BREEDING IMPROVEMENTS TOWARDS CANOLA QUALITY BRASSICA JUNCEA

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INTRODUCTION

The mustard species Brassica juncea Coss. is grown as a condiment crop on some 50,000 ha annually in the three prairie provinces of western Canada. Canadian condiment cultivars are higher yielding than cultivars of either of the two canola species B. napus and B. campestris (Woods, et al. 1991). They are early maturing and are also known to be more heat and drought tolerant, and more resistant to seed shattering than B. napus. They also have high resistance to blackleg. All these characteristics contribute directly to higher and more stable yields in mustard.

Despite these advantages, B. juncea is not grown as an edible oilseed crop in Canada because only Brassica oilseed cultivars that are low in erucic acid and glucosinolate content are suitable as edible oilseed crops. This paper reports on the results of breeding work designed to combine the zero erucic acid (Kirk and Oram 1981) and low glucosinolate traits (Love et al. 1990a) with high oil content and superior agronomic performance (high yield, early maturity and resistance to white rust) in a single strain of B. juncea. This work was conducted between 1985 and 1990 at the Agriculture Canada Research Station, Saskatoon, Saskatchewan.

PEDIGREE AND BREEDING

Parents

The strains 1058 and LDZ and the cultivar Cutlass were used as parents (Fig. 1).

Strain 1058, developed by Love et al. (1990a) from the interspecific cross [(B. juncea x B. campestris) x B. juncea], is a low glucosinolate B. juncea genotype. Seeds of strain 1058 contain less than 10 µmoles total of 3-butenyl and 2-hydroxy-3-butenyl glucosinolate. The seed is basically free of allyl glucosinolate and contains only trace amounts of 4-pentenyl and 2-hydroxy-4-pentenyl glucosinolates.

However, seed of strain 1058 is high in erucic acid, is very low in seed oil content (29% versus 42% for B. napus Westar in a field test in 1988), and has a brown colour. Plants of 1058 have very low fertility (low seed set) and are susceptible to white rust.

Figure 1. Pedigree of Brassica juncea canola mustard under development at the Agriculture Canada Research Station, Saskatoon, Sask.

Year	Pedigree	Purpose
1985	1058 x LDZ	incl. oil & intro. Z.E., inc. yield, imp. maturity, & int. WRR
1986	Cutlass x F_1	
	F_1	sel. het. E & GSL
1987	F_2	sel. low GSL
	F_3	sel. low GSL
	F_4	sel. WRR
1988	F_4 x LDZ	inc. oil
	F_1	prod. F_2
1989	F_2	sel. low GSL
	F_3	sel. Z.E., WRR
1990	F_4	sel. agr. type, high oil, low GSL

Abbreviations: inc.=increase; int.=introduce; sel.=select, prod.=produce; imp.=improve; Z.E.=zero erucic acid; het.E=heterozygous for erucic acid, GSL=glucosinolate, WRR=white rust resistance

Strain LDZ, developed at the Agriculture Canada Research Station, is a yellow-seeded zero erucic acid, high oil content strain selected from the cross [(Donskaja X Zem1) x Donskaja[3]]. It has high allyl glucosinolate content and is susceptible to white rust.

Cutlass, a B. juncea condiment mustard cultivar was developed at the Agriculture Canada Research Station at Saskatoon. This cultivar is well adapted to the climate of western Canada, it is high yielding and early maturing, and resistant to white rust. Seed of Cutlass is yellow in colour. Undesirable traits of Cutlass with respect to the development of an oilseed crop, are its high erucic acid and high allyl glucosinolate contents, and the low seed oil content.

Synthesis of first breeding population

Figure 1 depicts the crossing and selection scheme adopted for the development of canola quality B. juncea mustard in this study. Strain 1058 was crossed with strain LDZ and the F_1 generation of this cross with Cutlass. This 3-way cross combined genes for low glucosinolate content from 1058, with genes for zero erucic acid and high oil content from LDZ, and genes for high yield, early maturity and resistance to white rust from Cutlass in one breeding population.

