Project Title	Liverwort control using novel techniques
Project number:	HNS 175
Project leader:	Jill England, ADAS Boxworth
Report:	Year 3 report, March 2012
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AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

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GROWER SUMMARY

Headline

- Mulch application of *Brassica napus* and *Sinapis alba* ‘Braco’ seed meal can significantly reduce liverwort establishment
- Seed meal in combination with bark mulch can improve liverwort reduction.

Background and expected deliverables

Liverwort growing on the surface of growing media is a major problem to the horticulture industry, affecting both protected and outdoor grown hardy nursery stock. The cost of moss, liverwort and weed control at despatch alone is estimated at up to 3p per 3 L pot, equating to £5,625 per hectare based on 2012 figures, depending on the weed control regime in place (hand weeding, herbicide programme etc.) (Hewson, A. 2012). Zero tolerance of liverwort in certification schemes and a lack of approved chemical products make its control a technical priority for growers.

There are currently no herbicides approved for use over plants under protection that will control liverwort. Additionally, ensuring even distribution of pot toppers, such as bark remains a challenge for the industry, particularly for liner crops (i.e. those grown in small pot sizes).

The aim of this project has been to build on work completed in HDC projects HNS 126 and HNS 93c by investigating further the herbicidal effect of glucosinolate (GSL) hydrolysis products found in oil seeds on liverwort, and the suppression of liverwort growth by unknown biological or physical factors within certain growing media components.

GSLs and their hydrolysis products (isothiocyanates, ITCs) are responsible for the distinctive pungent smell and hot taste of cabbages, mustards and other brassicas and are known to have toxic effects against plants, root knot nematodes and fungal species; brassicas are successfully used in biofumigation of soils against weeds and diseases. GSLs could potentially be used to control weeds in containers; each brassica variety has a distinctive profile of one or more glucosinolates, each of which could have a different effect on liverwort.

In years 1 and 2 of this project, trials investigated the effect of different brassica oil seeds (*Brassica carinata, Sinapis alba* ‘Albatross’, *Sinapis alba* ‘Braco’*, Camelina sativa*, *Brassica* *napus* ‘RMF’ and *Brassica napus* ‘00’), and growing media amendments (Melcourt Sylvafibre®, Melcourt Growbark®, Perlite, Vital Earth Green Compost and sterilised loam) on liverwort establishment. *Sinapis alba* ‘Albatross’, *Sinapis alba* ‘Braco’, *Brassica napus* ‘00’ and *Camelina sativa*, and growing media amendment with Sylvafibre® and sterilised loam significantly reduced liverwort establishment.

The expected deliverables from this work include the development of an effective novel control for liverwort infestation based on:

- Growing media amendment with seed meal or a combination of seed meals to reduce liverwort establishment (either through herbicidal effect and/or any natural barrier effect).
- Growing media amendment with materials to provide natural microbial suppression of liverwort in addition to any physical effect.

Summary of the project and main conclusions

Two trials were carried out during 2011/12: 1) investigating the suppressive effect of mulch application of two seed meal varieties *Sinapis alba* ‘Braco’ (mustard) and *Brassica napus* (oilseed rape) to control liverwort at a single dose rate, and 2) investigating the effect of applying seed meal over the crop and removing the deposits using different methods.

Half of the seed meal was kept intact for the trial, and the rest was processed to a fine meal and the oil extracted. The trials were set up on 8 June 2011. For both trials, each pot was planted with a plug of *Clematis* ‘Ernest Markham’, which were grown according to commercial practice. Liverwort inoculum was provided by one ‘spreader’ pot per plot containing liverwort.

The results of several trial plots in both trials were excluded from statistical analysis following the accidental application of gluten by nursery staff on 22 September 2011, including the majority of the control treatment with bark, therefore these results are not reported for liverwort cover. The gluten also appeared to attract rats, which caused damage to a number of other pots.

Seed meal suppressive effect

This trial focused on two seed meal varieties, *Sinapis alba* and *Brassica napus*, applied at a single dose rate (6 g per pot), with six treatments, including two control treatments, seed meal only, a managed treatment, bark with seed meal, and a treatment using seed meal where the oil had not been extracted (whole seeds were ground just prior to setting up the trial). For the managed treatments a second application of seed meal was applied at the first sight of liverwort infestation on 1 August 2011, 9 weeks after treatment.

The most promising treatments from these trials were the seed meal with bark, and the managed treatment, where a second application of seed meal was applied (Figure 1). Of the two seed meals varieties tested, the *Brassica napus* provide greatest liverwort control; when mixed with bark less than 5% liverwort established in this treatment throughout the 33 weeks of the trial. Application of seed meal with bark would have the disadvantage of higher costs (including bark and its application). Liverwort cover was also low in the managed treatments, where a second application of seed meal was applied.

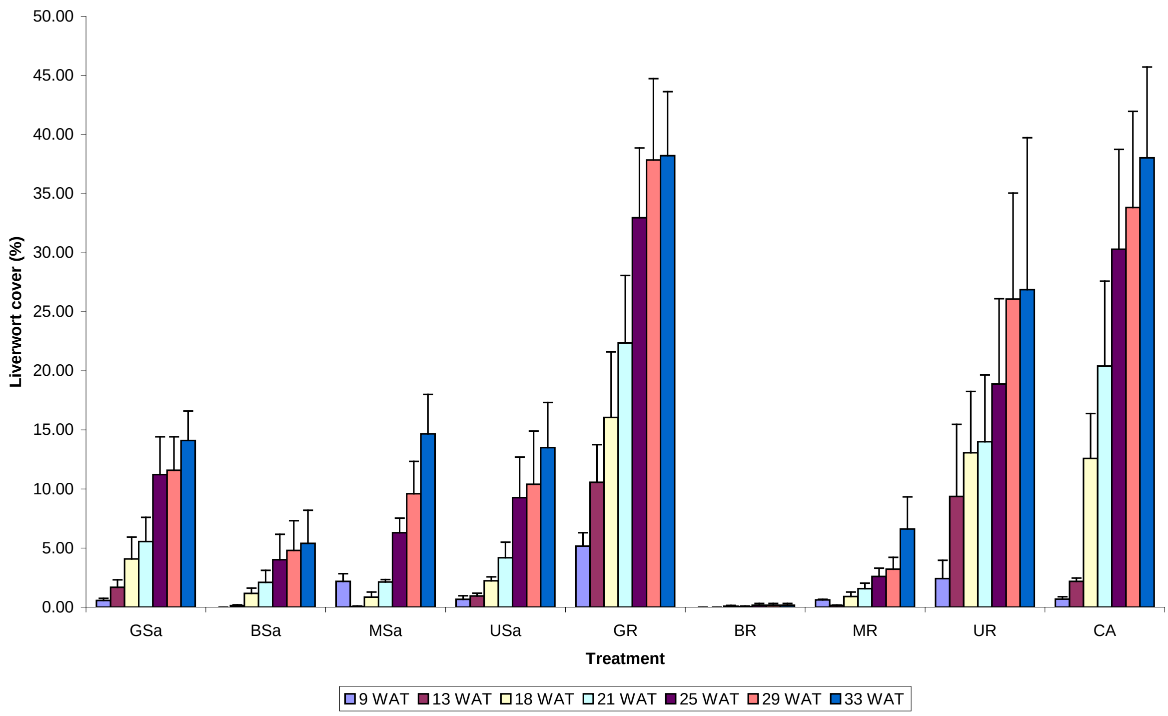

Figure 1. Percentage liverwort cover: GSa = Ground *Sinapis alba* 'Braco' seed meal*, BSa* = Ground *Sinapis alba* 'Braco' seed meal+bark, MSa = *Sinapis alba* 'Braco', managed treatment, USa = *Sinapis alba* 'Braco', unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus*, managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied. WAT = weeks after treatment

Phytotoxicity was recorded in all treatments, although only treatments using *Sinapis alba* 'Braco' four weeks after treatment were not commercially acceptable. After 13 weeks all treatments scored above 4 and were commercially acceptable.

There was a clear overall difference in plant height due to seed meal variety, with greater height recorded in the *Brassica napus* treatments. Plant height was greatest in the bark treatments, and treatments where the oil had not been extracted; the greatest adverse effect on plant height was due to the managed *Sinapis alba* ‘Braco’ treatments. No adverse effect on root development was recorded due to any of these treatments.

The seed meal with bark treatment showed greatest promise in terms of least liverwort cover with high plant quality and height, and low phytotoxicity. Bark mulch would be more expensive to apply, and it is recognized that it is difficult to achieve an even mulch layer in a commercial setting. Nevertheless, this treatment would make hand weeding generally more acceptable to nursery workers through the reduction of liverwort establishment.

The managed treatments (seed meal only) also showed good liverwort control, and could more easily be applied alone through a granular applicator, but the risk of phytotoxicity would be increased.

