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Effectiveness of Defatted Mustard Meals Used to Control Fungus Gnats

2000-2002

J. P. McCaffrey and M. J. Morra
University of Idaho
Moscow, Idaho

Subcontract Report
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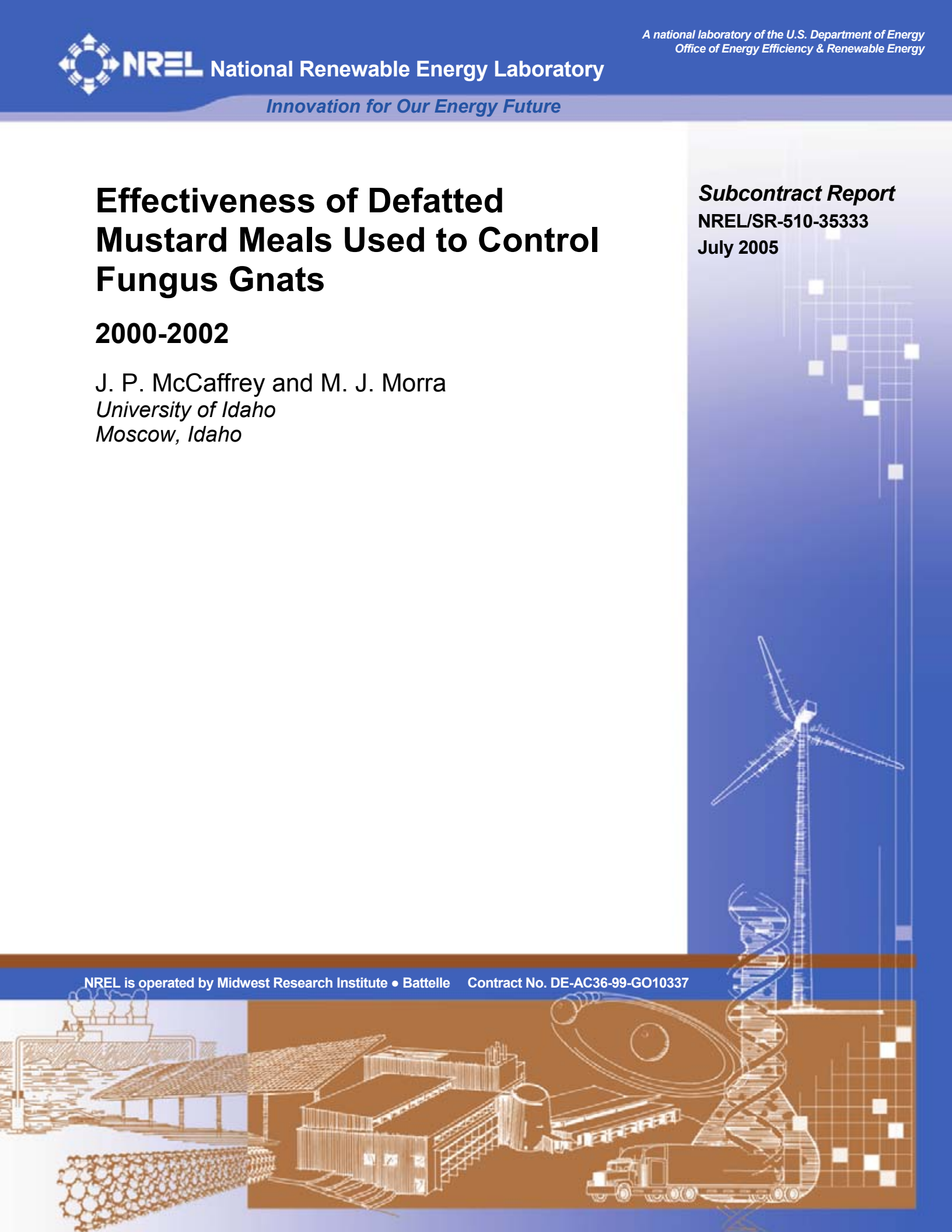

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J. P. McCaffrey and M. J. Morra
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NREL Technical Monitor: K. Shaine Tyson