Selection of intermediate parent

Because of the parents used in the 3-way cross, F_1 seeds were expected to be either homozygous (25%) or heterozygous (12%) for the erucic acid locus (Kirk and Hurlstone 1983). In order to maintain alleles for the zero erucic acid characteristics in the population, F_1 seeds were "half seeded" (Downey and Harvey 1963, Thies 1971) and only F_1 seeds containing 10 to 15% erucic acid grown.

It was expected also, as a result of the parents used, that seed of F_1 plants (F_2 seed) would contain either allyl or a mixture of allyl and 3-butenyl glucosinolate (Love et al. 1990b). The glucosinolate composition of vegetative tissue of F_1 plants was analyzed and used as a predictor for seed glucosinolate composition of seed from these plants (McGregor and Love 1987). One-half of the 162 F_1 plants contained allyl and 3-butenyl glucosinolates in their leaf tissue while the other half contained only allyl glucosinolate. Only F_1 plants that contained both allyl and 3-butenyl glucosinolate, in their leaves were grown to seed maturity and advanced since the presence of 3-butenyl glucosinolate was an indication that genes of the low glucosinolate strain 1058 were present in these plants thereby increasing chances for obtaining low glucosinolate plants in the F_2 generation.

Selections in the F_2 and F_3 generations concentrated on the reselection of low glucosinolate plants.

A total of 19862 F_2 plants were individually harvested from a field plot in 1987. Sixty-seven F_2 plants were classified as being low in glucosinolate content by "Glukotest" (Lein 1970). This very low frequency of low glucosinolate plants in the F_2 was not unexpected because of the fact that this material originated from an interspecific cross.

In the F_3 generation, plants were identified that had low contents of 3-butenyl glucosinolate (<20 µmoles per 1 g of meal) and were basically free of allyl glucosinolate. Glucosinolate analysis was conducted by gas chromatography using a modified version (J.P. Raney, unpubl. data) of the method of Daun and McGregor (1983).

Thus, selections of low glucosinolate plants in two subsequent generations (F_2 and F_3) were sufficient to reselect the low 3-butenyl glucosinolate characteristic of 1058 in progeny of the 3-way cross between 1058 and the two high allyl glucosinolate-containing parents, LDZ and Cutlass.

Plants of the F_4 generation, free of allyl glucosinolate and with low 3-butenyl glucosinolate contents, were selected for resistance to white rust under artificial inoculation conditions in the greenhouse. Because of the monogenic dominant inheritance of the white rust resistance trait (Tiwari et al. 1988), white rust resistant plants were easily reisolated from among low glucosinolate F_4 plants.

Synthesis of second breeding population

Since both the low glucosinolate strain 1058 and the condiment B. juncea cultivar Cutlass, used as parents in the synthesis of the first breeding population had low oil contents, it was concluded that the chances of selecting lines with oil contents similar to those of B. napus canola cultivars from the F_4 generation of the above 3-way cross were very low. Therefore, low glucosinolate, white rust resistant F_4 plants, selected from the first breeding population, were crossed with 3 zero erucic acid, high oil content lines selected out of LDZ, to create a second breeding population with a higher average oil content. The use of zero erucic acid LDZ lines as one parent made reselections of zero erucic acid plants from the F_4 generation (second parent) unnecessary.

F_2 seed production from F_1 plants of this cross completed the synthesis of the second breeding population.

Selection of experimental strains

As in the first breeding population, reselection of low glucosinolate plants in the F_2 generation had the highest priority because of the uncertainties of reselecting such plants from the cross with LDZ, an allyl glucosinolate-containing parent. A total of 12700 F_2 plants was individually harvested from a field plot in 1989. Three hundred F_2 plants had low "Glukotest" ratings. These 300 plants were analyzed for glucosinolate content by gas chromatography and 25 F_2 plants containing less than 40 µmoles of glucosinolates per 1 g meal identified. The frequency of low glucosinolate plants in this generation was again very low indicating the presence of meiotic abnormalities and incomplete incorporation of the low glucosinolate characteristic into the B. juncea genome.