Effects of seed meal deposit removal method

This small scale, unreplicated, trial investigated seed meal application; the results were not statistically analysed. Seed meal was applied over the plant leaves and then any deposits were removed by shaking or washing them off, or the deposits were left in place. Two control treatments were ground seed meal applied as a mulch and no seed meal, the results for which were collected from control treatments within the seed meal suppressive effect trial. For the treatment where the deposits were allowed to remain on the plants, the growing media was watered and the foliage allowed to dry before treatment, so that seed meal did not stick to wet foliage.

Least liverwort established in the treatments where seed meal deposits were left on the leaves, but greatest phytotoxicity and smaller plants were also recorded in these treatments. Seed meal was easily removed from plants, however the recommendation would be to apply seed meal to dry foliage and tap off to avoid any fungal or phytotoxicity problems due to any seed meal sticking to the foliage or lodging in leaf axils. If commercially adopted, seed meal may be quicker to apply over the crop using mechanised applicator than by mulch application.

Financial benefits

Potential financial benefits of using seed meal to control liverwort:

- The cost of moss, liverwort and weed control at despatch is estimated at 3p per 3 L pot, within a hand weeding regime, equating to £5,625 per hectare based on 2012 figures.
- There are currently no herbicides approved for use over plants under glass that will control liverwort.
- Seed meals have the potential to reduce the cost of liverwort control by reducing manual removal.
- Provision of plants to customers free from liverwort infestation.

Cost benefit analysis

Data (Table 11) for the cost of hand weeding, herbicide and loose-fill mulch application are based on 187,500 pots (3 litre, 19 cm diameter)/ha at 1.25 spacing (25 pots / m^2), allowing 25% non-cropped area for roads and general access (Hewson, A. 2012).

This study is aimed at hardy nursery stock grown as liners, therefore the analysis assumes 750,000 pots (9 cm)/ha, using a conversion factor of four to calculate the number of pots. The time involved in the operations described has been assumed to be the same for 9 cm pots as 3 L pots in this scenario. Figures are based on average costs and are for guidance only; there will be variations depending on situation, labour cost, and prevailing weed pressure.

Seed meal application would not replace a standard herbicide application as it is primarily for liverwort control. In recent trials at ADAS Boxworth, as part of the HDC Fellowship programme, seed meals gave good control of groundsel and annual meadow grass, which were used as test species. It may be possible, therefore, to substitute at least one application of Ronstar 2G with seed meal.

Action points for growers

- Further development of seed meal application, and refinement of dose rates on liverwort and phytotoxic effects on crop plants is required before any specific recommendations can be made for growers.
- Growers could consider including a proportion of Sylvafibre® or sterilised loam in potting mixes to aid liverwort reduction, particularly in short term crops (refer to Year 1 Annual Report for details).

Table 1. Cost comparison of nursery stock weed control operations (per hectare)

Programme	Task	Materials				Labour		
		Product	No. applications	Rate	Total £	Cost/hr £	Labour £	Total £
Hand weeding	Hand weeding (3/year)					8	37,500	
	Hand weeding during despatch					8	5,625	43,125
Standard herbicide programme		Ronstar 2G (£1,182/ha)	2	200 kg/ha	2,364	12	192	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	48	
		Venzar Flowable (£82/ha)	1	4.0 L/ha	82	12	24	
	Spot weeding					8	1,875	
	Hand weeding during despatch					8	1,875	6,570
***Brassica napus* + herbicide programme**		Seed meal (40p/kg)	1	4500 kg/ha	1,800	8	64	
		Oil extraction (15p/kg)	1	4500 kg/ha	675			
		Ronstar 2G (£1,182/ha)	1	200 kg/ha	1,182	12	96	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	48	
	Spot weeding					8	1,875	5,850
***Sinapis alba* 'Braco' + herbicide programme**		Seed meal (£2/kg)	1	4500 kg/ha	9,000	8	64	
		Oil extraction (15p/kg)	1	4500 kg/ha	675			
		Ronstar 2G (£1,182/ha)	1	200 kg/ha	1,182	12	96	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	24	
	Spot weeding					8	1,875	13,026
Loose-fill mulch		Bark	1	<20mm	5,625	8	1,875	
		Wastage			860			
		Flexidor 125 (£55/ha)	1	1.0 L/ha	55	12	24	
	Spot weeding/					8	936	
	Hand weeding during despatch					8	1,875	11,250

Figures, rounded up and based on average costs, are for guidance only. There will be variations depending on situation, labour cost, and prevailing weed pressure.

SCIENCE SECTION

Introduction

Liverwort growing on the surface of growing media is a major problem to the horticulturalindustry, affecting both protected and outdoor grown hardy nursery stock. The cost of moss, liverwort and weed control at despatch alone is estimated at up to 3p per 3 L pot, equating to £5,625 per hectare based on 2012 figures, depending on the weed control regime in place (Hewson, A. 2012). Zero tolerance of liverwort in certification schemes and a lack of approved chemical products make its control a technical priority for growers.

The aim of this project was to build on work completed in HDC projects HNS 126 and HNS 93c by investigating further the herbicidal effect of glucosinolate (GSL) hydrolysis products found in oil seeds on liverwort, and the suppression of liverwort growth by unknown biological or physical factors within certain growing media components.

GSLs and their hydrolysis products (isothiocyanates, ITCs) are responsible for the distinctive pungent smell and hot taste of cabbages, mustards and other brassicas and are known to have toxic effects against plants, root knot nematodes and fungal species; brassicas are successfully used in biofumigation of soils against weeds and diseases. GSLs could potentially be used to control weeds in containers; each brassica variety has a distinctive profile of one or more glucosinolates, each of which could have a different effect on liverwort.

Year 1 of this project was comprised of two trials investigating 1) the effect of brassica oil seeds (*Brassica carinata, Sinapis alba* 'Albatross'*, Camelina sativa, Brassica napus* 'RMG' and *Brassica napus* '00'), and 2) growing media amendments (Melcourt Sylvafibre®, Melcourt Growbark®, Perlite, Vital Earth Green Compost and sterilised loam) on liverwort establishment. In the seed meal trial *Sinapis alba* 'Braco', *Brassica napus* '00' and *Camelina sativa* significantly reduced liverwort establishment, whilst in the growing media amendment trial amendment with Sylvafibre® and sterilised loam significantly reduced liverwort establishment.

During year 2, two trials were carried out investigating 1) the suppressive effect of *Sinapis alba* 'Braco' seed meal application method (applied as pellets, mulch; ground seed meal, mulch; ground seed meal plus bark, mulch; ground seed meal incorporated into the growing media and a managed treatment, two applications of ground seed meal only, applied as a mulch), and dose rate on liverwort establishment and growth, and 2) the fate of the glucosinolate content of the seed meal**.** The *Sinapis alba* 'Braco' managed treatments and the seed meal with bark treatments were the most promising in terms of plant height and reduced phytotoxicity. Incorporating seed meal into growing media resulted in greater phytotoxicity and reduced root development compared to other treatments. Increased plant height occurred in the seed meal with bark (6 g) and the managed treatments (6 g), suggesting some growth promotion properties of the seed meal when applied at this dose rate.

Based on these results, year three of the project focused on the effect of *Sinapis alba* 'Braco' and *Brassica napus* ('00' oilseed rape) on liverwort establishment and growth, but using a single dose rate (6 g/pot), aiming to confirm the effect of the treatments using two different brassica varieties. A second trial investigated various methods of applying ground seed meal over the crop, to reduce time spent in carefully placing the seed meal on the growing media surface, to determine if such treatments would be practical commercially.

Year 3 Objectives

1. Seed meal suppressive effect: to investigate the use of various mulch applications of *Sinapis alba* 'Braco' and *Brassica napus* seed meals (containing glucosinolates) to control liverwort: ground seed meal, ground seed meal with bark, a managed treatment where the seed meal is re-applied at the first sight of liverwort infestation, and a mulch of seed meal ground on the day of application, from which the oil has not been extracted.

2. Effects of seed meal deposit removal method: to investigate any effect of applying the seed meal over plant leaves and either brushing or washing off any deposits, or leaving deposits in place.

Materials and methods

The trials were set up on 8 June 2011, sited under glass at John Richards Nurseries. Growing conditions, including irrigation, were managed as normal for the site. Trays were placed on benches lined with polystyrene with drainage holes.

5 kg each of *Sinapis alba* 'Braco' (Supplier: Farm Direct, Cumbria) and *Brassica napus* seeds (Selby House Farm, Stanton) were crushed and the oil extracted by Alan Brewis (Selby House Farm, Stanton, cold pressed), then reformed into pellets prior to supply. A further 5 kg of each seed variety was obtained without the oil being extracted.