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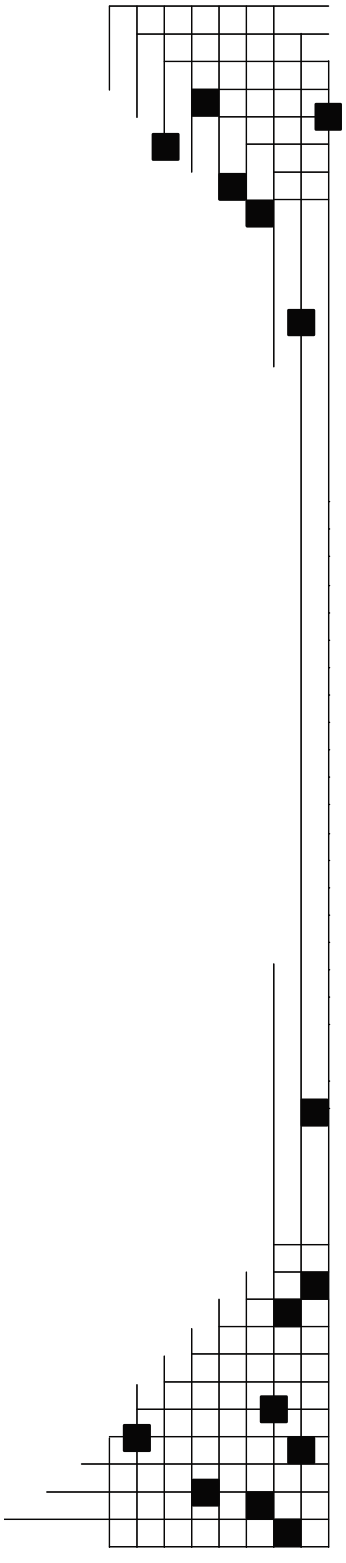

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I. CONTROL OF PLANT PESTS WITH MUSTARD MEALS

Glucosinolates, which are compounds that occur in agronomically important crops, may represent a viable source of allelochemic control for various soil-borne plant pests. Toxicity is not attributed to intact glucosinolates, but instead to biologically active products such as isothiocyanates (ITCs), organic cyanides, oxazolidinethiones, and ionic thiocyanate (SCN^-) released upon enzymatic degradation by myrosinase (thioglucoside glucohydrolase, EC 3.2.3.1) in the presence of water.

ITCs have historically been considered the "normal" products of glucosinolate breakdown. They are often volatile with pungent flavors or odors. The presence of propenyl ITC in mustards and horseradish is responsible for much of the flavor and thus, ITCs are sometimes called mustard oils. Formation requires that the initial unstable aglucon intermediate undergo a Loessen rearrangement to the R-N=C=S configuration. Isothiocyanates are quite reactive, although less so than the related isocyanates (R-N=C=O). A few commercially available soil fumigants depend on the activity of methyl ITC either as the parent compound or as produced from precursors such as sodium N-methyldithiocarbamate or tetrahydro-3,5-dimethyl-2*H*-1,3,5-thiadiazine-2-thione. Because of known toxicities, ITCs are often considered likely candidates for pesticidal activity.

Our objective is to develop a pesticidal product from mustard meals that can be used to control insect pests. We have focused our efforts on fungus gnats. This report details our current progress in developing a pesticidal product that can be used to control this plant pest.

Fungus Gnat Control with Mustard Meals

Fungus gnats and shore flies are persistent pests in flower and foliage production and are common in greenhouses, interiorscapes, and homes. The insects infest plants or algae within greenhouses, depending on the insect species. Fungus gnats commonly damage plant root systems because the larvae of these flies feed on roots, thus stunting plant growth. Root damage can occur in interiorscapes and in houseplants particularly if moist, organic-rich soils are present. Fungus gnat larval damage can be especially serious in greenhouses where the gnats feed on seedlings and cuttings. In addition to the larvae chewing on roots, both larvae and adults can spread plant pathogens and promote disease in commercial crops. The adult flies are a nuisance to consumers and they are not always evident when the plants are purchased since the larvae are in the soil (Price et al., 2001). The importance of the fungus gnat is made evident by a report in a 1993 Washington State survey of pests. It indicated that aphids, spider mites, fungus gnats, and thrips were reported as pests by 70%, 49%, 23%, and 21%, respectively, of greenhouse bedding-plant producers (Tanigoshi and Antonelli 1994, as reported by Copes (2001)).