F_3 seed of low glucosinolate F_2 plants was analyzed for fatty acid composition by the half seed method using gas chromatography. Zero erucic acid half seeds were found at high frequencies; they were planted in the greenhouse and selected for resistance to white rust. White rust resistant plants were present at high frequencies among zero erucic acid plants. 480 zero erucic acid, white rust resistant plants were identified in 12 of the 25 F_3 families. Selfed seed was produced from these 480 plants for agronomic and quality evaluation in 1990.

PERFORMANCE

The 480 low glucosinolate. zero erucic acid, white rust resistant F_3 plants, were field evaluated for agronomic performance and seed quality in a 3-replicate, 3m single row nursery (F_4 rows) in 1990 at Saskatoon. Rows were rated for agronomic type, individually harvested with a plot combine and the seed analyzed for oil content by nuclear magnetic resonance and for glucosinolate content by gas chromatography.

Field observations indicated that the material was, on average, one week earlier maturing than B. napus Westar (data not shown). Average oil contents of the 12 families varied greatly from a low content of 37.3% for family H4 to a high content of 40.6% for family H5 (Table 1). Seed of all families contained allyl and 3-butenyl glucosinolate and only trace amounts of the other alkenyl glucosinolates (2-hydroxy-3-butenyl, 4-pentenyl, and 2-hydroxy-4-pentenyl).

Table 1. Average agronomic performance and oil and glucosinolate content of 12 F_3 families in a field test at Saskatoon in 1990.

F_3 family	No. of F_4 rows	Agr. (1-5)[1]	Oil (%)	Glucosinolates, µmoles g^{-1} meal allyl	3-butenyl	others
H1	26	3.3	40.5	20.6	35.4	5.2
H2	127	3.7	39.8	27.8	16.5	2.0
H3	46	3.1	38.9	38.7	13.5	.9
H4	36	3.5	37.3	22.1	45.8	4.8
H5	117	4.3	40.6	21.4	15.8	2.6
H6	30	4.4	39.4	23.7	18.8	1.7
H9	29	4.2	37.6	20.6	39.3	10.7
H10	12	4.1	37.4	22.7	38.9	3.1
H11	8	3.7	38.6	28.9	22.8	2.7
H12	15	3.7	39.5	23.3	23.8	3.3
H13	13	3.7	38.3	33.0	17.1	2.3
H14	21	4.1	37.7	6.2	31.7	4.5

[1]Agr. 1=poor, 5=best

The F_3 family H5 segregated F_4 lines that combined superior agronomic performance with high oil content and less than 30 µmoles (per 1 g of meal) of aliphatic glucosinolates (Table 2). These lines are also low in erucic acid and are yellow seeded; they also carry genes for white rust resistance.

Replicated yield testing of 25 selected lines at four locations in Saskatchewan in 1991 will provide quantitative data on agronomic performance and quality of these lines.

Table 2. Agronomic performance and oil and glucosinolate content of 5 selected F_4 lines from family H5, average for H5 and average for B. napus Westar in a field test at Saskatoon in 1990.

Entry	Agr. (1-5)[†]	Oil (%)	Glucosinolates, µmoles g^{-1} meal allyl	3-butenyl	others
J90-3392	4.3	42.2	12.0	9.3	1.8
J90-3430	5.0	42.4	11.4	9.7	2.8
J90-3435	4.0	42.4	11.3	11.2	2.0
J90-3447	4.3	42.8	12.9	10.5	1.8
J90-3450	4.7	42.0	10.5	7.5	1.6
Average H5	4.3	40.6	21.4	15.8	2.6
B. napus Westar	4.0	42.6		4.5	9.5

[†]Agr. 1=poor, 5=best

CONCLUSION

The results of the breeding work indicate that canola quality Brassica juncea strains with improved agronomic performance and high oil content can be developed through cross-breeding and selection utilizing existing germplasm resources.

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