Glucosinolate content of each variety of ground seedmeal (oil extracted) and whole seed (unextracted), was measured by NIAB (National Institute of Agricultural Botany) using test procedures based on British Standard BS 4289 Part 9: 1993 ISO 9167-1 1992.

The potting mix (pH 4.0) was comprised of:

- 100% Bulrush peat (standard)
- Osmocote Exact + trace elements (3-4 months, 4.0 kg/m^3)
- Lime (2.4 kg/m^3)
- Thiacloprid (Exemptor, vine weevil control, 0.3 kg/m^3)

The irrigation water (pH 8.41) used was collected rainwater, purified through iris beds:

- Alkalinity as HCO_3 84 mg/l
- Conductivity 183 µS/cm
- Nitrate-N <0.1 mg/l

For both trials, each pot was planted with a plug of *Clematis* 'Ernest Markham' (Supplier: Micropropagation Services Ltd). The plants were grown according to commercial practice, tied to canes when large enough, and cut back to the top of the cane as necessary. Three applications of maltodextrin (Majestik, 25 ml/L) were made to the *Clematis* to control two spotted mite and thrips (18 August, 25 August 2011 and 1 September 2011).

Objective 1: Seed meal suppressive effect

Experimental design

Treatments were arranged in a randomised block design with 4-fold replication (Appendix 1). Liverwort inoculum was provided by a 'spreader' pot (one pot per plot) containing liverwort. Plots consisted of 10 x 9 cm pots, placed in trays each with one liverwort 'spreader' pot.

The experiment consisted of six treatments and two seed meal varieties, within which were two control treatments (no seed meal applied, CA; and bark only applied, CB) (Table 2). *Sinapis alba* 'Braco' and *Brassica napus* seed meal were applied at a single dose rate (6 g per pot). For the managed treatments (MSa and MR) a second application of seed meal was applied at the first sight of liverwort infestation on 1 August 2011, 9 weeks after treatment.

Table 2. Seed meal suppressive effect

	Seed meal variety	Treatment
GSa	*Sinapis alba* 'Braco'	Mulch - ground seed meal
BSa	*Sinapis alba* 'Braco'	Ground seed meal + bark
MSa*	*Sinapis alba* 'Braco'	Managed treatment
USa	*Sinapis alba* 'Braco'	Unextracted seed meal, ground on day of application
GR	*Brassica napus*	Mulch - ground seed meal
BR	*Brassica napus*	Ground seed meal + bark
MR*	*Brassica napus*	Managed treatment
UR	*Brassica napus*	Unextracted seed meal, ground on day of application
CA	None	No seed meal
CB	None	Bark only, no seed meal

*For the managed treatment a second seed meal mulch application was applied once liverwort first appeared in the treatments.

Assessments

Assessments were carried out as follows:

Date	WAT*	Action	Data collection
8 June 2011	0	Set up	General observations
4 July 2012	4	Assessment	Phytotoxicity Plant height
1 August 2011	9	Assessment	Liverwort cover
1 September 2011	13	Assessment	Phytotoxicity Liverwort cover
5 October 2011	18	Assessment	Liverwort cover
24 October 2011	21	Assessment	Liverwort cover Plant height
21 November 2011	25	Assessment	Liverwort cover
19 December 2011	29	Assessment	Liverwort cover
19 January 2011	33	Assessment	Liverwort cover Root development

*WAT = weeks after treatment

Statistical analysis was carried out using GenStat Release 12.1 (PC/Windows XP).

Objective 2: Effects of seed meal deposit removal method

This unreplicated trial consisted of five treatments (Table 3) and two seed meal varieties (with the oil extracted). *Sinapis alba* 'Braco' and *Brassica napus* seed meal was applied over the plant leaves and then any deposits were removed either by brushing or washing them off, or they were left in place. For the treatment where the deposits were allowed to remain on the plants, the growing media was watered before the seed meal was applied and the foliage allowed to dry before treatment, so the seed meal would not stick to wet foliage.

The two control treatments were ground seed meal applied as a mulch and no seed meal, the results for which were collected from control treatments within Objective 1. Each plot consisted of a tray of 17 x 9 cm pots.

Observations assessed any symptoms of phytotoxicity on the *Clematis* leaves, also noting the ease of removing any deposits left during the application process, if the seed meal landed within or outside of pots during deposit removal, and how successful the removal method used was in cleaning the plants.

Table 3. Effects of seed meal deposit removal method treatments

	Seed meal variety	**Treatment**
SSa	*Sinapis alba* 'Braco'	Apply over foliage and shake off seed meal deposits
WSa	*Sinapis alba* 'Braco'	Apply over foliage and wash off seed meal deposits
GSa**	*Sinapis alba* 'Braco'	Mulch - ground seed meal
LSa	*Sinapis alba* 'Braco'	Apply over foliage and do not remove deposits
SR	*Brassica napus*	Apply over foliage and shake off seed meal deposits
WR	*Brassica napus*	Apply over foliage and wash off seed meal deposits
GR**	*Brassica napus*	Mulch - ground seed meal
LR	*Brassica napus*	Apply over foliage and do not remove deposits
CA**	None	No seed meal

**Results were collected using plots from within Objective 1 for the untreated (no seed meal) control and ground seed meal mulch.

Results and Discussion

The trials were set up and treatments applied on 8 June 2011, and the second application of ground seed meal was applied to the managed treatments at the first sight of liverwort infestation (MSa, MR) on 1 August 2011 (9 weeks after treatment). The *Clematis* were irrigated as normal for the crop, but this resulted in the liverwort inoculum drying out to an unacceptable degree, therefore they were stood in saucers of water from 27 June 2011 (Figure 2). However, pots adjacent to the liverwort spreaders were affected disproportionately, regardless of treatment, and were therefore excluded from statistical analysis.

Figure 2. Desiccated liverwort 'spreader' pots were stood in saucers of water (left), following which they grew vigorously into neighbouring pots.

The results of several trial plots were confounded by accidental application of gluten by nursery staff on 22 September 2011, therefore these plots have been excluded from the statistical analysis for liverwort area; these plots included three of the four control treatment plots with bark (CB), therefore these results are not be reported. Plant height and phytotoxicity data were collected prior to this date, therefore all plots were included in the statistical analysis for these datasets. The gluten appeared to attract rats, which caused damage to a number of other pots, and these were also excluded from statistical analysis.

Objective 1: Seed meal suppressive effect

Liverwort cover

Throughout the majority of this trial, least liverwort established under the *Brassica napus* seed meal with bark treatment (BR), although low levels also established in the managed *Brassica napus* (MR) and *Sinapis alba* 'Braco' with bark (BSa) treatments; the greatest amount of liverwort established in the ground *Brassica napus* (GR) and the untreated control (CA) (Figure 3). Lowest levels of *liverwort* cover were most successfully maintained for 33 weeks in the two treatments including bark (BR, BSa); with less than 5% liverwort cover maintained throughout the trial in the *Brassica napus* treatment.

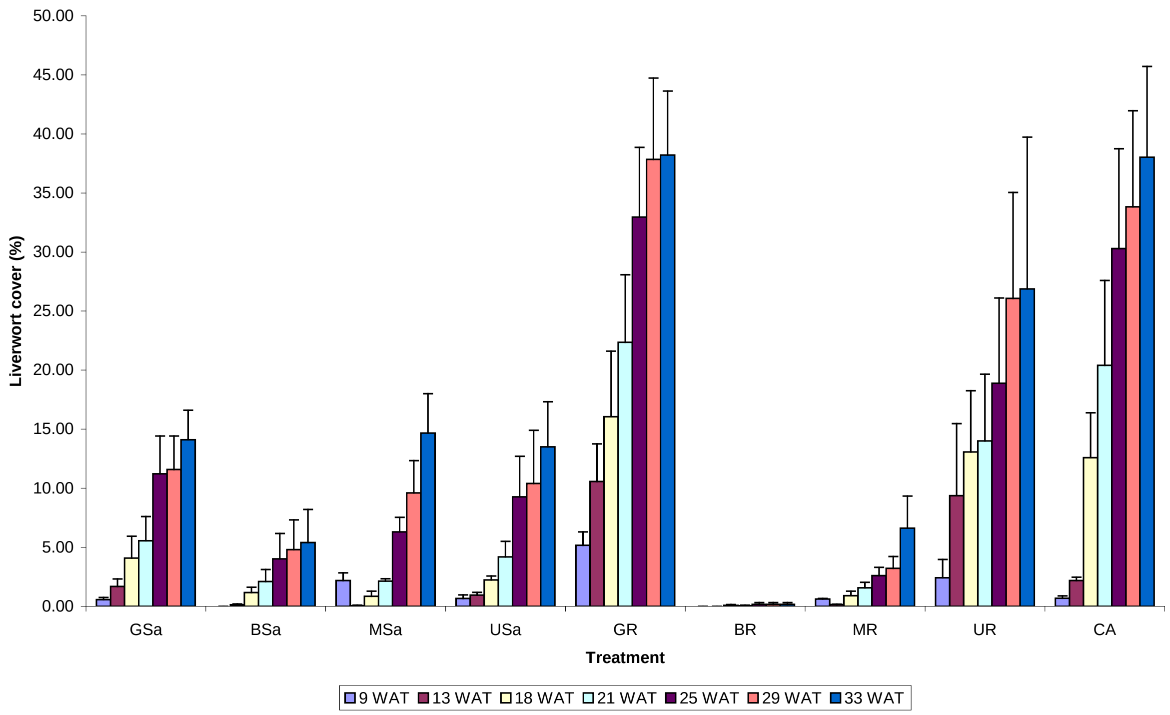

Figure 3. Percentage liverwort cover: GSa = Ground *Sinapis alba* 'Braco' seed meal*, BSa* = Ground *Sinapis alba* 'Braco' seed meal+bark, MSa = *Sinapis alba* 'Braco', managed treatment, USa = *Sinapis alba* 'Braco', unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus*, managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied. WAT = weeks after treatment