Pesticides for controlling fungus gnats are available and usually applied as drenches to pots for larval control and foliar sprays for adult control. Insect growth regulators, organophosphate and carbamate insecticides, and Bt products are available. Furthermore, the botanical insecticide, azadirachtin and a relatively new insecticide, imidaclorprid, are available. While there are some registered insecticides, particularly for the greenhouse industry, little is available to the retailer or the consumer who may have to deal with a problem that originated with the plant wholesaler. Biological control agents are also available, and with water management and rescue treatments of the insecticides, gnats can often be controlled, but more often the applications are a disappointment because flies emerging from untreated larvae or from other areas can re-infest the plants. Because there are no economic threshold levels for managing these pests, the decision to suppress populations is largely subjective with marketing forces relating to state and federal regulation and plant quality playing an important role in the decision-making process. Finally, excessive reliance on one or just a few pesticides could easily result in the development of insecticide resistance because of the number of generations of flies that are often produced under the near optimal environments of greenhouses and homes. The availability of a product with a different mode of action will increase our ability to control this plant pest.

Materials and Methods

Four different meals were used in a series of bioassay experiments to determine pesticidal behavior against fungus gnats. The meals included two mustards (*B. juncea* Pacific Gold and *S. alba* IdaGold) and two *B. napus* varieties (Dwarf Essex and Athena). Chemical characteristics of the meals are shown in Table 1. Details concerning the methods of analysis are contained in a previous report entitled "Chemical Characterization and Release Efficiency of Defatted Mustard Meals."

We first determined the effectiveness of volatiles in closed containers in which the meal was physically separated from the test organism. Toxicity of the volatiles from the four meals to both fungus gnat adults and larvae were determined. Only volatiles from the wetted meal were allowed to contact the bioassay organism. Once we established which meal was most effective, we determined the most appropriate methods and rates of application of the meal in order to promote efficacy and increase consumer acceptance. Experiments included comparing meal incorporated in a potting medium, top dressing to the potting medium, and a novel "tea bag" preparation in which a sealed mesh bag was added to the surface of the potting medium. Specific details of the individual experiments are shown as footnotes in Tables 2 through 12.

Table 1. Glucosinolate content of cold pressed seed meals[1].

Glucosinolate trivial name	Glucosinolate structure	"Athena" B. napus	"Dwarf Essex" B. napus	"Pacific Gold" B. juncea	"IdaGold" S. alba
Desulfoglucoiberin	3-Me-SO-pentyl[2]	1.59±0.74			
Desulfoprogoitrin	2-OH(R)-3-butenyl	6.08± 0.29	60.98± 1.54		
Desulfoepi-progoitrin	2-OH(S)-3-butenyl		1.03± 0.23		6.38± 0.09
Desulfosinigrin	Propenyl[2]			109.87± 3.00	
Desulfoglucoraphanin	4-Me-SO-butyl[2]	trace	trace		0.80±0.05
Desulfonapoleiferin	2-OH-4-pentenyl	trace	3.55± 0.23		
Desulfoglucosinalbin	4-OH-benzyl[2]				549.57± 30.47
Desulfoglucoalyssin	4-Me-SO-pentyl[2]	trace	3.55± 3.50		
Desulfogluconapin	3-butenyl[2]	4.67± 0.34	41.40± 4.23		
Desulfo-4-hydroxyglucobrassicin	4-OH-indolyl-3-methyl	18.92± 4.47	5.48± 1.05	2.91± 0.25	
unknown					trace
Desulfoglucobrassicanapin	4-pentenyl[2]	1.17± 0.04	8.85± 0.91		
Desulfoglucotropaeolin	benzyl (4-Me-S-butyl)[2]	trace	0.54± 0.19		
Desulfoglucobrassicin	indolyl-3-methyl	3.61±0.93	trace		trace
Desulfogluconasturtin	2-phenylethyl[2]	trace	2.42±0.31		
Desulfo-4-methoxyglucobrassicin	4-MeO-indolyl-3-methyl	trace	0.43± 0.01	1.67 (± 0.10)	
unknown		trace		1.34 (± 0.08)	
Desulfoneoglucobrassicin	N-MeO-indolyl-3-methyl	trace	0.86±0.2		
	TOTAL	36.04± 4.06	129.09± 7.13	115.79 (± 3.12)	556.75± 29.69
	ITC-producing	7.43	56.76	109.87	550.75

[1]All values expressed in units of μmol g^{-1} of sample (parentheses values are the standard deviation of the sample set). All reported values are the average of 12 replications from three analysis runs. All meals analyzed were completely defatted and freeze-dried prior to extraction procedure.
[2]Isothiocyanate-producing glucosinolates; ITC- Isothiocyanate.

