 - 97 and 1997 - 98)

Treatment	Seed Yield (q .ha ⁻¹)	Height (cms)	Siliqua/plant	1000 seed Wt. (gm)	Oil content (%)	Protein content (%)
B ₀	10.14	98.7	60.7	2.5	37.1	19.4
B _{2.5}	10.78	99.6	64.6	2.7	37.4	20.4
B _{5.0}	12.47	104.2	65.8	2.9	38.2	22.1
B _{7.5}	12.06	101.9	65.5	2.8	38.3	20.5
CD _(P=0.05)	0.78	NS	0.6	NS	NS	0.3
S ₀	11.31	99.5	63.8	2.7	38.0	19.7
S ₁₀	12.45	100.4	65.4	2.9	42.5	20.0
S ₂₀	14.23	101.7	67.1	3.1	43.2	20.4
S ₄₀	13.67	101.2	66.5	2.8	43.3	22.1
CD _(P=0.05)	0.84	NS	NS	NS	1.6	0.5

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THERMAL USE EFFICIENCY IN *BRASSICA* SPECIES UNDER DIFFERENT NITROGEN LEVELS

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Temperature is very important climatic variable which determines the seeding time and consequently the rate and duration of growth and ultimately the productivity of crop. A change in optimum temperature during its vegetative or reproductive growth, adversely affects the onset and duration of phenophases and yield of a crop. The concept of thermal time has been used in scheduling planting and prediction of different phenophases (Nield, 1977). Hence the present investigations was undertaken to study the thermal use efficiency in *Brassicā* species under different nitrogen levels.

A field experiment was conducted at research farm of Department of Agricultural Meteorology Chaudhary Charan Singh Haryana Agricultural University, Hisar (29° 10' N Latitude, 75° 46' E longitude and 215m altitude) during *rabi* season 1995-96. Four *Brassica* spp : *Brassica juncea*, *B. campestris*, *B. napus* and *B. carinata* were sown under four different nitrogen levels/ha (N₀ - No application, N₁ - 40 Kg, N₂ - 80 Kg and N₃ - 120 Kg) in factorial randomized block design with three replications. All recommended package of practices for brassica crop were followed. Dry matter and seed yield (Kg/ha) were recorded in each plot. Daily temperature data were recorded at agrometeorological observatory situated about 100 m away from the experimental field. Cumulative heat units were computed using the expression :

$$HU = \sum_{ds}^{dm} \frac{T_{mx} + T_{mn}}{2} - T_b$$

Where, HU - Heat units

T_{mx} and T_{mn} - Daily maximum and minimum temperatures respectively

T_b - Base temperature = 5°C (Bishnoi, 1989)

ds and dm - Dates of sowing and maturity of the crop respectively.

Thermal use efficiency was computed as :

Dry matter (Kg/ha)

 Cumulative heat units (°C days)

Thermal use efficiency and seed yield of four *Brassica* species under different nitrogen levels are presented in table 1. *Brassica juncea* showed highest thermal use efficiency for dry biomass production followed by *Brassica campestris*, *B. carinata* and *B. napus*. However,

in later brassica species thermal use efficiency values were statistically at par. Singh *et al* (1995) also studied the thermal use efficiency in toria crop.

Nitrogen application has significantly influenced the thermal efficiency of *Brassica* species. Thermal efficiency was increased with increase in nitrogen application. The thermal efficiency of brassica was 111.0 per cent higher in crop applied with 120 Kg/Nitrogen over crop with no nitrogen application.

Brassica juncea produced significantly highest seed yield among all the *Brassica* species. This could be because of better thermal use efficiency of *Brassica juncea* over other *Brassica* species. Brassica crop fertilized with 120 Kg/ha nitrogen produced maximum seed yield. An increase in seed yield was observed with increase in nitrogen application.

Nitrogen application in *Brassica* species has significantly improved thermal use efficiency for dry matter production and seed yield.

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Table 1 : Effect of nitrogen application on thermal use efficiency for drymatter production and yield of *Brassica* species.