Statistical analysis using ANOVA indicated significant differences in liverwort cover due to the treatments from 9 weeks after treatment ($F_{8,14}$ = 5.17, P<0.01, Table 4), the differences were very highly significant from 25 weeks after treatment ($F_{8,14}$ = 6.30, P<0.001,Table 5).

Table 4. Analysis of variance (ANOVA) comparing liverwort cover, 9 WAT.

Source of variation	d.f.	s.s.	m.s.	v.r.	F. pr.	
Block	2	1.634	0.817	0.50	0.617	
Treatment	8	67.673	8.459	5.17	0.004	**
Residual	14	22.907	1.636			
Total	24	87.699				

Table 5. Analysis of variance (ANOVA) comparing liverwort cover, 33 WAT.

Source of variation	d.f.	s.s.	m.s.	v.r.	F. pr.	
Block	2	84.70	42.35	0.53	0.599	
Treatment	8	4007.64	500.96	6.30	<.001	***
Residual	14	1114.07	79.58			
Total	24	4685.28				

Untreated control. From 18 weeks after treatment, liverwort cover was greater in the untreated control (CA) than all other treatments except for the ground *Brassica napus* seed meal (GR); however, the differences were only consistently significant in the treatments with bark (BSa, BR), the managed treatments (MSa, MR), and the unextracted *Sinapis alba* 'Braco' treatment.

Ground seed meal. More liverwort established in the ground *Brassica napus* (GR) seed meal treatments than the ground *Sinapis alba* 'Braco' (GSa, significant) and untreated control (CA, not consistently significant) treatments throughout the trial. There was, however, significantly less liverwort in the ground *Sinapis alba* 'Braco' (GSa) compared with the untreated control (CA) 21, 29 and 33 weeks after treatment. Although there was generally more liverwort cover in the ground *Sinapis alba* 'Braco' (GSa) treatment than other *Sinapis alba* 'Braco' treatments (BSa, MSa, USa), the differences were not significant.

Managed treatments. Liverwort cover was against the trend 9 weeks after treatment in the managed *Sinapis alba* 'Braco' treatment; this was due to three pots with greater than average liverwort cover, and evidenced by the greater variability within plots compared with subsequent assessments. From 13 weeks after treatment, although liverwort cover was greater in the ground *Sinapis alba* 'Braco' treatment (GSa) than the managed *Sinapis alba* 'Braco' treatment (MSa), the differences were not significant. However, for *Brassica napus*, liverwort cover due to the managed treatment (MR) was significantly less than in the ground seed meal (GR) throughout the trial, and the untreated control (CA) from 18 weeks after treatment.

Bark. The addition of bark to the seed meal treatments (BSa, BR) resulted in reduced liverwort cover when compared to application of seed meal alone (GSa, GR), and the untreated control (CA). The differences between the *Sinapis alba* 'Braco' seed meal treatments, with and without bark (GSa, BSa), were not significant. There was significantly less liverwort cover in the *Brassica napus* treatment with bark (BR) than without (GR) from nine weeks after treatment. Significantly less liverwort cover established in the seed meal treatments with bark (BSa, BR) than the untreated control (CA) from 18 weeks after treatment.

Oil extraction. There was no consistent trend in the effect of extracting the oil from the seed meal on liverwort cover. There was no significant difference in liverwort cover in the *Sinapis alba* 'Braco' seed meal treatments where the oil had been retained (USa) compared to treatments where the oil was extracted (GSa, BSa, MSa), suggesting there may be no need to remove the oil. However, for the *Brassica napus* seed meal treatments, there was significantly more liverwort cover in the treatment where the oil was retained (UR) than the seed meal with bark (BR) and managed (MR) treatments from 18 weeks after treatment.

Regression analysis (Table 6) indicated that by 13 WAT 100% of pots four treatments (BSa, MSa, BR, MR) had less than 5% liverwort cover, and the *Brassica napus* treatment with bark (BR) continued to maintain this level until 33 weeks after treatment.

Table 6. Liverwort cover: proportion of pots with less than 5% liverwort cover. GSa = Ground *Sinapis alba* 'Braco' seed meal, BSa = Ground *Sinapis alba* 'Braco' seed meal+bark, MSa = *Sinapis alba* 'Braco', managed treatment, USa = *Sinapis alba* 'Braco', unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus*, managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied, CB = Bark only applied, no seed meal. WAT = weeks after treatment

	13 WAT		18 WAT		33 WAT	
Treatment	**Mean (%)**	**s.e.**	**Mean (%)**	**s.e.**	**Mean (%)**	**s.e.**
GSa	87	0.05	78	0.093	31	0.115
BSa	100	0	92	0.061	79	0.099
MSa	100	0	95	0.052	55	0.133
USa	88	0.045	76	0.092	36	0.114
GR	52	0.085	35	0.123	18	0.111
BR	100	0	100	0.001	100	0.001
MR	100	0	96	0.040	65	0.111
UR	75	0.108	43	0.180	34	0.194
CA	85	0.055	60	0.117	10	0.080

Phytotoxicity and plant quality

Phytotoxicity was assessed prior to the application of gluten, therefore the results were unaffected by this event. Phytotoxicity symptoms were recorded as quality scores on a scale of 1-5 (1 = plant death, 4 = commercially acceptable, 5 = no damage observed). Data was collected 4 and 13 weeks after treatment. Greatest phytotoxicity was recorded 4 weeks after treatment, mainly in the *Sinapis alba* 'Braco' treatments. Although there was some significant phytotoxicity due to both seed meal varieties after 13 weeks, only treatments using *Sinapis alba* 'Braco' treatments in week four were not commercially acceptable (Figure 4).

Statistical analysis using ANOVA indicated a very highly significant difference in plant quality due to the treatments four weeks after treatment ($F_{9,27}$ = 22.88, P<0.001, Table 7), and a significant difference in plant quality 13 weeks after treatment ($F_{9,27}$ = 2.66, P<0.05, Table 8). The data indicates significantly greater phytotoxicity due to all treatments than control treatments (CA and CB), except for the *Brassica napus* where the oil had not been extracted

(UR) four weeks after treatment; however by 13 weeks after treatment significant phytotoxicity only occurred in the *Sinapis alba* ‘Braco’ treatment with bark (BSa) and the managed treatment (MSa).

For both the *Sinapis alba* ‘Braco’ (4 and 13 WAT) and *Brassica napus* (4 WAT) less phytotoxicity resulted from treatments where the oil was not extracted prior to application.