Results and Discussion

Tables 2 and 3 show data collected in preliminary experiments designed to determine the effects of meal volatiles on fungus gnat adults. *B. juncea* Pacific Gold showed complete control, whereas *S. alba* IdaGold was ineffective (Table 2). The high glucosinolate *B. napus* "Dwarf Essex" showed partial control and, as expected, low glucosinolate *B. napus* Athena had no effect on adult fungus gnat survival (Table 3). Preliminary results with larvae were similar (Tables 4 and 5), except that Dwarf Essex showed no effect.

Propenyl glucosinolate, which dominates *B. juncea* meal (Table 1), produces propenyl ITC when water is added to the meal. Our data indicate that propenyl ITC is an effective insecticide against both fungus gnat adults and larvae (Tables 2 and 4). In contrast, 4-hydroxybenzyl glucosinolate contained in *S. alba* meal produces 4-hydroxybenzyl ITC, an unstable compound that hydrolyzes to form SCN^- and 4-(hydroxymethyl)phenol. SCN^- is not volatile and therefore had no effect on fungus gnat adults or larvae in this bioassay (Tables 2 and 4). Although the exact amount of 4-(hydroxymethyl)phenol in the atmosphere of bioassay containers is not known, the compound also showed no toxicity. *B. napus* Dwarf Essex meal will produce mainly 3-butenyl ITC (Table 1) that, based on our bioassays, shows limited toxicity only (Tables 3 and 5). *B. juncea* meal should thus be used when only volatile products from the meal will contact fungus gnats. Additional experiments must be performed to determine if non-volatile glucosinolate hydrolysis products are biologically active.

Table 2. Biological activity of volatiles produced from *B. juncea* or *S. alba* meal on adult fungus gnats (n=1).[1]

	Number adults alive:		
Treatment[2]	after 90 minutes	after 17 hrs	after 24 hrs
Peat moss (control)	10	8	8
B. juncea (Pacific Gold)	0	0	0
S. alba (IdaGold)	10	8	5

[1]Bioassay chamber consisted of a 50-dram snap-cap plastic vial, with 10 adults placed in a 9-dram snap cap vial with organdy top and drop of apple sauce for sustenance. Treatment material was placed at the bottom of the 50-dram vial.
[2] Treatments: 1) 0.75 g peat moss + 4 ml water; 2) 0.75 g B. juncea meal + 4 ml water; and 3) 0.75 g S. alba meal + 4 ml water.

Table 3. Biological activity of volatiles produced from *B. napus* Athena or *B. napus* Dwarf Essex meal on adult fungus gnats (n=2).[1]

	Mean number adults alive after:					
Treatment[2]	30 min.	60 min.	90 min.	6 hrs	18 hrs	24 hrs
B napus (Athena)	10	10	10	10	9.5	7.5
B. napus (Dwarf Essex)	10	10	10	9.5	7.0	3.0

[1]Bioassay chamber consisted of a 50-dram snap-cap plastic vial, with 10 adults place in a 9-dram snap cap vial with organdy top and drop of apple sauce for sustenance. Treatment material was placed at the bottom of the 2-dram glass vial.
[2] Treatments: 1) 0.75 g B. napus (Athena) + 4 ml water; 2) 0.75 g B. napus (Dwarf Essex) meal + 4 ml water.

Table 4. Biological activity of volatiles produced from *B. juncea* or *S. alba* meal on larval fungus gnats (n=2).[1]

	Mean number larvae alive:		
Treatment[2]	after 90 minutes	after 20 hrs	after 43 hrs
Peat moss (control)	10	10	10
B. juncea (Pacific Gold)	10	0	0
S. alba (IdaGold)	10	10	10

[1]Bioassay chamber consisted of a 50-dram snap-cap plastic vial, with 10 last-instar larvae placed on small piece of agar sprinkled with small amount of sifted alfalfa meal in open 4-dram glass vial. Treatment material was placed in a separate open 4-dram glass vial.
[2] Treatments: 1) 0.75 g peat moss + 4 ml water; 2) 0.75 g B. juncea meal + 4 ml water; and 3) 0.75 g S. alba meal + 4 ml water.

Table 5. Biological activity of volatiles produced from *B. napus* Athena or *B. napus* Dwarf Essex meal on larval fungus gnats (n=2).[1]

	Mean number larvae alive after:		
Treatment[2]	90 min.	17 hrs	24 hrs
*B. napus (*Athena)	10	9.5	9.5
B. napus (Dwarf Essex)	10	9.5	9.5

[1]Bioassay chamber consisted of a 50-dram snap-cap plastic vial, with 10 last-instar larvae placed on a small piece of agar sprinkled with small amount of sifted alfalfa meal in open 4-dram glass vial. Treatment material was placed in a separate open 4-dram glass vial.
[2] Treatments: 1) 0.75 g B. napus (Athena) + 4 ml water; 2) 0.75 g B. napus (Dwarf Essex) meal + 4 ml water.

