Control of Dandelions with Mustard Meal or Extract

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Summary

A growth chamber study, conducted by the PTRC, identified oriental mustard as having an ability to control dandelion seed germination and also the ability to kill dandelion seedlings for at least 4 weeks after germination. This past spring, a field experiment was initiated to determine if mustard could be used to control dandelion seed germination and dandelion seedlings in a field situation. Unfortunately, there was no control. Therefore, a series of non-replicated screening studies were initiated to determine why the mustard was effective in the growth chamber, but not in the field.

These screening studies showed that a greater effect was achieved if the mustard product was watered in. However, it was determined that much of the active in oriental mustard was tied up in the thatch layer of turf. It was also determined that the active was very volatile and that recovery of the active peaked at two hours following application and then declined after that. These results illustrated the need for re-formulation of the product in order to reduce volatility and improve its effectiveness.

Rationale for Using Mustard Meal to Control Dandelions

Mustard meal and other members of the Brassicaceae contain glucosinolates. Both the volatiles (Brown and Morra, 1995) and the water soluble products of glucosinolate hydrolysis can inhibit seed germination (Brown and Morra, 1996; Mason-Sedun et al., 1986) inhibit seedling growth (Brown and Morra, 1997) and can kill a number of fungi (Sarwar et al., 1998) and insect species (Borek et al., 1998). However, mammalian systems metabolize and eliminate these products rapidly so they do not pose a problem to either humans or other mammals (Brown and Morra, 1997). Consequently, mustard meal can be used as a feed supplement.

The breakdown products of glucosinolates have been widely reported to inhibit seed germination or seedling emergence (Bialy et al., 1990; Boydston and Hang, 1995; Dale, 1986; Haramoto and Gallandt, 2005; Ju et al., 1983; Oleszek, 1987; Stiehl and Bible, 1989; Teasdale and Taylorson, 1986; Vaughn and Boydston, 1997; Vaughn et al., 2006). Most of these studies were conducted on agricultural crops and studied the effect either on weed seed or crop seed germination and emergence.

Glucosinolates and Their Breakdown Products

There are many different types of glucosinolates. However, two broad classes of glucosinolates are the aromatic glucosinolates which contain a benzene ring in their chemical structure (referred to as benzyl) and the aliphatic glucosinolates which do not contain a benzene ring (referred to as allyl) (Norsworthy and Meeham, 2005). Within these two broad classes, there are many different glucosinolates (Brown and Morra, 1997).

Different species may contain different types of glucosinolates. For example, *Sinapis alba* or yellow mustard contains hydroxybenzyl glucosinolate which is one of the benzyl

glucosinolates (Borek and Morra, 2005). In contrast, *Brassica juncea* or oriental mustard contains sinigrin which belongs to the allyl class of glucosinolates (Vaughn et al., 2006).

Glucosinolates are produced by species that belong to the *Brassicacea* or mustard family. However, there can be a lot of variation in the type and concentration of glucosinolate between species, cultivar or even plant part (Eberlein et al., 1998).

It is actually the breakdown products of the glucosinolates, the isothiocyanates (or ITC's) that have allelopathic qualities. When the cell vacuoles are ruptured the glucosinlates are hydrolyzed by the enzyme myrosinase to form a variety of potential allelochemicals (Borek et al., 1996)(Vaughn et al., 2006). Different glucosinolates produce different breakdown products which can have different activity against specific target organisms. Freezing and thawing helps to increase the concentration of ITC's (Morra and Kirkegaard, 2002). Oriental mustard, for example is an aliphatic (or allyl) glucosinolate so the breakdown product that is produced is Allyl Isothicyanate (AITC).

Different breakdown products have different residence times in the soil. It is possible that differences in inhibition of seed germination may be related to residence times of the breakdown products as well as to amount and type of glucosinolates produced (Eberlein et al., 1998). Isothiocyanates remain in the soil for as little as a few days to a few weeks and volatile losses are a major route of disappearance (Brown and Morra, 1997). Soil residence times are reduced as soil temperature increases and as soil moisture decreases. Losses are also more rapid in soils containing greater concentrations of organic carbon (Borek et al., 1995).