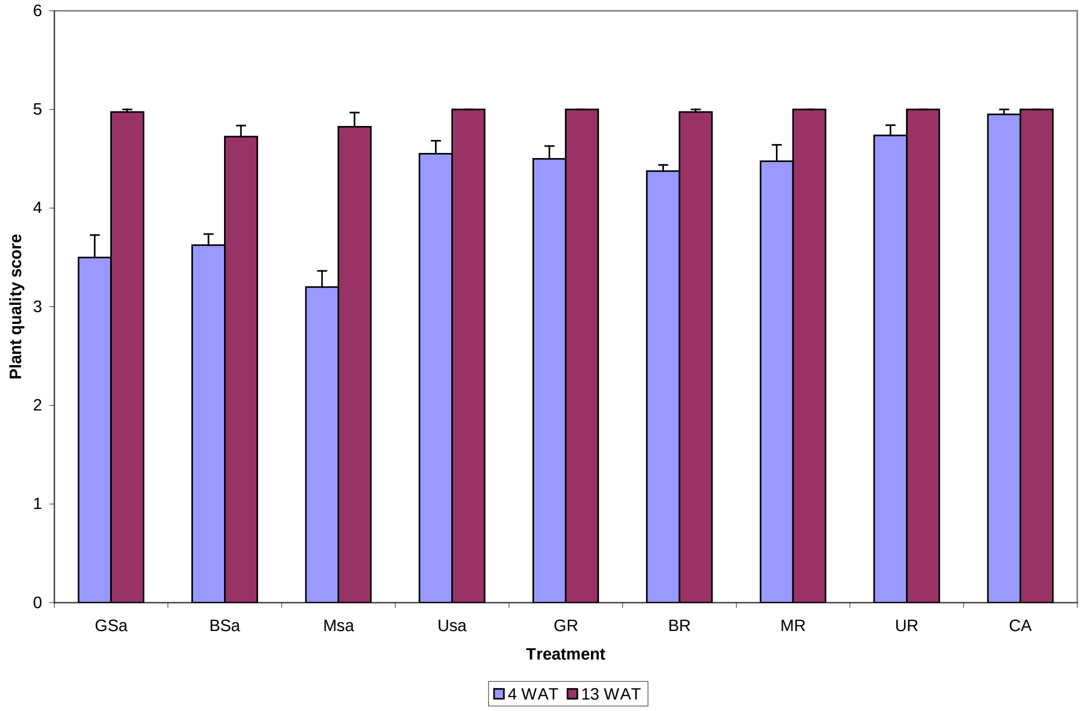

Figure 4. Plant quality scores: 1 (plant death) to 5 (no damage observed). GSa = Ground *Sinapis alba* ‘Braco’ seed meal*, BSa* = Ground *Sinapis alba* ‘Braco’ seed meal+bark, MSa = *Sinapis alba* ‘Braco’, managed treatment, USa = *Sinapis alba* ‘Braco’, unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus*, managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied, CB = Bark only applied, no seed meal. WAT = weeks after treatment

Table 7. Analysis of variance (ANOVA) comparing plant quality 4 WAT.

Source of variation	d.f.	s.s.	m.s.	v.r.	F. pr.	
Block	3	0.17489	0.05830	0.85	0.481	
Treatment	9	14.18511	1.57612	22.88	<.001	***
Residual	27	1.85956	0.06887			
Total	39	16.21956				

Table 8. Analysis of variance (ANOVA) comparing plant quality 13 WAT.

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F. pr.**	
Block	3	0.03800	0.01267	0.92	0.445	
Treatment	9	0.33000	0.03667	2.66	0.024	*
Residual	27	0.37200	0.01378			
Total	39	0.74000				

Regression analysis (Table 9) indicated that by 13 WAT over 97% of all plants had plant quality scores greater than 4, indicating that although there was some phytotoxicity, which may have caused setback to the plants, they were able to grow through it.

Table 9. Plant quality: percentage of plants with quality scores greater than 4, assessed 4 and 13 weeks after treatment. Scores: 1 (plant death) to 5 (no damage observed). GSa = Ground *Sinapis alba* 'Braco' seed meal, BSa = Ground *Sinapis alba* 'Braco' seed meal+bark, MSa = *Sinapis alba* 'Braco', managed treatment, USa = *Sinapis alba* 'Braco', unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus*, managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied, CB = Bark only applied, no seed meal. WAT = weeks after treatment

Treatment	4 WAT		13 WAT	
	Mean (%)	s.e.	Mean (%)	s.e.
GSa	18	0.084	98	0.017
BSa	28	0.099	85	0.039
MSa	20	0.088	93	0.029
USa	63	0.107	100	0
GR	65	0.105	100	0
BR	55	0.110	98	0.017
MR	60	0.108	100	0
UR	79	0.092	100	0
CA	98	0.034	100	0
CB	100	0.001	100	0

Plant height

Plant height was assessed only once as the plants were subsequently trimmed back to the top of the cane. Plant height was assessed prior to the application of gluten, therefore the results were unaffected by this event (Figure 5). Statistical analysis using ANOVA indicated a very highly significant effect on plant height due to treatment ($F_{9,27} = 5.85$, $P<0.001$, Table 10). There was a clear overall difference in plant height due to seed meal variety, with greater plant height achieved in the *Brassica napus* seed meal treatments (GR, BR, MR and UR), compared to the *Sinapis alba* 'Braco' treatments (GSa, BSa, MSa and USa).

Untreated control. Plant height was greater in all *Brassica napus* seed meal treatments than the untreated control (CA); the difference was significant where the oil had not been extracted (UR) and where bark had been applied (BR). Plant height was less than the untreated control (CA) in all *Sinapis alba* 'Braco' seed meal treatments where the oil had been extracted from the seed meal (GSa, BSa and MSa), although this difference was only significant in the managed treatment (MSa); where the oil had not been extracted (USa) plant height was greater than in the untreated control (CA).

Managed treatment. The second application of seed meal in both managed treatments (MSa and MR), where dose rate was doubled by a second application of seed meal, did have an adverse effect on plant height within seed meal varieties. However, although plant height was significantly less than both control treatments (CA, CB) due to application of *Sinapis alba* 'Braco' seed meal (MSa) in the managed treatment, there was no significant difference between *Brassica napus* managed treatment (MR) and the controls (CA, CB).

Bark. Plant height was significantly greater in the control treatment with bark (CB) than the untreated control (CA), all *Sinapis alba* 'Braco' seed meal treatments (MSa, BSa, GSa and USa) and the managed *Brassica napus* seed meal treatment (MR). Plant height was also significantly greater in the *Brassica napus* seed meal treatment with bark (BR) than most *Sinapis alba* 'Braco' treatments (treatment USa, where the oil had not been extracted was the exception), and the untreated control (CA). However, plant height due to the *Sinapis alba* 'Braco' treatment with bark (BSa) was not significantly greater than other *Sinapis alba* 'Braco' treatments.

Oil extraction. The extraction of the oil had a generally positive effect on plant height within each seed meal variety, however for *Sinapis alba* 'Braco', the difference was only significant between unextracted seed meal treatment (USa), and the ground seed meal (GSa) and managed (MSa) treatments. Plant height in the *Brassica napus* unextracted seed meal was significantly greater than the untreated control (CA).

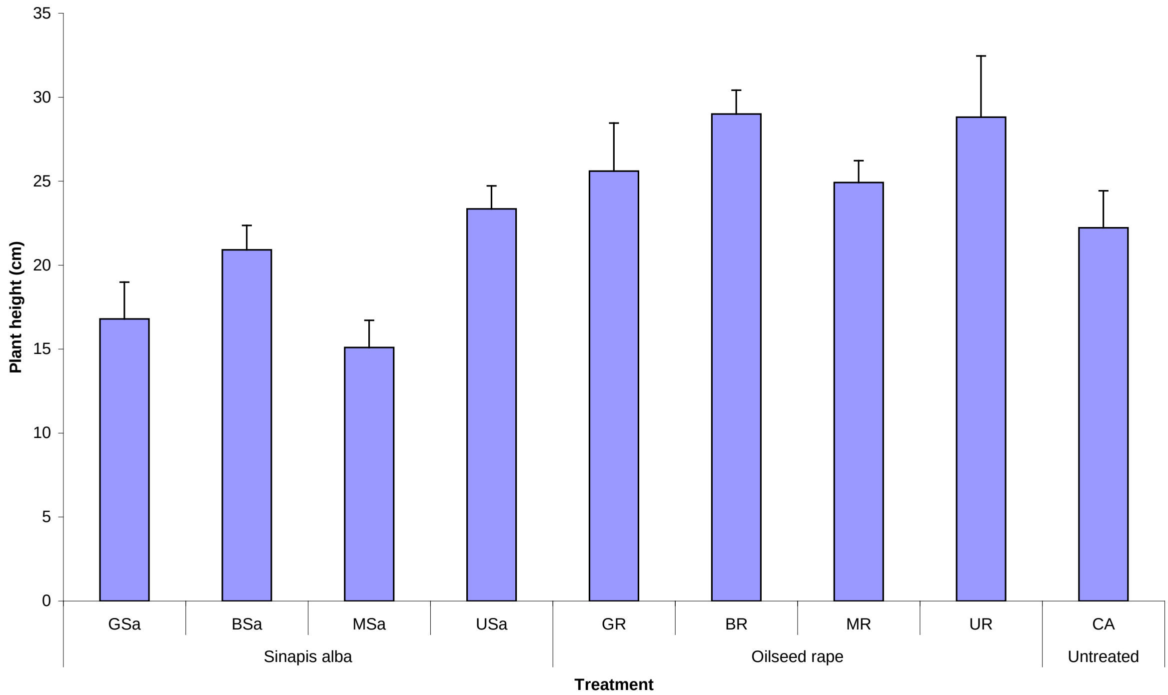

Figure 5. Plant height 4 WAT. GSa = Ground *Sinapis alba* 'Braco', BSa = Ground *Sinapis alba* 'Braco'+bark, MSa = *Sinapis alba* 'Braco' managed treatment, USa = *Sinapis alba* 'Braco', unextracted seed meal, GR = Ground *Brassica napus* seed meal, BR = Ground *Brassica napus* seed meal+bark, MR = *Brassica napus* managed treatment, UR = *Brassica napus*, unextracted seed meal, CA = No seed meal applied, CB = Bark only applied, no seed meal. WAT = weeks after treatment

Table 10. Analysis of variance (ANOVA) comparing plant height 4 WAT.