Results obtained in the preliminary experiments described above were confirmed in an experiment with larger numbers of replicates and a full complement of the meals (Table 6). We chose to focus on only the larval stage of the organism, since it is this stage that will be easiest to target using meal products. *B. juncea* meal was the only meal to produce volatiles that caused a toxic effect on fungus gnat larvae. Partial toxicity was observed at the first measurement time of 2 h with complete kill measured at 4 h. None of the other three meals showed any significant toxicity towards fungus gnat larvae. Either the

volatiles were not produced, or they were produced at levels that were below a threshold level of toxicity. This work confirms that *B. juncea* meal should be the focus of any future efforts to develop a soil amendment to control fungus gnats. The use of other meals such as *B. napus* or *S. alba* are unlikely to result in acceptable efficacy. It should be noted that other plant pests, including other insects, may behave differently and that testing against the specific pest should be performed before making further generalizations.

Toxicity expressed in volatile experiments may not necessarily correspond to efficacy when the meals are used as a soil amendment given the partitioning behavior of the active ingredient. Because the soil or potting medium will behave as a three-phase system in which the active ingredient (propenyl ITC) distributes itself, toxicity may be altered. Experiments were necessary to determine if partitioning of propenyl ITC within the soil environment decreases efficacy.

Table 6. Biological activity of volatiles produced from four meals on larval fungus gnats (n=10).[1]

	Mean number larvae per container alive (% alive) after:		
Treatment	2 hrs	4 hrs	24 hrs[2]
Brassica napus (Athena)	20.0 (100%)	20.0 (100%)	19.9 (99.5%)
Brassica napus (Dwarf Essex)	20.0 (100%)	20.0 (100%)	19.6 (98%)
Brassica juncea (Pacific Gold)	16.6 (83%)	0.0 (0%)	0.0 (0%)
Sinapis alba (IdaGold) (batch 1)	20.0 (100%)	20.0 (100%)	19.4 (97%)

[1]Bioassay chamber consisted of a 50-dram snap-cap plastic vial, with 20 last-instar larvae placed on small piece of agar sprinkled with small amount of sifted alfalfa meal in open 4-dram glass vial. Treatment material (1.0 g meal) was placed in a separate open 4-dram glass vial. Five milliliters of water were added to meal at start of experiment. Experiment set up on February 21, 2002.

[2]Dead larvae in B. napus and S. alba treatments appear to have drowned, except possibly one larva in Dwarf Essex treatment.

Meal incorporation into the potting medium showed similar trends with respect to fungus gnat toxicity as did volatile bioassays (Tables 7 and 8). *B. juncea* meal showed complete fungus gnat control at a rate of 3% (Table 7), whereas *S. alba* showed little impact at a rate of 6% (w:w) (Table 8). This lack of any effect of *S. alba* meal along with a similar survival rate for *B. napus* treatments indicates that it is the specific type of glucosinolate that is significant. As described above, *B. juncea* meal contains primarily propenyl glucosinolate that hydrolyzes to form the corresponding ITC upon wetting the meal. Efforts to control insects should thus focus on *B. juncea* meal as compared to *S. alba* containing 4-OH benzyl glucosinolate, despite the fact that *S. alba* meal contains much higher glucosinolate concentrations (Table 1).

This conclusion was confirmed by observations that *B. juncea* meal was also toxic to nematodes, but *S. alba* and *B. napus* meals were not (Tables 7 and 8). Although nematodes were not target organisms for this research, this observation opens the possibility of using *B. juncea* meal as a soil amendment to control plant-feeding nematodes. These data also illustrate that propenyl ITC is a general biocide that will have activity with a wide spectrum of organisms, both pests as well as non-pests. The potential of phytotoxicty to the crop of interest is a potential concern that has not yet been addressed.