Use of Oriental Mustard to Control Dandelions in the Field

A growth chamber study, conducted by the PTRC, identified oriental mustard as having an ability to control dandelion seed germination and also the ability to kill dandelion seedlings for at least 4 weeks after germination.

This past spring, a field experiment was initiated to determine if mustard could be used to control dandelion seed germination and dandelion seedlings in a field situation. Unfortunately, there was no control. Therefore, a series of non-replicated screening studies were initiated to determine why the mustard was effective in the growth chamber, but not in the field. If it is possible to determine why the mustard is effective in one situation and not the other it still may be possible to apply the mustard in a way that would improve efficacy.

Some possible explanations for the difference between the growth chamber experiments and the field experiments include:

- meal getting caught up in the thatch layer and being volatilized too quickly
- lack of moisture to increase movement of the chemicals (i.e. AITCs) into the soil

A series of screening experiments were conducted to explore these two options.

Analysis of Allyl Isothiocyanate (AITC) in the Thatch or in Soil Collected From Field Plots

Oriental mustard meal or a combination of 80% oriental mustard and 20% yellow mustard meal was applied to the turf plots in the field. Samples were collected after 1 hour or 1 week using Headspace Solid Phase Microextraction (SPME). A qualitative analysis of the AITC present in the thatch layer or different levels in the soil was performed using a gas chromatograph. This methodology was used in each of the following experiments and will be referred to as SPME-GC.

This first study was conducted to determine if applying water (or water + surfactant) after application of the meal could improve the ability to extract AITC from either the bottom cm of the thatch layer or from different layers in the soil. The ability to extract AITC in greater amounts or for longer periods of time would indicate reduced losses to volatilization.

The application of water improved soil penetration and presumably reduced volatilization losses as it was possible to extract more AITC from the thatch and the soil than in the control treatment which did not receive water (Table 1). However after even 1 week, the amount of AITC that was able to be extracted from any of the treatments was very low, especially in the soil (Table 2). Mixing in some yellow mustard (containing more of the benzyl form of glucosinolates) did not improve the situation (Table 3).

Table 1. Qualitative Analysis of AITC in the thatch or soil by SPME-GC. Samples were collected on Sept. 18, 2008, 1 hr after application of oriental mustard meal.

Treatment	AITC (mg/g soil)			
	Thatch	0-1 cm	1-2 cm	2-3 cm
Control	4.4	0.0	0.0	0.0
Water	16.5	8.9	2.0	0.0
Water + Surfactant	3.4	0.0	0.0	0.0

Table 2. Qualitative Analysis of AITC in the thatch or soil by SPME-GC. Samples were collected on Sept. 25, 2008, 1 week after application of oriental mustard meal.

Treatment	AITC (mg/g soil)			
	Thatch	0-1 cm	1-2 cm	2-3 cm
Control	0.9	0.0	0.0	0.0
Water	1.5	0.2	0.0	0.0
Water + Surfactant	0.7	0.2	0.0	0.0

Table 3. Qualitative Analysis of AITC in the thatch or soil by SPME-GC. Samples were collected on Sept. 25, 2008, 1 hr after application of oriental mustard mixed with 20% yellow mustard meal.

Treatment	AITC (mg/g soil)			
	Thatch	0-1 cm	1-2 cm	2-3 cm
Control	4.8	0.0	0.0	0.0
Water	3.9	3.0	0.7	0.2
Water + Surfactant	8.5	0.7	0.3	0.0

Analysis of AITC in the Thatch or Soil – Growth Chamber Experiments

Pots were established in the growth chamber containing turf that was transplanted from the field. Oriental mustard meal or extract, was applied in a variety of rates and forms including:

- oriental mustard meal at 240 g/m^2
- oriental mustard meal at 480 g/m^2
- oriental mustard extract at 480 g/m^2
- oriental mustard extract at 480 g/m^2 + defatted cake at 240 g/m^2
- AITC powder at 480 g/m^2

In each case, the rate refers to the amount of product. For oriental mustard, the estimated AITC content is 17.8 mg/g. Therefore, the treatment applying AITC powder would be applying a much higher rate of the AITC than the other treatments.

