Identification and evaluation of flea beetle (*Phyllotreta cruciferae*) resistance within Brassicaceae

J. E. Gavloski[1], U. Ekuere[2], A. Keddie[2], L. Dosdall[2], L. Kott[3], and A. G. Good[2,4]

[1]*Manitoba Agriculture and Food, Soils and Crops Branch, Box 1149, Carman, Manitoba, Canada R0G 0J0;* [2]*Department of Biological Sciences, University of Alberta, Edmonton, Alberta, Canada T6G 2E9;* [3]*Department of Crop Science, University of Guelph, Guelph, Ontario, Canada N1G 2W1.*
Received 1 December 1999, accepted 14 June 2000.

Gavloski, J. E., Ekuere, U., Keddie, A., Dosdall, L., Kott, L. and Good, A. G. 2000. **Identification and evaluation of flea beetle (*Phyllotreta cruciferae*) resistance within Brassicaceae.** Can. J. Plant Sci. **80**: 881–887. All currently registered varieties of canola/oilseed rape, *Brassica napus* and *B. rapa*, are susceptible to attack by flea beetles, although to varying degrees. The development of resistant cultivars would be an environmentally acceptable means to reduce the damage caused by flea beetles. Seedlings from 10 species of Brassicaceae were evaluated for levels of antixenosis resistance to flea beetles in the laboratory, along with 308 *Sinapis alba/B. napus* hybrids. *Thlaspi arvense* and 11 cultivars of *S. alba* were resistant to feeding by flea beetles. In addition, 34 *S. alba/B. napus* hybrids were resistant to feeding by flea beetle in at least one test, although many of these failed to demonstrate resistance with repeated testing. One hybrid line was resistant to feeding by flea beetles each of the four times it was tested, while another was resistant in three out of four tests. These data indicate that resistance to flea beetles within the Brassicaceae is a genetic trait and can be transferred by interspecific hybridization. This information is the first step towards introgression of genetic sources of flea beetle resistance from resistant relatives into canola varieties.

Key words: Flea beetles, *Phyllotreta cruciferae*, *Brassica*, resistance, antixenosis, introgression

Gavloski, J. E., Ekuere, U., Keddie, A., Dosdall, L., Kott, L. et Good, A. G. 2000. **Identification et évaluation de la résistance à l'altise (*Phyllotreta cruciferae*) chez les Brassicacées**. Can. J. Plant Sci. **80**: 881–887. Toutes les variétés actuellement homologuées de colza (et navettes) oléagineux de type canola, *Brassica napus* et *B. rapa*, sont, à plus ou moins fort degré, sensibles aux attaques de l'altise. La sélection de cultivars résistants serait un moyen écologiquement convivial de réduire les endommagements causés par cet insecte. Nous avons recherché en laboratoire des signes de résistance par antixénose à l'altise sur des jeunes plants appartenant à dix espèces de Brassicacées ainsi qu'à 308 hybrides *Sinapis alba* × *B. napus*. *Thlaspi arvense* (le tabouret des champs) et onze cultivars de *S. alba* manifestaient de la résistance aux déprédations de l'altise. Trente-quatre des hybrides moutarde × soja se montraient résistants dans un moins un essai, mais pour plusieurs d'entre eux cette résistance ne s'observait pas de façon régulière. Une lignée hybride était résistante dans quatre essais, tandis qu'une autre ne l'était que dans trois essais sur quatre. Ces observations montrent que la résistance à l'altise chez les Brassicacées est un caractère génétique transférable par hybridation interspécifique. Elles constitue, par ailleurs, la première étape vers l'introgression de source génétique de résistance à l'altise dans les variétés de colza canola à partir d'espèces apparentées de Brassicacées.

Mots clés: Altise, *Phyllotreta cruciferae*, *Brassica*, résistance, antixenose, introgression

The crucifer flea beetle, *Phyllotreta cruciferae* (Goeze), and the striped flea beetle, *Phyllotreta striolata* (F.), are the most important chronic pests of canola/oilseed rape, *B. napus* L. and *B. rapa* L., in Canada (Lamb 1989). They are oligophagous herbivores that feed primarily on plants in the family Brassicaceae (Cruciferae). Adult flea beetles feed on cotyledons and slender stems of seedling cruciferous plants, and continue to attack the leaves as the plant develops (Feeny et al. 1970). The typical feeding damage of flea beetles to plants consists of small holes or pits in the epidermis of leaves. Although initial feeding does not penetrate the leaf completely, tissues below the injury eventually dry up and break or fall out giving a shot-hole appearance (Westdal and Romanow 1972; Brandt and Lamb 1993). Flea beetle damage to canola has been estimated to cause an average annual yield loss of about 10% (Lamb and Turnock 1982). The damage caused by this insect has been managed primarily with applications of chemical insecticides. The financial cost and negative impacts of chemical control on the environment emphasizes the need to develop *Brassica* cultivars with enhanced genetic insect resistance.