Source of variation	d.f.	s.s.	m.s.	v.r.	F. pr.	
Block	3	46.69	15.56	0.85	0.480	
Treatment	9	966.71	107.41	5.85	<.001	***
Residual	27	496.11	18.37			
Total	39	1509.51				

Root development

Root development, assessed 33 WAT, was compared with the control and graded on a scale of 1-5, as in year 2 (Figure 6), ranging from no roots to root development comparable with those found in the control plots. The data collected showed no adverse effects of any of the treatments, with the majority of plants having a score of 5, and a few having a score of 4.

Figure 6. Root scores (left to right): 1 = no visible root, 2 = some visible root, 3 = visible root growing down, 4 = slightly reduced rooting (compared to control), 5 = root growth comparable with control (Image from HNS 175 year 2 report).

Objective 2: Effects of seed meal deposit removal method

This was an unreplicated trial, therefore no statistical analysis was carried out. The results of the trial treatments were confounded by accidental application of gluten by nursery staff on 22 September 2011(16 WAT) therefore only the data collected prior to this event is reported. The data for the mulch applications (GSa and GR) and the untreated control (CA) were sourced from Objective 1.

Liverwort cover

Liverwort cover was least in the untreated control (CA) and ground *Sinapis alba* 'Braco' (GSa) than all treatments, both 9 and 13 weeks after treatment (Figure 7). Of the treatments where seed meal was applied over the crop, after 13 weeks there was least liverwort cover where seed meal deposits were not removed from leaves (LSa, LR). Treatments where seed meal deposits were shaken off leaves (SSa, SR) resulted in more liverwort cover than other treatments where seed meal was applied, suggesting that too little seed meal landed on the pots, effectively reducing the dose rate.

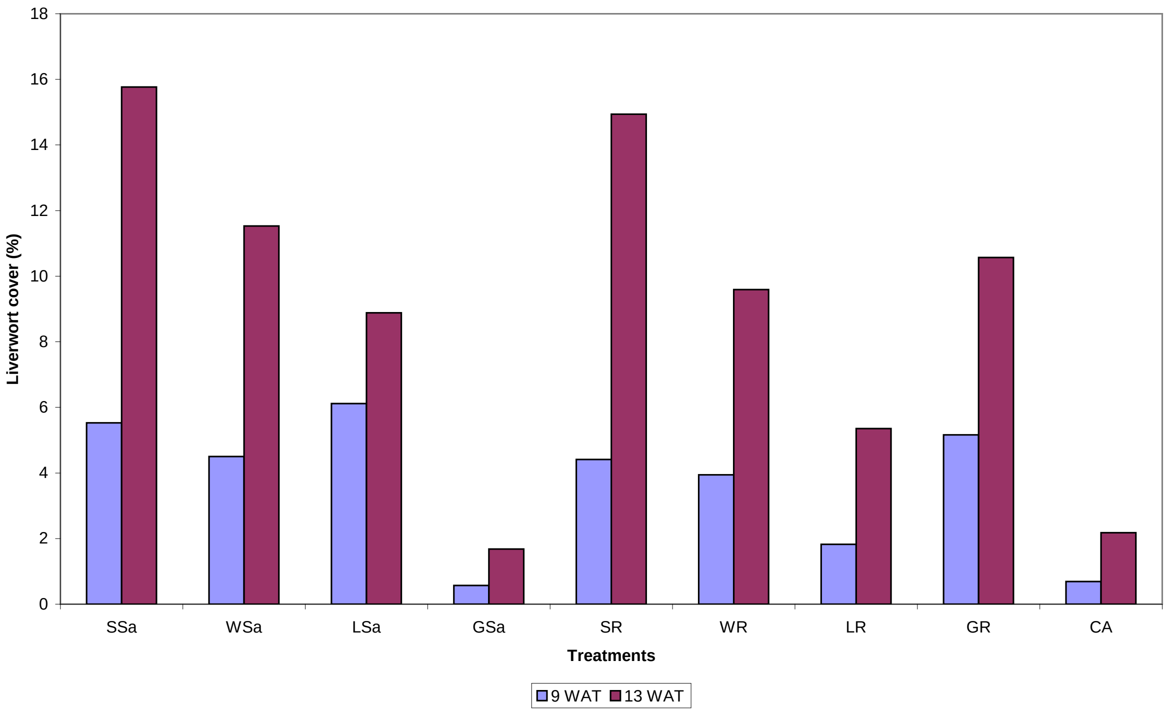

Figure 7. Liverwort cover up to 9 and 13 WAT. SSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and shake off seed meal deposits, WSa = Apply *Sinapis alba* 'Braco' seed meal over foliage and wash off seed meal deposits, LSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and deposits not removed, GSa = Ground *Sinapis alba* 'Braco' seed meal mulch, SR = Apply *Brassica napus* seed meal over foliage and shake off seed meal deposits, WR = Apply *Brassica napus* over foliage and wash off seed meal deposits, LR = Apply *Brassica napus* over foliage and deposits not removed, GR = Ground *Brassica napus* seed meal mulch, CA = No seed meal applied. WAT = weeks after treatment

Plant quality

Plant quality scores (Figure 8) of 4 and above were deemed commercially acceptable, and were recorded in the control treatment (CA) at both assessments (4 and 13 WAT). Plant quality scores below 4 were only recorded in three *Sinapis alba* 'Braco' treatments (4 WAT), where the seed meal was applied over the foliage and then shaken off (SSa), where seed meal deposits were not removed (LSa), and where the seed meal was applied as a mulch (GSa). Less phytotoxicity was observed when the seed meal deposits were washed away (WSa) than if they were shaken off (SSa), left in place (LSa) or applied as a mulch (GSa). However, for the *Brassica napus* less phytotoxicity was observed where seed meal deposits were shaken off (SR) rather than washed off (WR), applied as a mulch (GR) or left in place (LR). Plant quality had improved after 13 weeks with all plants achieved a score of four and above, and the majority achieving a score of five, suggesting recovery from the adverse effects of seed meal application.

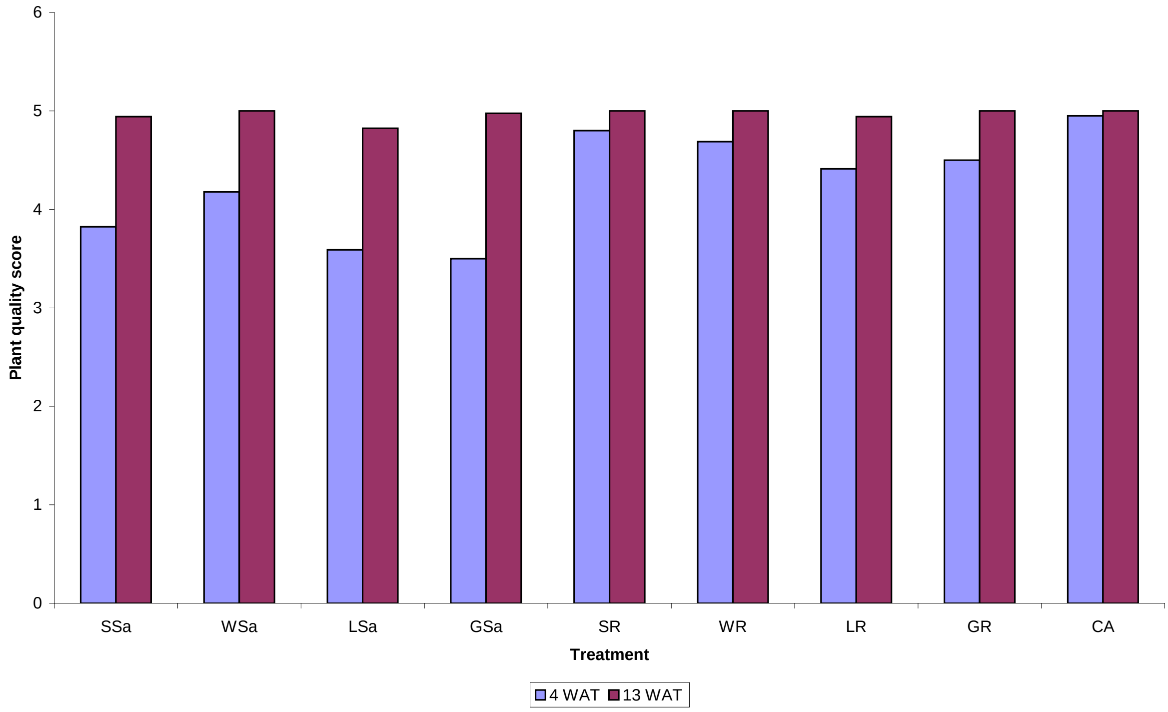

Figure 8. Plant quality scores: 1 (plant death) to 5 (no damage observed). SSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and shake off seed meal deposits, WSa = Apply *Sinapis alba* 'Braco' seed meal over foliage and wash off seed meal deposits, LSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and deposits not removed, GSa = Ground *Sinapis alba* 'Braco' seed meal mulch, SR = Apply *Brassica napus* seed meal over foliage and shake off seed meal deposits, WR = Apply *Brassica napus* over foliage and wash off seed meal deposits, LR = Apply *Brassica napus* over foliage and deposits not removed, GR = Ground *Brassica napus* seed meal mulch, CA = No seed meal applied. WAT = weeks after treatment