Once again, most of the AITC that was extracted was still in the thatch layer (Table 4). However, the combination of oriental mustard extract mixed with defatted cake produced increased levels of the AITC in the thatch and especially in the soil. Even after 24 hours there were higher levels in the soil. Therefore, the use of the extract with the defatted cake did keep the AITC in the soil for a longer period of time.

Table 4. Qualitative analysis of AITC in the thatch or soil by SPME-GC from samples collected 1h or 24 hours after application of oriental mustard (OM) or AITC treatments.

Treatment	AITC (nmoles/g – dry wt. basis)			
	Thatch		Soil – Top 1 cm	
	1 Hour	24 Hours	1 Hour	24 Hours
OM meal - 240 g/m^2	18,344	33	381	6
OM meal - 480 g/m^2	25,740	459	472	11
OM extract – 480 g/m^2	22,495	711	8,787	128
OM extr. – 480 g/m^2 + defatted cake – 240 g/m^2	63,514	25,663	30,543	12,528
AITC 4 – 480 g/m^2	33,150	458	9,815	18

In the next study, oriental mustard cake was applied at the lowest rate (240 g/m^2) and then watered. The amount of AITC that could be extracted from the thatch or the soil was monitored over a variety of times. The amount of AITC peaked at 2 hours after application. However, even after 12 hours, the level of AITC in the soil was very low (Table 5).

Table 5. Qualitative analysis of AITC in the thatch or soil by SPME-GC from samples collected at various times after application of oriental mustard meal at a rate of 240g/m^2.

Treatment	AITC content (nmoles/g - dry wt. basis)					
	1 Hour	2 Hours	3 Hours	6 Hours	12 Hours	24 Hours
Thatch	287	343	186	91	19	10
Soil	18	39	29	16	2	1

This next experiment tracked AITC content in the thatch or the soil over a period of time when the oriental mustard was applied in a variety of ways:

- No water, 240 g/m^2

- Extract + defatted cake, 240 g/m^2 of each
- Extract, 240 g/m^2
- AITC powder, 240 g/m^2

These results would seem to indicate that the combination of extract mixed with the defatted cake or the AITC powder were most effective (Table 6). Not adding water to the meal, following application resulted in very low levels of AITC in the thatch or the soil. Also, the extract, by itself was not as effective as the other treatments.

Table 6. Qualitative analysis of AITC in the thatch or soil by SPME-GC from samples collected at various times after application of oriental mustard applied in different forms.

Treatment	AITC (nmoles/g - dry wt)					
	1 hr	2 hr	3 hr	6 hr	12 hr	24 hr
Thatch						
No water	590	290	471	238	159	98
Extr. + Defat. cake	1,145	1,247	288	239	26	50
Extract	105	125	102	94	75	44
Powder	1,026	189	529	540	146	16
Soil						
No water	4	7	4	7	5	1
Extr. + Defat. cake	59	102	57	75	4	5
Extract	27	50	49	29	3	3
Powder	137	53	82	54	16	6

Conclusions

From these series of experiments, we can conclude that the application of water is essential in moving the AITC into the soil. It is felt that in order to be effective against dandelion seeds and seedlings, the AITC must move into the soil. Therefore, one of the differences between the original growth chamber experiment and these experiments was that much of the AITC was tied up in the thatch.

The form in which the product was applied produced some very large differences in the amount of AITC that could be extracted. For example, the mixture of oriental mustard meal extract and defatted cake produced greater levels of AITC in both the thatch and the soil with a greater residual time. Therefore, the same work with formulation should be able to greatly improve efficacy in killing dandelion seeds and seedlings.

There is a concern with the methodology. It takes an hour to extract the AITC from each of the samples, so much of the AITC may be being lost during the sampling procedure. Consequently, these results are probably consistently underestimating both AITC levels and residual time. Therefore, in future studies we will be changing the sampling procedure.

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