One useful component of any integrated pest management program is resistant cultivars. Resistance can be due to antixenosis (nonpreference), antibiosis, and tolerance or by some combination of these (Painter 1951). The level of antibiosis to flea beetles seems to vary less among *Brassica* species and related plants than does the level of antixenosis (Palaniswamy et al. 1997). Antixenosis is thus a more promising type of resistance for use against flea beetles in

[4]*To whom correspondence should be addressed (e-mail: allen.good@ualberta.ca).*

canola. A laboratory method to screen crucifer seedlings for antixenosis-based resistance to flea beetles was described by Palaniswamy et al. (1992). Using this method, no significant antixenosis was found among 19 cultivars of *B. napus* L. and *B. campestris* L.; however one accession of *B. carinata* L. and two accessions of *S. alba* L. exhibited antixenosis (Palaniswamy et al. 1992).

Species of Brassicaceae and cultivars of oilseed rape differ in their susceptibilities to attack by flea beetles (Lamb 1980, 1984, 1988, Lamb and Palaniswamy 1990; Bodnaryk and Lamb 1991). Seedlings of yellow mustard, *S. alba* 'Ochre', have a high level of resistance to feeding by flea beetles (Bodnaryk and Lamb 1991). Antixenosis and tolerance are two of the mechanisms responsible for this resistance. Cotyledons of *S. alba* may lose some of their antixenotic properties as they age (Bodnaryk and Lamb 1991; Palaniswamy and Lamb 1992). *Thlaspi arvense* and *Lunaria annua* are largely unacceptable to *P. striolata* because of the feeding deterrents they possess (Meisner and Mitchell 1983), whereas resistance to feeding by *P. cruciferae* in false flax, *Camelina sativa*, appears due to the absence of cues that initiate feeding (Palaniswamy et al. 1998). Leaves of *B. villosa* Biv. and *B. villosa* Biv. subsp. *drepanensis*, which contain trichome densities of >2172 cm^{-2}, are also highly resistant to flea beetle feeding (Palaniswamy and Bodnaryk 1994). The high density of trichomes acts as a physical barrier to flea beetle feeding by preventing the flea beetles from firmly settling on the leaf surface to initiate feeding. Feeding preferences may change with leaf type or growth stage, as well as with host species. True leaves of *B. oleracea* were less preferred by *P. striolata* than those of *S. arvensis*, whereas the opposite was observed for cotyledons (Palaniswamy and Lamb 1992).

In this study we screened many genotypes of Brassicaceae to identify sources of antixenotic resistance to flea beetles. Doubled haploid (DH) interspecific hybrids derived from *S. alba* × *B. napus* crosses were evaluated to determine their levels of antixenotic resistance to flea beetles. A genomic slot-blot hybridization analysis (Anamthawat-Johnson et al. 1990; Besse et al. 1997) was used to confirm that these interspecific hybrids contained portions of the *S. alba* genome. These procedures comprise the first steps towards introgression of flea beetle resistance from resistant relatives into canola varieties.

MATERIALS AND METHODS

Plant Material

All plant materials used in this study were obtained from the University of Alberta germplasm collection, except the interspecific hybrids (*S. alba* × *B. napus*//*B. napus* 3×), which were produced at the University of Guelph, Ontario, using embryo rescue techniques (Ripley and Arnison 1990).

Screening for Antixenosis Resistance to Flea Beetles

Flea Beetle Collection

Adult crucifer flea beetles, *P. cruciferae*, were collected from field populations from May to August each year near Vegreville, Kitscoty, and Edmonton, Alberta, using allylisothiocyanate-baited traps (Burgess and Wiens 1980), hand aspiration, or a D-vac vacuum insect sampler. Beetles were held in screen cages in a growth cabinet at 18°C (day) and 15°C (night) with a 16L:8D photoperiod. Beetles were fed canola, and occasionally cabbage leaves, and had access to a honey solution through dental wicks protruding from a reservoir. Before each screening, beetles were starved for 24 h and had access only to the honey solution.