Species/Treatments	Thermal use efficiency (Kg/ha/°Cday)	Seed yield (q/ha)
<i>Brassica juncea</i>	4.26	15.30
<i>B. campestris</i>	3.72	11.78
<i>B. napus</i>	2.50	7.60
<i>B. carinata</i>	2.53	10.70
C.D. at 5%	0.29	0.90
Nitrogen levels		
N ₀	2.09	6.47
N ₁	2.59	8.31
N ₂	3.65	14.17
N ₃	4.41	16.43
C.D. at 5%	0.38	0.87

HETEROSIS IN RELATION TO GENETIC DIVERSITY IN INDIAN MUSTARD

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In heterosis breeding programme, progenies derived from diverse crosses are expected to give high heterosis for seed yield. But, there are many cases where high magnitude of heterosis is not always directly relate to extreme parental diversity. The present study was therefore, under taken to know the relationship of heterosis and genetic diversity among 28 genotypes of Indian mustard.

The material for the present study consisted of 7 genotypes (RSK 28 PR 8903, NDR 8501, RK 8902, RH 8814, DIRA 313 AND DLM 2) along with their 21 F_1 s were grown separately in RBD with three replications. Observations were recorded for days to 50% flowering, seed yield/plant g), 1000-seed weight (g), oil₂ content and seedling vigour. Genetic diversity was estimated using D^2 statistic (Rao, 1952) and heterosis was measured as deviation of F_1 from better parent.

Seven parents and their 21 hybrids were grouped into five clusters. Parents were distributed over two clusters, IV and V, while hybrids spread over cluster I, II and III. Genotypes belonging to different states of India constituted a single cluster (V) which may be due to similarity in requirements and selection approaches followed under domestic cultivation (Arunachalam and Ram, 1967). Further, there was a free exchange of seed material among the different regions. Consequently, character constellation that might be associated with a particular region in nature loose their individuality under human interference. Cluster I, II and III consisting only of hybrids indicated that considerable variation was created by hybridization and they were also widely dispersed from the parents. Another feature that came to light was that two varieties from New Delhi were placed to separate clusters indicating wide genetic diversity among genotypes originating from the same geographic region. The free clustering of genotypes suggested dependence upon the directional selection pressure applied for realizing maximum yield in different regimes; the nicely evolved homeostatic device will favour constancy of the associated characters and will thus show indiscriminate clustering. All the lines from Uttar Pradesh were grouped into a single cluster (IV) indicating commonness in genetic structure and selection history.

In general, there was not a fare agreement between the degree of heterosis and the distance between the parental clusters (Table 1). The high manifestation of heterosis for seed yield was evidenced by the significant superiority of heterosis over better parent ranging from 7.48% (PR 8903 x RH 8814) to 58.09% (PR 8903 x DLM 2). Hybrids RK 8902 x DLM 2, PR 8903 x RK 8902 and RSK 28 x DLM 2 with medium divergence between parental clusters exhibited higher magnitude of desirable heterosis for seedling vigour, seed yield, days to 50% flowering, oil content and 1000-seed weight. Thakur and Zarger (1989) also reported similar findings in Indian mustard. Crosses with high and low divergence between parental clusters exhibited negative heterosis for seed yield and other traits except PR 8903 x DIRA 313. The present study revealed that there should be an optimum level of genetic divergence to obtain economic heterosis in F_1 for seed yield in Indian mustard.

Table-1 Relationship between genetic diversity and heterosis in Indian mustard.