Plant height

Plant height (Figure 9), assessed four weeks after treatment, was adversely affected by all seed meal treatments with the exception of the ground *Brassica napus* treatment (GR), when compared with the untreated control (CA); the greatest effect was seen where seed meal deposits were not removed from the leaves (LSa, LR). Comparing treatments where the seed meal was applied over the plants, within each seed meal variety, least adverse effect was caused where the seed meal was shaken off the leaves.

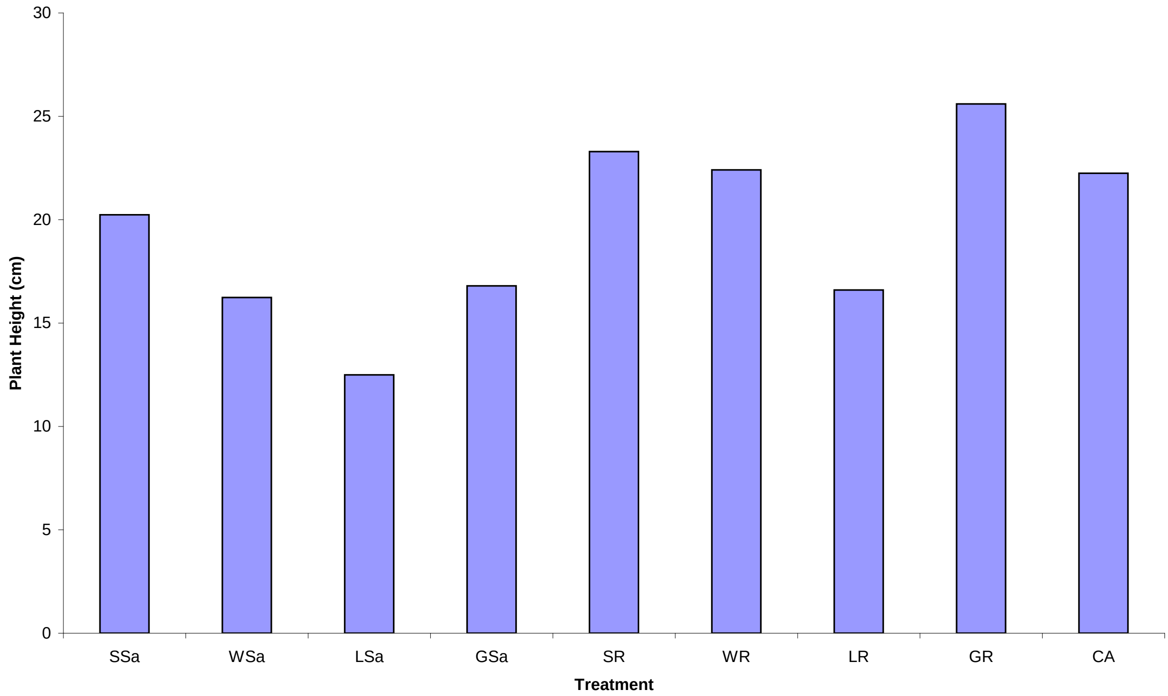

Figure 9. Plant height (cm). SSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and shake off seed meal deposits, WSa = Apply *Sinapis alba* 'Braco' seed meal over foliage and wash off seed meal deposits, LSa = Apply *Sinapis alba* 'Braco' ground seed meal over foliage and deposits not removed, GSa = Ground *Sinapis alba* 'Braco' seed meal mulch, SR = Apply *Brassica napus* seed meal over foliage and shake off seed meal deposits, WR = Apply *Brassica napus* over foliage and wash off seed meal deposits, LR = Apply *Brassica napus* over foliage and deposits not removed, GR = Ground *Brassica napus* seed meal mulch, CA = No seed meal applied. WAT = weeks after treatment

Financial benefits

Potential financial benefits of using seed meal to control liverwort:

- The cost of moss, liverwort and weed control at despatch is estimated at 3p per 3 L pot, within a hand weeding regime, equating to £5,625 per hectare based on 2012 figures.
- There are currently no herbicides approved for use over plants under glass that will control liverwort.

- Seed meals have the potential to reduce the cost of liverwort control by reducing manual removal.
- Provision of plants to customers free from liverwort infestation.

Cost benefit analysis

Data (Table 11) for the cost of hand weeding, herbicide and loose-fill mulch application are based on 187,500 pots (3 litre, 19 cm diameter)/ha at 1.25 spacing (25 pots / m^2), allowing 25% non-cropped area for roads and general access (Hewson, A. 2012). This study is aimed at hardy nursery stock grown as liners, therefore the analysis assumes 750,000 pots (9 cm)/ha, using a conversion factor of four to calculate the number of pots. The time involved in the operations described has been assumed to be the same for 9 cm pots as 3 L pots in this scenario. Figures are based on average costs and are for guidance only; there will be variations depending on situation, labour cost, and prevailing weed pressure.

Seed meal programme

The cost of the seed and oil extraction would vary depending on quantity and source, larger quantities being more cost effective. For this analysis oil extraction costs are based on tonnes of seed and seed prices per 50 kg. The labour required for seed meal application has been assumed comparable to Ronstar 2G, although some development would be required to produce an even grade of ground seed meal that could be applied via an applicator. The unextracted seed meal would need to be ground prior to application.

Seed meal application would not replace a standard herbicide application as it is primarily for liverwort control. In recent trials at ADAS Boxworth, as part of the HDC Fellowship programme, seed meals gave good control of groundsel and annual meadow grass, which were used as test species. It may be possible, therefore, to substitute at least one application of Ronstar 2G with seed meal.

Hand weeding

It is likely that a high percentage of liners will generally be grown under protection and therefore hand weeded; there are currently no herbicides approved for use under glass that will control liverwort. Hand weeding costs are calculated using ADAS figures.

Standard herbicide programme – outdoor production

For outdoor liner production, a standard herbicide programme, including liverwort control, would include two alternating applications each of Ronstar 2G (oxadiazon, 200 kg/ha,.

Table 11. Cost comparison of nursery stock weed control operations (based on Hewson, 2012)

Programme	Task	Materials				Labour		
		Product	No. applications	Rate	Total £	Cost/hr £	Labour £	Total £
Hand weeding	Hand weeding (3/year)					8	37,500	
	Hand weeding during despatch					8	5,625	43,125
Standard herbicide programme		Ronstar 2G (£1,182/ha)	2	200 kg/ha	2,364	12	192	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	48	
		Venzar Flowable (£82/ha)	1	4.0 L/ha	82	12	24	
	Spot weeding					8	1,875	
	Hand weeding during despatch					8	1,875	6,570
***Brassica napus* + herbicide programme**		Seed meal (40p/kg)	1	4500 kg/ha	1,800	8	64	
		Oil extraction (15p/kg)	1	4500 kg/ha	675			
		Ronstar 2G (£1,182/ha)	1	200 kg/ha	1,182	12	96	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	48	
	Spot weeding					8	1,875	5,850
***Sinapis alba* 'Braco' + herbicide programme**		Seed meal (£2/kg)	1	4500 kg/ha	9,000	8	64	
		Oil extraction (15p/kg)	1	4500 kg/ha	675			
		Ronstar 2G (£1,182/ha)	1	200 kg/ha	1,182	12	96	
		Flexidor 125 (£55/ha)	2	1.0 L/ha	110	12	24	
	Spot weeding					8	1,875	13,026
Loose-fill mulch		Bark	1	<20mm	5,625	8	1,875	
		Wastage			860			
		Flexidor 125 (£55/ha)	1	1.0 L/ha	55	12	24	
	Spot weeding/					8	936	
	Hand weeding during despatch					8	1,875	11,250

Figures, rounded up and based on average costs, are for guidance only. There will be variations depending on situation, labour cost, and prevailing weed pressure.