Seedling Preparation

Seedlings were grown individually in a greenhouse in 3 cm by 5 cm plastic vials filled with Redi-mix (Apache Seeds, Edmonton, AB). Vials were held in greenhouse flats and watered by sub-irrigation. The bottom of each vial was punctured to facilitate watering. Seven-day-old seedlings (cotyledon stage) selected to be roughly at the same growth stage were used in the screenings.

Screening

Screening was done in Plexiglas arenas with nylon-screen tops essentially as described by Palaniswamy et al. (1992). Arenas were 35 cm tall and 43 × 43 cm square, with a plastic foam base (43 × 43 cm square and 2.5 cm thick) supported on a Plexiglas ridge about 6 cm from the bottom of the arena. The base had 100 holes (3 cm diameter) evenly spaced in a 10 × 10 layout with a spacing of 0.8 cm between holes. Vials containing seedlings were inserted into the holes so that vial rims were even with the foam surface. Holes were numbered sequentially from 0 to 99, producing a 10 × 10 layout of 10 rows and 10 columns. Ten plants of 10 lines were placed in the arena as a 10 by 10 Latin square design (i.e., each entry appeared once in each row and each column in random order). *Brassica napus* 'Quantum' was used in each test as a standard against which the level of resistance for the nine other lines was compared. This enabled data screened in different tests to be compared. Quantum was selected to be the standard since it is a commonly grown cultivar of canola. For ease of identification, a small mark was placed in vials containing Quantum. Approximately 1000 beetles were introduced to the arena through an opening on the front of the arena, and the opening was then sealed with a plastic cover. The beetles were distributed as evenly as possible and additional beetles were added as needed to maintain populations near 1000.

Feeding damage to each cotyledon was rated visually using a scale from 0 (no damage) to 10 (cotyledon destroyed); thus, a rating of 1 was assigned if approximately 10% of the area of the cotyledon was damaged (Palaniswamy et al. 1992). Each cotyledon was rated separately and then the values for the two cotyledons were added and multiplied by 5 to estimate the percentage of the foliage area of the whole seedling that was damaged. The beetles sometimes fed on petioles and stems, which caused death of cotyledons even if the cotyledon surface had not been fed upon. Regardless of the amount of damage to the cotyledon, a rating of 10 was assigned when the petiole was cut and the cotyledon dropped off or wilted beyond recovery. Both cotyledons were given a rating of 10 when the stem was cut

Table 1. Mean damage ratings of feeding by the crucifer flea beetle (*Phyllotreta cruciferae*) on canola cultivars of *Brassica napus* and *Brassica rapa*

				1998[z]					
		1996		Round 1		Round 2		Round 3	
Species	Cultivar	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]
B. napus	Bronowski	39	N	42	S	57	N		
B. napus	Cresor					39	N	36	N
B. napus	Cyclone	45	N			44	N		
B. napus	Delta	51	N	50	N	63	N	47	N
B. napus	Westar	47	N	45	N	40	N		
B. napus	Zephyr	80	S			37	R		
B. napus	Altex			61	N	55	N		
B. napus	Pivot	64	N						
B. napus	Stellar	51	N						
B. napus	Torch	49	N						
B. rapa	Horizon	67	S	42	N	52	N		
B. rapa	C8711			77	S	41	N		
B. rapa	AC Sunshine	46	N						
B. rapa	Chinensis	41	N						
B. rapa	Echo	71	N						
B. rapa	Eclipse	53	N						
B. rapa	Reward	63	N						
B. rapa	Tobin	41	N						

[z]Three rounds of testing (each round including trials in two cages) were performed in 1998.
[y]Damage rating relative to the control cultivar (*B. napus* 'Quantum') (see Materials and Methods for details).
[x]Significance rating based on Duncan's new multiple range test ($P < 0.05$). R = resistant, S = susceptible, and N = not significantly different from the control cultivar (*B. napus* 'Quantum').

and the seedling felled. The amount of damage to Quantum (the standard) was rated regularly. Once damage to Quantum was approximately 50%, damage ratings were taken for all plants and the test ended.