Table 7. Toxicity of *B. juncea* meal to fungus gnat larvae as determined by the numbers of emerged adults (n=3).[1]

Treatment	Mean number fungus gnat adults emerged per pot	Nematodes present (day 13)
B. napus 3% (control)	15.7	yes
B. juncea 1%	11.3	yes
B. juncea 3%	0.0	no
B. juncea 6%	0.0	no

[1]Treatments consisted of approximately 18 g dry weight of a Sunshine mix no. 2 / composted bark mixture (7:3); mixed with 1%, 2%, or 3% meal (B. napus Athena or B. juncea Pacific Gold); plus approximately 1.6 g dry pinto beans (soaked for 24 hrs in water) for larval food; plus the appropriate amount of water to have a moist mixture. This mixture was placed in plant pots (6 cm x 6 cm x 8 cm ht). Twenty fungus gnat larvae were added to the mixture in each of the pots. Numbers of adults emerging were recorded daily.

Table 8. Toxicity of *S. alba* meal to fungus gnat larvae and nematodes as determined by the numbers of emerged adults (n=3).[1]

Treatment		Mean number fungus gnat adults emerged per pot	Nematodes present (day 14)
B. napus	3% (control)	15.0	yes
S. alba	*1%*	15.0	yes
S. alba	3%	14.7	yes
S. alba	6%	12.7	yes

[1]Treatments consisted of approximately 18 g dry weight of a Sunshine mix no. 2 / composted bark mixture (7:3); mixed with 1%, 2%, or 3% meal (B. napus Athena or S. alba IdaGold); plus approximately 1.6 grams dry pinto beans (soaked for 24 hrs in water) for larval food; plus the appropriate amount of water to have a moist mixture. This mixture was placed in plant pots (6 cm x 6 cm x 8 cm ht). Twenty fungus gnat larvae were added to the mixture in each of the pots. Numbers of adults emerging were recorded daily.

Table 9 shows fungus gnat survival after large amounts of meal were mixed into the potting mix. A second sample of *S. alba* meal was used to confirm that previous results were not caused by some peculiar aspect related to seed processing or meal storage (batch 2). Survival was determined by counting the number of adults that emerged from the respective treatments. High glucosinolate *B. napus* and *S. alba* meals showed some control at rates of 10%, but never toxicity equivalent to that of *B. juncea*. We also determined nematode survival in the potting mix and noted that only *B. juncea* completely eliminated nematodes. These data confirm the need to concentrate efforts on using *B. juncea* meal for fungus gnats, and possibly nematode control. 4-OH Benzyl ITC produced from *S. alba* meal is not an effective insecticide against fungus gnats even at very high meal amendment rates.

Table 9. Toxicity of large amounts of *B. napus*, *S. alba,* and *B. juncea* meals to fungus gnat larvae as determined by the numbers of emerged adults (n=5).[1]

Treatment	Mean number fungus gnat adults emerged per pot[2]	Survival (%) (larvae to adult)	Nematodes present (day 14)
B. napus (Athena) 20%	13.6 ± 0.9 a	68	yes
B. napus (D. Essex) 20%	10.8 ± 2.0 ab	54	yes
B. napus (D. Essex) 10%	8.2 ± 1.2 bc	41	yes
B. napus (D. Essex) 30%	7.4 ± 0.9 cd	37	yes
S. alba 10% (batch 2)	5.0 ± 1.4 d	25	yes
S. alba 20% (batch 2)	2.0 ± 0.4 e	10	yes
S. alba 30% (batch 2)	1.8 ± 0.0 e	9	yes
B. juncea 20%	0.0 ± 0.0 e	0	no
B. juncea 10%	0.0 ± 0.0 e	0	no
B. juncea 30%	0.0 ± 0.0 e	0	no

[1]Treatments consisted of approximately 18 g dry weight of a Sunshine mix no. 2 / composted bark mixture (7:3); mixed with 10%, 20%, or 30% meal; plus approximately 1.6 grams dry pinto beans (soaked for 24 hrs in water) for larval food; plus the appropriate amount of water to have a moist mixture. This mixture was placed in plant pots (6 cm x 6 cm x 8 cm ht). Twenty fungus gnat larvae were added to the mixture in each of the pots. Pots were placed in 1-quart canning jars with organdy top. Numbers of adults emerging were recorded daily. Soil mix was oven-dried overnight before use. Experiment set up on February 28, 2002.

[2]Means in a column followed by the same letter are not significantly different (P = 0.05) as determined using a protected LSD.

Table 10 shows final data on fungus gnat larval survival for experiments with larger numbers of replicates. The most effective treatments were *B. juncea* meal amendments at 3% and 6%. A range in meal amendment of near 3% has thus been established for the specific potting mix used in the current experiments. Rates for other potting mixes or soils may require additional testing. However, because this particular mix is highly organic, we consider the rates determined here to be near the extreme and probably worst case. It is likely that lower meal amendment rates will be required in lower organic carbon-containing mixes or in soils. Decreased fungal gnat survival with amendment of *B. napus* Dwarf Essex and *S. alba* meals was determined, but even 6% amendment did not result in acceptable fungus gnat control.