£1,182/ha) and Flexidor 125 (isoxaben, 1.0 L/ha, £55/ha) (Hewson, 2012). Of these, liverwort is moderately susceptible to Ronstar 2G; liverwort is susceptible to Venzar Flowable (lenacil, 4.0 L/ha, £82/ha), which may be applied over dormant crops during late January, but there is risk of crop damage, and this product is not applicable in many situations.

Loose-fill mulch

Loose-fill mulch based on one application of bark (<20mm depth) by machine, excluding depreciation and annual charges. Costs may be higher for hand application

Conclusions

Objective 1: Seed meal suppressive effect

The treatments using both seed meal varieties with bark showed greatest promise in these trials, with the *Brassica napus* maintaining exceptionally low levels of liverwort cover (<5%) for the full 33 weeks of the trial. During year 2 of this study it was demonstrated that glucosinolates levels can reduce to unmeasurable quantities within a few weeks, suggesting that the main effect could be due to the mulching effect of the seed meal, particularly at higher dose rates. In the absence of a 'bark only' control during year 3 it has not been possible to established if least liverwort would have established in the bark treatments with or without seed meal. However, in commercial practice, where bark mulch has been applied, liverwort often colonises around the edges of pots, or where the growing media has become exposed, particularly where the bark depth is insufficient.

Plant quality and plant height both followed a similar trend, with a greater adverse effect due to treatments with *Sinapis alba* 'Braco' seed meal than the *Brassica napus*, and greatest adverse effect due to the managed treatments for both seed meal varieties; these effects had lessened by 13 weeks after treatment, but as this study did not include finished plants the longer term effect is unknown. Clematis was used as the subject due to its sensitivity to herbicides, suggesting that minimal phytotoxic effects may be found on other subjects, and this may allow scope to optimise the dose rate. In year 2 less liverwort established in the managed treatments than the bark treatments, but the reverse occurred in year 3.

The cost of seed meal application as part of a herbicide programme compared favourably with current alternatives, particularly hand weeding. The increased cost of including seed meal in the bark treatments may be offset by greater liverwort control and reduced hand weeding.

There appeared to be no difference in the condition of the *Sinapis alba* 'Braco' seed meal regardless of whether the oil had been extracted or not, however more mould appeared to grow on the *Brassica napus* seed meal where the oil had not been extracted than where it had. One or two seedlings of both seed meal varieties emerged in the unextracted treatments, suggesting a potential weed issue where seeds remain are not adequately ground and remain viable. Less phytotoxicity and greater plant height were generally found in the unextracted treatments, but more liverwort established, although at relatively low levels using *Sinapis alba* 'Braco'. The benefits of using unextracted seed meal are that the seeds could be stored whole on the nursery, and then ground on site prior to application. This would reduce the potential for glucosinolate levels to dissipate during storage, although studies have established that under the correct conditions seed meal can be stored for up to 30 months without deterioration (Morra, M. J. & Borek, V. 2010).

Objective 2: Effects of seed meal deposit removal method

Application of seed meal over the crop on a trial basis, with a small batch of plants, was easily managed, but on a commercial scale production of consistently sized seed meal granules would be required to enable application using a motorised knapsack.

Least liverwort established in the treatments where seed meal deposits were left on the leaves, but greatest phytotoxicity and smaller plants were also recorded in these treatments. Seed meal was easily removed from plants, however the recommendation would be to apply seed meal to dry foliage and tap off to avoid any fungal or phytotoxicity problems due to any seed meal sticking to the foliage or lodging in leaf axils. If commercially adopted application of seed meal over the crop may be more practical than mulch application.

Summary

The most promising treatments from these trials were the seed meal with bark, and the managed treatment, where a second application of seed meal was applied. Of the two seed meals varieties tested, the *Brassica napus* provide greatest liverwort control and when mixed with bark the control continued for the duration of the trial. However, application of seed meal with bark would have the disadvantage of higher costs (bark and application). Seed meal could more easily be applied alone through a granular applicator, but the risk of phytotoxicity would be increased.

These results confirm the conclusions from year 2, in that future work should be based around the managed and seed meal with bark treatments, further investigating any effect on plant growth and development, liverwort establishment and phytotoxicity across a range of plant species. Comparison of the effects of a ‘bark only’ control with a seed meal plus bark treatment is necessary to clarify the effects of the individual components of these treatments, potentially highlighting any synergistic effects. As these studies have been targeted at liners, the longer term effects of seed meal application have not been studied, but would merit investigation. The work looking at the effect of seed meal left on leaves suggested that there may be activity via the laminar, but further work would be required to establish this.

Preparation and application of the seed meal needs to be refined. Use of industrial grinding equipment to produce a consistent, optimum granule size, and application using a motorised knapsack to improve efficiency are needed to help bring costs down, as would extracting oil from larger quantities, and bulk buying of seed or seed meal.

Seed meals could be used as part of a herbicide programme, and there could be a synergistic interaction with standard herbicides (including Flexidor 125, Ronstar 2G and Venzar Flowable), which requires investigation.

Seed meal treatments could also be combined with Melcourt Sylvafibre® and sterilised loam to identify if a combination of seed meal treatments and growing media amendments could reduce liverwort cover, possibly providing the opportunity to use lower seed meal dose rates. Such studies should take account of the current move towards use of reduced peat and peat free growing media.

Further work testing seed meals on a broad range of nursery weed species could also identify further options for weed control, and reducing herbicide application.

Technology transfer

- Article for HDC News

Acknowledgements

The assistance of John Richards Nurseries who provided growing media, fertiliser, and bark, and managed the trials is gratefully acknowledged.

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Morra, M. J. & Borek, V. 2010. Glucosinolate preservation in stored Brassicaceae seed meals. *Journal of Stored Products Research*, 46(2): 98-102.

Appendix 1. Trial layout

Objective 1: Seed meal suppressive effect

	Seed meal variety	Treatment
1	*Sinapis alba* 'Braco'	Mulch - ground seed meal
2	*Sinapis alba* 'Braco'	Bark + seed meal
3	*Sinapis alba* 'Braco'	Managed treatment
4	*Sinapis alba* 'Braco'	Unextracted seed meal, ground on day of application
5	*Brassica napus*	Mulch - ground seed meal
6	*Brassica napus*	Bark + seed meal
7	*Brassica napus*	Managed treatment
8	*Brassica napus*	Unextracted seed meal, ground on day of application
9	None	No seed meal
10	None	Bark only, no seed meal

Objective 2: Effects of seed meal deposit removal method

	Seed meal variety	Treatment
SSA	*Sinapis alba* 'Braco'	Applied over the top of whole tray of plants and shaken off
LSA	*Sinapis alba* 'Braco'	Applied over the top of whole tray of plants and left
WSA	*Sinapis alba* 'Braco'	Applied over the top of whole tray of plants and washed off
SR	*Brassica napus*	Applied over the top of whole tray of plants and shaken off
LR	*Brassica napus*	Applied over the top of whole tray of plants and left
WR	*Brassica napus*	Applied over the top of whole tray of plants and washed off

Plot	Treatment	Plot	Treatment
3	7	1	8
4	4	2	5
7	3	5	10
8	2	6	1
11	8	9	9
Plot	**Treatment**	**Plot**	**Treatment**
12	10	10	6
15	2	13	9
16	3	14	1
19	7	17	6
20	4	18	5
Plot	**Treatment**	**Plot**	**Treatment**
23	2	21	10
24	1	22	6
27	7	25	8
28	3	26	4
31	3	29	9
Plot	**Treatment**	**Plot**	**Treatment**
32	1	30	5
35	9	33	8
36	7	34	2
39	4	37	5
	Concrete path and door		
40	10	38	6
	LR		SR
	WSA		LSA
	WR		SSA
	LR		SR
	WSA		LSA
	WR		SSA

Block 1

Block 2

Block 3

Block 4

Appendix 2. *Glucosinolate content*

Year 3: Glucosinolate content of *Sinapis alba* 'Braco', and *Brassica napus,* seed ground or whole

	Brassica napus whole	***Brassica napus*** ground	***Sinapis alba*** **'Braco'** whole	***Sinapis alba*** **'Braco'** ground
	Glucosinolate content (µmol/g)			
Glucoberin	0.18	0.47	-	-
Progoitrin	5.25	7.99	-	-
Epi Progoitrin	-	-	-	-
Glucoraphanin	-	-	-	-
Glucoalyssin	0.89	1.13	-	-
Gluconapin	2.31	3.39	-	-
40H glucobrassicin	1.68	2.59	-	-
Glucobrassicanapin	0.43	0.69	-	-
Glucobrassicin	0.09	0.14	-	-
Gluconarsturtiin	-	-	-	-
Neo glucobrassicin	0.08	0.13	-	-
Glucosinalbin	-	-	202.1	179.69
Total glucosinolate content	10.91	**16.53**	**202.1**	**179.69**