All tests were conducted in a room at 23 ± 1°C with light cycle of 16L:8D. Because flea beetles are sensitive to light and tend to move to the side of the arena with the highest light intensity, diffuser panels were placed below the overhead fluorescent lighting and cages were situated so that light was as uniform as possible on the cage (Palaniswamy et al. 1992).

Analysis of Data

Each group of 10 lines was tested in two arenas, resulting in 20 replications per line. Values for percent damage were analyzed. Data were analyzed as a Latin square design using analysis of variance (SAS Institute, Inc. 1985). Duncan's new multiple range test ($P < 0.05$) was used to separate the means. Analysis was run for the pooled data of the two arenas, and also separately by arena. Results presented and analyzed here are from pooled data.

It was not always possible to stop the tests at exactly 50% damage to Quantum and to correct for this and to make the results comparable between the lines in different tests, the mean score for every individual line was corrected relative to Quantum damage at 50% (D) using the following equation:

$$D = DM \times 50/QD$$

where DM (%) = mean % damage of an individual line, except Quantum, and QD (%) = mean % damage for Quantum. Data was adjusted in this way after first being analyzed.

Slot-blot Analysis

To determine the amount of *S. alba* DNA in selected hybrids, DNA extraction and quantification were carried out according to Sharpe et al. (1995). Ten nanograms of DNA from each of six lines (listed in Table 3) were denatured by 0.4 M NaOH and then loaded per slot to a Hybond N^+ nylon membrane (Amersham Pharmacia Biotech, Buckinghamshire, UK), under vacuum using a manifold II slot apparatus as described by the manufacturer (Schleicher and Schuell, Dassel, Germany). Membranes were neutralized by incubation in 2× SSC (0.3 M NaCl plus 0.03 M sodium citrate) at room temperature.

Sinapis alba 'Kirby' and *B. napus* 'Westar' DNA were used as standards as well as artificial hybrids. The artificial hybrids consisted of a mixture of genomic DNA from the two species, in the ratios; 5:5, 6:4, 8:2, 9:1, 9.5:0.5, 9.9:0.01 (to give a final concentration of 10 ng of DNA mixture per artificial hybrid).

Hybridization of slot-blot membranes was carried out essentially as described by Sharpe et al. (1995) except that blocking DNA also consisted of 250 μg of denatured *B. napus* 'Westar' DNA and the labeled probe was 60 ng of denatured *S. alba* 'Kirby' DNA. The radioactivity on the slot-blot membrane was measured using a radioactivity scanner (PhosphoImager 445 SI, Molecular Dynamics, Sunnyvale, CA) and signal intensity was quantified using ImageQuant (Molecular Dynamics).

RESULTS

No consistent resistance to flea beetles was detected in any cultivars of *B. napus* or *B. rapa* relative to the control cultivar, *B. napus* 'Quantum' (Table 1). *Brassica napus* 'Cresor' had the lowest rating among *B. napus* cultivars, although it

Table 2. Mean damage ratings of feeding by the crucifer flea beetle (*Phyllotreta cruciferae*) on various genotypes of cruciferous plants

						1998[z]					
		1996		1997		Round 1		Round 2		Round 3	
Species	Cultivar	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]
B. juncea	Blaze					59	N	59	S		
B. juncea	Cutlass	57	N			77	S	45	N		
B. juncea	Domo	48	N			46	N	40	N		
B. oleracea	Dwarf Green					41	N	45	N		
B. oleracea	Calabrese					56	N	57	N		
B. oleracea	Snowball T3					68	S	74	S	84	S
B. oleracea	Danish Ballhead					65	N				
B. oleracea	Copenhagen Market					30	R	54	N		
B. oleracea	Long Island					44	N				
B. oleracea	Purple					65	S	56	N		
B. oleracea	Forage	67	S								
B. tournefortii						54	N	33	N		
B. villosa						64	N				
S. alba	Albatros					30	R	20	R		
S. alba	Arda					30	R	30	R	41	R
S. alba	Emergo	41	N			28	R	18	R		
S. alba	Kirby	4	R			40	R				
S. alba	Gisilba					26	R	27	R		
S. alba	Kreta					26	R	22	R		
S. alba	Lethbridge22					33	R	34	R		
S. alba	Ochre	4	R			29	R	27	R		
S. alba	Stona					38	R	28	R		
S. alba	Tilney	30	N			28	R	22	R		
S. alba	Trico							29	R	21	R
S. pubescens						70	S				
Thlaspi arvense		2	R	0	R	0	R				
Crambe abyssinica				45	N	9	R				

[z]Three rounds of testing (each round including trials in two cages) were performed in 1998.
[y]Damage rating relative to the control cultivar (*B. napus* 'Quantum') (see Materials and Methods for details).
[x]Significance rating based on Duncan's new multiple range test ($P < 0.05$). R = resistant, S = susceptible, and N = not significantly different from the control cultivar (*B. napus* 'Quantum').

was not significantly different from the control cultivar. *Brassica napus* 'Zephyr' was rated as resistant to feeding by flea beetles in a 1998 test, but previously rated susceptible in a 1996 test.