Table 10. Toxicity of *B. napus*, *S. alba,* and *B. juncea* meals to fungus gnat larvae as determined by the numbers of emerged adults (n=10).[1]

Treatment	Mean number fungus gnat adults emerged per pot[2]	Survival (%) (larvae to adult)
S. alba 1% (batch 2)	15.6 ± 0.7 a	78.0
S. alba 3% (batch 2)	15.3 ± 0.6 ab	78.0
B. juncea 1%	14.8 ± 1.1 ab	73.5
B. napus (D. Essex) 3%	14.3 ± 0.9 ab	71.5
B. napus 6% (Athena)	14.0 ± 1.1 ab	70.0
S. alba 6% (batch 2)	12.7 ± 0.9 b	63.5
B. napus (D. Essex) 1%	12.5 ± 1.6 b	62.5
B. napus (D. Essex) 6%	7.9 ± 1.5 c	39.5
B. juncea 3%	0.7 ± 0.6 d	3.5
B. juncea 6%	0.0 ± 0.0 d	0.0

[1]Treatments consisted of approximately 18 g dry weight of a Sunshine mix no. 2 / composted bark mixture (7:3); mixed with 1%, 2%, or 6% meal, plus 4 pinto bean halves (soaked for 24 hrs in water) for larval food; plus the appropriate amount of water to have a moist mixture. This mixture was placed in plant pots (6 cm x 6 cm x 8 cm ht). Twenty fungus gnat larvae were added to the mixture in each of the pots. Pots were placed in 1-quart canning jars with sealed tops for 24 hrs, at which time organdy cloth replaced the lid. Numbers of adults emerging were recorded daily. Soil mix was oven-dried overnight before use. First five reps were set up on March 5 and second five reps were set up on March 13, 2002.

[2]Means in a column followed by the same letter are not significantly different (P = 0.05) as determined using a protected LSD.

Current trials rely on complete incorporation of the meal into the potting medium. Increased utility and user acceptance will be achieved by marketing a product that is less labor intensive with respect to application. We therefore chose to explore alternate application methods to determine if fungus gnat control could be achieved using a less labor intensive method.

Table 11 shows the effect of meal top-dressing and minimal soil incorporation on the survival of fungus gnat larvae. Significant decreases occurred in emerging numbers of adult fungus gnats when *B. juncea* meal was either top-dressed or incorporated into the top 6-7 mm of potting mix. There was no difference between any of the 3% and 6% treatments. Top dressing may be a viable option for controlling fungus gnats, thus increasing the ability of the grower to control this pest on established plants prior to shipment. Homeowners likewise, could eliminate fungus gnats in pots containing established plants.

Table 11. Toxicity of top-dressed and surface-incorporated *B. juncea* meal to fungus gnat larvae as determined by monitoring the number of emerged adults (n=4).[1]

Treatment	Mean number fungus gnat adults emerged per pot[2]	Survival (%) (larvae to adult)
No meal, no disturbance	13.3 ± 0.9 a	66.3
No meal, disturbance	13.0 ± 1.6 a	65.0
B. juncea 1%, top-dressing	9.3 ± 1.4 ab	46.3
B. juncea 1%, incorporated 6-7 mm	7.8 ± 2.4 bc	38.8
B. juncea 3%, top-dressing	3.8 ± 1.9 cd	18.8
B. juncea 3%, incorporated 6-7 mm	5.8 ± 1.8 bcd	28.8
B. juncea 6%, top-dressing	2.3 ± 1.3 d	11.3
B. juncea 6%, incorporated 6-7 mm	2.8 ± 1.3 d	13.8

[1]Pinto bean seeds were planted into soil mixture (19 or 20 grams dry weight) in plant pots (6 cm x 6 cm x 8 cm ht) on March 9 (block 1) and March 21, 2002 (block2). Soil mixture consisted of Sunshine mix no. 2 / composted bark mixture (7:3). Twenty fungus gnat larvae were added March 25 (block 1) and April 2 (block 2) to the soil mixture (~1-2 cm deep) in each of the pots. Pots were placed in 1-quart canning jars with organdy tops. Numbers of adults emerging were recorded daily. Soil mix was oven-dried overnight before use. Twenty-five milliliters of water were added to soil surface of each pot (block 1) on March 28, March 31, April 3, and April 7. Twenty milliliters of water were added to soil surface of each pot (block 2) on April 5, April 8, April 11, and April 14.