Cross	Heterosis over better parent(%)					
	Divergence between parental lines	Seedling vigour	Days to 50% flowering	Seed yield	Cil content	1000-seed weight
RSK 28 x PR 8903	160.40	27.89**	-10.90**	-20.09**	-3.31**	20.84**
NDR 8501 x RH 8814	160.40	-26.91**	-16.61**	-31.53**	-10.08**	-5.58**
PR 8903 x RH 8814	160.40	5.57*	- 7.88	7.48	5.58**	6.10**
RSK 28 x NDR 8501	160.40	15.98**	- 9.00**	9.65	1.33	5.38*
RK8902xRH 8814	160.40	27.90**	-18.41**	-19.98**	3.94**	-13.30**
RSK 28 x RK 8902	160.40	-29.38**	6.45	- 3.53	1.81	-7.08**
RSK 28 x RH 8814	89.80	19.46**	-24.16**	11.24	3.68**	15.72**
RH 8814 x DIRA 313	66.87	- 5.99**	10.40*	-13.32	-0.22	-6.70
RSK 28 x DLM 2	66.87	25.12**	-26.93**	53.37**	2.34*	11.70**
RH 8814 x DLM 2	66.87	28.51**	-14.40**	7.71	2.52*	20.62**
NDR 8501 x DIRA 313	61.23	-12.10**	1.74	-45.07**	-7.43*	-30.48**
RK 8902 x DLM 2	61.23	12.19**	- 0.33	25.36**	2.87*	25.86**
RK 8902 x DIRA 313	61.23	- 2.77	9.18*	27.24**	-5.09**	-10.90**
PR 8903 x DLM 2	61.23	34.67**	-29.90**	58.09**	3.56**	8.75**
NDR 8501 x DLM 2	61.23	9.82**	-12.20	10.69	1.85	14.61**
PR 8903 x NDR 8501	50.49	-14.35**	9.35*	-33.12	- 3.01*	6.27**
PR 8903 x RK 8902	50.49	10.41**	-23.24**	19.65**	4.22**	4.64**
NDR 8501 x RK 8902	35.27	-11.12**	0.06	- 8.51	0.91	-0.26
RSK 28 x DIRA 313	23.72	12.30**	-10.38*	-13.73	3.30**	-12.80**
PR 8903 x DIRA 313	23.72	28.51**	-16.40**	8.69*	-4.76**	-5.58**
DIRA 313 x DLM 2	23.72	-15.84	10.86	- 2.15	5.64**	2.05

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**RECENT ADVANCES
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EUCARPIA CRUCIFERAE NEWSLETTER Nr. 23

Instructions to authors 2000

Deadlines : december 31st 2000

Next Cruciferae Newsletter Nr 23 will be produced and edited at the end of 2000. The editing group of Rennes will take charge but, as previously, it will be produced by direct photocopying the material you submit. Therefore, we should be grateful if you would, please, follow instructions below, from which depend the quality of your script.

- 1 – Contributions should be en English.
 - 2 – The scripts will not be retyped and must be produced with the best typing quality. Laser printing is expected with a clear black character.
 - 3 – As previously Contributions must not exceed **2 pages**, including tables and figures. Whole pages are preferred (eventually reduce the format through computer printing). Consecutive pages must be numbered in pencil. **Times 12** character is expected.
 - 4 – The heading of the paper must include the title (**1st line in bold letter**), followed by the authors names (lines below).
 - 5 – Single spacing is required. If not, it will be published at the editors discretion. The **A 4 format (21 x 29,7)** is strictly required with margins (not less than 3 cm on the left, 2 cm above and below). No other format will be accepted.
 - 6 – Tables and figures must be included in, or at the end of the text. Photographs cannot be included and reproduced.
 - 7 – Please do not fold the script and do not clip the pages together. Please send two copies of each protected by card.
- All invoices to :**

The Editor
Cruciferae Newsletter
INRA
Station d'Amélioration des Plantes
BP 29
F 35650 LE RHEU
FRANCE

All contributions must be sent before :

December 31st 2000 to the above address for edition in the next issue :
Cruciferae Newsletter Nr. 23



**RECENT ADVANCES
IN
OILSEED BRASSICAS**

**H.R. KALIA
S.K. GUPTA**

Oilseed Brassicas occupy a prominent third place ranking next only to Soybean and Groundnut.

It is, thus, evident that oil seed Brassicas will continue to uphold their prominence in the future as well.

However, the book deals with current trends in Brassicas research.

The book deals with current trends in Brassica research. The frontline scientists from different countries of the world have contributed chapters relative to their specialisations to pack in one volume for the benefit of students and researchers. The book includes topics such as importance, nomenclature and origin of Brassica and its allies, taxonomy based on conventional approach and restriction fragment length polymorphism (RFLP) analysis, wide hybridization and meiotic pairing, breeding objectives and methods, male sterility and its utilization in hybrid seed production recent advances in induction of male sterility by chimeric ribonuclease gene, chimeric LAT-59 and fms genes, mitochondrial DNA structure and reformation of Brassica mitochondrial genome and expression, molecular basis of cytoplasmic male sterility, bio-regulation of oil-filling in Brassica seeds etc. Recent developments as gene transfer and expression, microspore culture, and its importance in Brassica breeding research, polyamines in relation to abiotic stresses mark the distinctness of this book. Inclusion of topics like disease resistance, insect-pest resistance, shatter-resistance, oil and seed meal quality together with chemical and biological techniques to mitigate/eliminate deleterious effects of toxic seed contents make for the completeness of the book.