No resistance was found in the *B. juncea* cultivars screened (Table 2). *Brassica oleracea* 'Copenhagen Market' was resistant to flea beetle feeding in the first round of testing in 1998, but not in the second round of testing in 1998. *Brassica oleracea* 'Snowball T3' was consistently susceptible to flea beetle feeding in three rounds of testing in 1998. *Brassicsa tournefortii* and *B. villosa* lines screened had no resistance to flea beetle feeding.

Cultivars of *S. alba* were consistently resistant to feeding by flea beetles (Table 2). All of the 26 ratings for the 11 *S. alba* cultivars were below 50, with 17 of the 26 ratings being between 20 and 30. *Sinapis alba* 'Kirby' and *S. alba* 'Ochre' rated as low as 4 in the 1996 tests. The *Sinapis alba* cultivars Emergo and Tilney were not resistant to flea beetle feeding in the 1996 screening. However, both cultivars were resistant in each of two rounds of testing in 1998. The reason for this difference in results is not known. Another species of *Sinapis*, *S. pubescens* was susceptible to flea beetle feeding. *Thlaspi arvense* was very resistant to feeding by flea beetles and had the lowest feeding damage rating amongst all the lines screened. *Crambe abyssinica* had a high level of resistance in a 1998 test, but not in a 1997 test.

Three hundred and eight hybrid lines were tested for resistance to feeding by flea beetles and 34 of these lines were resistant in at least one test. Hybrid lines with the greatest resistance and susceptibility to feeding by flea beetles are shown in Table 3. BNH-006 exhibited consistent resistance to feeding by flea beetles, each of the four times it was tested over a 3-yr period. The lowest damage rating for the resistant hybrids was in BNH-524, with a rating of 23 in a 1997 test. This hybrid had a higher damage rating, 48, in a 1996 test, however. BNH-518, BNH-525, and BNH-532 all had resistance ratings below 40 both times each of these hybrids were tested. Each of these hybrids rated as resistant to feeding by flea beetles in one of the tests, and not resistant relative to the control cultivar in the other test. BNH-574 was resistant to feeding by flea beetles three of the four times it was tested, with resistance ratings ranging from 26 to 41. The highest susceptible rating observed was for BNH-448, with a rating of 101. A rating greater than 100 happened because damage ratings were adjusted so the control cultivar, *B. napus* 'Quantum', would always have a rating of 50 in each test.

Six of the 308 hybrid lines screened for flea beetle resistance were analyzed for amount of *S. alba* DNA content. These lines were selected to represent the three phenotypic classes observed from the feeding experiments (resistant, susceptible and not significantly different from the control

Table 3. Mean damage ratings of feeding by the crucifer flea beetle (*Phyllotreta cruciferae*) on 19 of the 308 *S. alba* × *B. napus* hybrids that had the greatest resistance or susceptibility to feeding by flea beetles, and *S. alba* DNA content in six of these hybrids

					1998[z]						
	1996		1997		Round 1		Round 2		Round 3		% *S. alba*
Hybrid	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	D[y]	S[x]	DNA[w]
BNH-006	36	R	34	R	33	R	32	R			0.35
BNH-094			31	R	54	N					
BNH-138	60	N	40	R	54	N	57	N	57	N	
BNH-139	31	R			43	N	33	N			
BNH-197			38	R	58	N	57	N			
BNH-366	59	N	29	R							
BNH-418			35	R			48	N	45	N	
BNH-518			34	R	38	N					
BNH-524	48	N	23	R							
BNH-525	35	R	39	N							
BNH-532	38	N	35	R							
BNH-574	38	R	26	R	41	N	39	R			0.33
BNH-106	92	S									0.28
BNH-124			74	S							
BNH-132	74	S	51	S	65	S					0.37
BNH-161	87	S									
BNH-448	101	S									
BNH-014	59	N			53	N	56	N			0.42
BNH-015	65	N			53	N	42	N			0.28