[2]Means in a column followed by the same letter are not significantly different (P = 0.05) as determined using a protected LSD.

Additional bioassays were conducted to optimize the use and application of the meal and to determine the viability of an innovative method of delivery. In this experiment, pots were bottom-watered for comparison with the previous experiment. In addition, we included a "tea bag" treatment in which the meal was sealed inside of a mesh bag that measured 6 cm x 6 cm. The bag was placed on the soil surface and wetted by the water carried upward through the soil. A liquid extract from the meal was also included as a treatment. In general, top dressing of *B. juncea* provided the best control under the conditions of bottom watering (Table 12), apparently better than when top watering was practiced (Table 11). This was also true of a tea bag formulation of *B. juncea* in which control was equal to that of top dressing. The tea bag formulation offers a benefit of ease of use and should be appealing to the homeowner market. Unfortunately, the extract solution of the meal had no effect on fungus gnat survival. A more thorough extraction of pesticidal agents from the meal may be necessary to obtain activity in extracts. The potential use of adjuvants such as surfactants is being considered. The development of a soil drench would have broader appeal than a solid meal amendment and further work in this area is worthwhile.

Table 12. Effect of application method and rate of *B. juncea* mustard meals on survival of fungus gnats using bottom watering (n=10).

Application method	Rate[1]	% Survival[2]
Incorporation	1%	42.5b
"Tea bag"	1%	11.0c
No meal --	1%	71.0a
Solution	<1%	61.0a
Top dressing	1%	8.5c

[1]As a percentage of the dry weight of the soil
[2]Larva to adult

Summary of Significant Results and Conclusions

1. *S. alba* IdaGold has the highest glucosinolate concentration with ITC-producing glucosinolates present in amounts of 551 µmol/g meal. This is an extremely high glucosinolate concentration and *S. alba* meal is therefore a potentially valuable pesticidal source of ITC. However, toxicity against specific plant pests must be determined. Current bioassays indicate that this meal is ineffective against fungus gnats and nematodes. In contrast, *B. juncea* Pacific Gold meal contains ITC-producing glucosinolate concentrations of only 110 µmol/g meals. Although lower in concentration, glucosinolates in Pacific Gold meal (propenyl) produce much more biologically active ITCs than those in *S. alba* meal (4-OH benzyl). Although this is true for our current bioassay organisms, this is not true for other organisms such as weeds. We recommend that breeding efforts be directed at

producing meals containing propenyl glucosinolate concentrations closer to 250 μmol/g. Breeding efforts to produce higher concentrations of 4-OH benzyl glucosinolate in the meal are of lower priority since the meal already appears to have quite high glucosinolate concentrations.

2. *B. juncea* Pacific Gold meal suppresses fungus gnats when homogeneously mixed in potting soil at a rate of 3%. This is a realistic rate of soil amendment that will be commercially acceptable. Tests for phytoxicity are necessary to determine any negative effects on the plants in which fungus gnats are a problem. The development of meals with higher propenyl glucosinolate concentrations will decrease the amount of meal necessary to achieve fungus gnat control. The development of a commercially viable product based on meals containing propenyl glucosinolate is feasible.

3. Top dressing reduced fungus gnat populations when top watered, but not to the same extent as homogeneous incorporation of the meal into the potting mix. Breeding for higher seed propenyl glucosinolate concentrations will increase the effectiveness of top dressing. Horticultural markets will probably be much more receptive to a top dressing treatment than to complete incorporation because of the ease of application. It is thus important to produce new mustard varieties with propenyl glucosinolate concentrations closer to 250 μmol/g meal.

4. Bottom watering increased the efficacy of fungus gnat control. The practical application of this method may be limited since bottom watering is not commonly practiced. However this work demonstrates that increased fungus gnat control is possible if the reason for increased efficacy could be determined and applied.

5. Packaging techniques such as using a mesh bag may have broad consumer appeal for the homeowner market given the fact that *B. juncea* meal was quite effective when applied to soil in this manner. Optimizing the use of this type of amendment product should be pursued.

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Our objective is to develop a pesticidal product from mustard meals that can be used to control insect pests. We have focused our efforts on fungus gnats. This report details our current progress in developing a pesticidal product that can be used to control this plant pest.

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