[z]Three rounds of testing (each round including trials in two cages) were performed in 1998.
[y]Damage rating relative to the control cultivar (*B. napus* 'Quantum') (see Materials and Methods for details).
[x]Significance rating based on Duncan's new multiple range test ($P < 0.05$). R = resistant, S = susceptible, and N = not significantly different from the control cultivar (*B. napus* 'Quantum').
[w]*S. alba* DNA content values were calculated based on the assumption of an exponential relationship between radioactive signal and *S. alba* DNA content (Slope = 0.02138). Numbers are means of two replicates.

cultivar). *Sinapis alba* DNA content in these lines was low and varied between 0.28 to 0.42% (Table 3). The level of *S. alba* DNA content detected in the six hybrid lines was similar. Thus, there is no apparent relationship between the amount of *S. alba* DNA content and resistance to feeding by flea beetles among the hybrid lines that were tested.

DISCUSSION

This approach allows for rapid screening of canola cultivars for resistance to flea beetles. However, there was often large variation in damage ratings between tests of individual lines. This may be because the lines a particular genotype was tested against varied from test to test. For example, a line with a low level of resistance would appear more resistant when tested with a group of lines that are highly susceptible to the beetles, than when tested with a group of lines more resistant to the beetles. Another factor that may have contributed to the variation in damage ratings of the same line among tests is that the tests were conducted over the summer. Summer feeding flea beetles often feed less voraciously and less consistently than do flea beetles that have overwintered. Thus, multiple rounds of screening are advisable using this methodology to increase confidence in the results.

The lack of resistance in *B. napus* and *B. rapa* to feeding by flea beetles is consistent with the results of previous studies. Palaniswamy et al. (1992) found no significant antixenosis among 19 cultivars of *B. napus* and *B. rapa* when tested against the standard entries *B. napus* 'Westar' and *B. rapa* 'Tobin'. The same study also found no significant antixenosis in *B. juncea*, which is consistent with the present study.

Brassica villosa was not resistant to feeding by flea beetles in this study. Previous reports, however, have shown that the upper leaves of 6- to 8-wk-old *B. villosa* plants are highly resistant to feeding by flea beetles (Palaniswamy and Bodnaryk 1994). A high density of trichomes on *B. villosa* acted as a physical barrier to feeding by flea beetles by preventing them from firmly settling on the leaf surface to initiate feeding. The seedlings of *B. villosa* used in the present experiment lacked trichomes, in contrast to older plants, and thus failed to give the resistance described by Palaniswamy and Bodnaryk (1994). Since flea beetles tend to be most destructive to canola/oilseed rape when the crop is in the seedling stage, trichome-based resistance would be of most economical importance if it could be bred into the crop and expressed at an early growth stage.

The higher level of resistance to flea beetle feeding in *S. alba* relative to *B. napus* and *B. rapa* is consistent with previous findings. Damage scores for *S. alba* were often half those of the *B. napus* check, which is consistent with previous studies (Bodnaryk and Lamb 1991; Palaniswamy and Lamb 1992; Brandt and Lamb 1993). *Sinapis alba* lines are tolerant of low levels of flea beetle feeding damage to their cotyledons, and show antixenosis in tests with the susceptible species, *B. napus* 'Westar' (Bodnaryk and Lamb 1991). Cotyledons of *S. alba* may lose their antixenotic properties as they age, however (Bodnaryk and Lamb 1991; Palaniswamy and Lamb 1992). The cotyledons of *S. alba*

contain high concentrations of sinalbin, which does not occur in *B. napus* (Bodnaryk 1991), and which may be responsible for the antixenosis exhibited by *S. alba* (Bodnaryk and Lamb 1991).

Stinkweed, *T. arvense* L., was shown in this study to have a high level of antixenosis to flea beetles. Palaniswamy et al. (1997) also demonstrated a high level of antixenosis in *T. arvense* to flea beetles, but found that antixenosis decreased if foliage was excised or the plant was wilted.

The hybrids BNH-106, BNH-014, and BNH-015 rated as susceptible, not resistant, and not resistant, respectively, to feeding by flea beetles. These three hybrids all have antixenotic resistance to feeding by root maggots (*Delia* spp.) (Dosdall et al. 2000). This demonstrates that resistance against root maggots does not affect flea beetles.

Potential sources of resistance to feeding damage by flea beetles were found in some *S. alba*/*B. napus* hybrids that were tested. The low level of detectable *S. alba* DNA content in the hybrid lines reflects the enrichment for the recurrent *B. napus* genotype background, to which the initial F_1 hybrids (from the *S. alba* × *B. napus*) were backcrossed for several generations. The genomic slot blot hybridization technique enhances the ability to detect genomic introgressions not detectable by conventional morphological procedures used in breeding programs (Besse et al. 1997). Although the six hybrid lines analyzed for *S. alba* DNA content showed differences in flea beetle damage, these differences were not associated with *S. alba* DNA content. This indicates that portions/segments of the *S. alba* genome retained in these hybrid plants were different for the three phenotypic classes.

The data from these lines further suggest that the different DH hybrid lines have inherited different segments of the *S. alba* genome. These findings provide useful material for finding the genetic locations of these *S. alba* introgressed segments and ultimately the genes from *S. alba* responsible for conferring resistance against flea beetle and other insect pests. There are reports of other successful introgressions of useful traits from wild/distant relatives into crop species such as the introgression of the Ogura cytoplasmic male sterility (a maternally inherited gene) from a fodder radish cytoplasm into *B. napus* (Renard et al. 1992; Stiewe et al. 1994).

This research confirms the high level of resistance to feeding by flea beetles in *S. alba* and *T. arvense*. The majority of hybrid lines tested in this study demonstrated no useful resistance to feeding by flea beetles. However, some hybrid lines did demonstrate resistance in these laboratory tests, and are worth further evaluation as potential sources for increasing flea beetle resistance in commercial cultivars of canola with good agronomic traits.

ACKNOWLEDGMENTS

We thank Jim Tansey, Mike McFarland, Kim Zeleny and Lesley Davis for excellent technical support and Dr. R. Lamb for advice and discussions. Funding was provided by the Natural Sciences and Engineering Research Council of Canada (NSERC), Alberta Agriculture Research Institute (AARI), Alberta Wheat Pool (AWP) and Zeneca Seeds.

Anamthawat-Johnson, K., Schwarzacher, T., Leitch, A. R., Bennett, M. D. and Heslop-Harrison, J. S. 1990. Discrimination between closely related Triticeae species using genomic DNA as a probe. Theor. Appl. Genet. **79**: 721–728.

Besse, P., McIntyre, C. L., Burner, D. M. and de Ameida, C. G. 1997. Using slot blot hybridization to assess intergeneric *Saccharum* × *Erianthus* hybrids (Andropogoneae – Saccarinae). Genome **40**: 428–432.

Bodnaryk, R. P. 1991. Developmental profile of sinalbin (ρ-hydroxybenzyl glucosinolate) in mustard seedlings, *Sinapis alba* L., and its relationship to insect resistance. J. Chem. Ecol. **17**: 1543–1556.

Bodnaryk, R. P. and Lamb, R. J. 1991. Mechanisms of resistance to the flea beetle, *Phyllotreta cruciferae* (Goeze), in mustard seedlings, *Sinapis alba* L. Can. J. Plant Sci. **71**: 13–20.

Brandt, R. N. and Lamb, R. J. 1993. Distribution of feeding damage by *Phyllotreta cruciferae* (Goeze) (Coleoptera: Chrysomelidae) on oilseed rape and mustard seedlings in relation to crop resistance. Can. Entomol. **125**: 1011–1021.

Burgess, L. and Wiens, J. E. 1980. Dispensing allyl-isothiocyanate as an attractant for trapping crucifer-feeding flea beetles. Can. Entomol. **112**: 93–97.

Dosdall, L. M., Good, A., Keddie, B. A., Ekuere, U. and Stringam, G. 2000. Identification and evaluation of root maggot (*Delia* spp.) (Diptera: Anthomyiidae) resistance within Brassicaceae. Crop Protect. **19**: 247–253.

Feeny, P., Paauwe, K. L. and Demong, N. J. 1970. Flea beetles and mustard oils: host plant specificity of *Phyllotreta cruciferae* and *P. striolata* adults (Coleoptera: Chrysomelidae). Ann. Entomol. Soc. Am. **63**: 832–841.

Lamb, R. J. 1980. Hairs protect pods of mustard (*Brassica hirta* 'Gisilba') from flea beetle feeding damage. Can. J. Plant Sci. **60**: 1439–1440.

Lamb, R. J. 1984. Effects of flea beetles, *Phyllotreta* spp. (Chrysomelidae: Coleoptera), on the survival, growth, seed yield and quality of canola, rape and yellow mustard. Can. Entomol. **116**: 269–280.

Lamb, R. J. 1988. Assessing the susceptibility of crucifer seedlings to flea beetle (*Phyllotreta* spp.) damage. Can. J. Plant Sci. **68**: 85–93.

Lamb, R. J. 1989. Entomology of oilseed *Brassica* crops. Ann. Rev. Entomol. **34**: 211–229.

Lamb, R. J. and Palaniswamy, P. 1990. Host discrimination by a crucifer-feeding flea beetle, *Phyllotreta striolata* (F.) (Coleoptera: Chrysomelidae). Can Entomol. **122**: 817–824.

Lamb, R. J. and Turnock, W. J. 1982. Economics of insecticidal control of flea beetles (Coleoptera: Chrysomelidae) attacking rape in Canada. Can. Entomol. **114**: 827–840.

Meisner, J. and Mitchell, B. K. 1983. Phagodeterrency induced by two cruciferous plants in adults of the flea beetle *Phyllotreta striolata* (Coleoptera: Chrysomelidae). Can Entomol. **115**: 1209–1214.

Painter, R. J. 1951. Insect resistance in crop plants. University of Kansas, Lawrence, KS.

Palaniswamy, P. and Bodnaryk, R. P. 1994. A wild *Brassica* from Sicily provides trichome-based resistance against flea beetles, *Phyllotreta cruciferae* (Goeze) (Coleoptera: Chrysomelidae). Can. Entomol. **126**: 1119–1130.

Palaniswamy, P. and Lamb, R. J. 1992. Host preferences of the flea beetles *Phyllotreta cruciferae* and *P. striolata* (Coleoptera: Chrysomelidae) for crucifer seedlings. J. Econ. Entomol. **85**: 743–752.

Palaniswamy, P., Lamb, R. J. and Bodnaryk, R. P. 1997. Antibiosis of preferred and non-preferred host-plants for the flea

beetle, *Phyllotreta cruciferae* (Goeze) (Coleoptera: Chrysomelidae). Can. Entomol. **129**: 43–49.

Palaniswamy, P., Lamb, R. J. and Bodnaryk, R. P. 1998. Resistance to the flea beetle *Phyllotreta cruciferae* (Coleoptera: Chrysomelidae) in false flax, *Camelina sativa* (Brassicaceae). Can. Entomol. **130**: 235–240.

Palaniswamy, P., Lamb, R. J. and McVetty, P. B. E. 1992. Screening for antixenosis resistance to flea beetles, *Phyllotreta cruciferae* (Goeze) (Coleoptera: Chrysomelidae), in rapeseed and related crucifers. Can. Entomol. **124**: 895–906.

Renard, M., Delourme, R., Mesquida, J., Pelletier, G., Primard, C., Boulidard, L., Dore, C., Ruffio, V., Herve, Y., Morice, J., Dattee, Y., Dumas, C. and Gallais, A. 1992. Male sterilities and F_1 hybrids in *Brassica*. Reprod. Biol. Plant Breed. **38**: 107–119.

Ripley, V. L. and Arnison, P. G. 1990. Hybridization of *Sinapis alba* L. and *Brassica napus* L. via embryo rescue. Plant Breed. **104**: 26–33.

SAS Institute, Inc. 1985. SAS user's guide: statistics. SAS Institute, Inc., Cary, NC.

Sharpe, A. G., Parkin, I. A. P., Keith, D. J. and Lydiate, D. J. 1995. Frequent nonreciprocal translocations in the amphidiploid genome of oilseed rape (*Brassica napus*). Genome **38**: 1112–1121.

Stiewe, G., Witt, U., Hansen, S., Theis, R., Abel, W. O. and Robbelen, G. 1994. Natural and experimental evolution of CMS for rapeseed breeding. Pages 59–76 *in* U. Kuck and G. J. Wricke, eds. Advances in plant breeding: Genetic mechanisms for hybrid breeding. Plant Breed. **18**: (Suppl.).

Westdal, P. H. and Romanow, W. 1972. Observations on the biology of the flea beetle, *Phyllotreta cruciferae* (Coleoptera: Chrysomelidae). Manitoba Entomol. **6**: 35–45.