Grower summary

HNS 119

Reducing levels of Verticillium wilt and nematodes in soils using green manure crops

Final Report 2004

Project Title:	Reducing levels of Verticillium wilt and nematodes in soils using green manure crops.
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Field-grown nursery stock: Field-grown nursery stock: Field levels of *Verticillium* wilt (*V.dahliae*) and nematode reduction by the use of two different green manure crops

Headline

- Specific green manure crops with known nematicidal effects reduce the number and type of free-living nematodes and incidence of *Verticillium* wilt propagules in a field situation during a 12-month growing period.
- Using Sudan grass can save around £5200/ha for nematode and *Verticillium* wilt control compared to methyl bromide fumigation.
- Using *Tagetes* (French marigold) can save around £6200/ha for nematode control compared to methyl bromide fumigation.
- High levels of organic matter can be successfully produced.
- Environmentally sound application with marketing potential for customers.

Background and expected deliverables

There is increasing pressure on all growers who produce field crops to become more focused on the effects of their current production methods on the environment. The use of chemical fumigants is becoming a more expensive procedure and it can also leave the grower open to the risk of pest and disease resistance build-up. Controls for soil-borne pests and diseases are currently based on chemical applications. The continual review and analysis of their environmental effect has led to many being withdrawn.

The presence of both *Verticillium* wilt and nematodes in any numbers can stunt root development and cause weak growth, leading to quality issues. Withdrawal of methyl bromide is set for the end of 2005 and providing alternative options for growers, especially through successful and easily applied cultural control is essential. Previous work on green manures (see Appendix 1) has suggested that these crops can rehabilitate a stressed field environment.

The expected deliverables from this project are:

- An evaluation of a species of Sudan grass appropriate for maximum growth in the UK and its efficacy on known nematode populations and *Verticillium* wilt levels.
- An evaluation of Sudan grass and its suitability to replace soil sterilisation by methyl bromide or methyl isothiocyanate.
- An evaluation of French marigold and its ability to reduce nematode populations.
- An evaluation of both crops to see if weed suppression is successfully achieved.

Summary of the project and main conclusions

1. The number of nematode types and population levels were reduced throughout both green manure crops' lives and for some time after incorporation.
2. Both crops achieved some wilt reduction. The grass in the control plot acted as a non-host plant for nematodes and realised some wilt control too.
3. Sudan grass achieved a good level of *Verticillium* wilt reduction during the growth period and after incorporation. French marigold achieved some wilt suppression but mainly dealt with the free-living nematode populations.
4. Adding a green manure crop into a crop rotation does not significantly increase labour costs or require specialist-growing knowledge.
5. The benefits of using green manures are wide-ranging and considerably important when looking at an environmental approach to growing or using integrated crop management (ICM) on a nursery.
6. Good soil moisture levels and a flail cut to damage the plant tissue will encourage the hydrogen cyanide to be released successfully.

Figure 1 shows the nematode population levels prior to planting, during growth and after incorporation of the green manures compared to the chemical fumigants used. Figure 2 shows the amount of propagules of *Verticillium* wilt found per 200g of soil analysed during 2003/2004.

Figure 1: Nematode populations

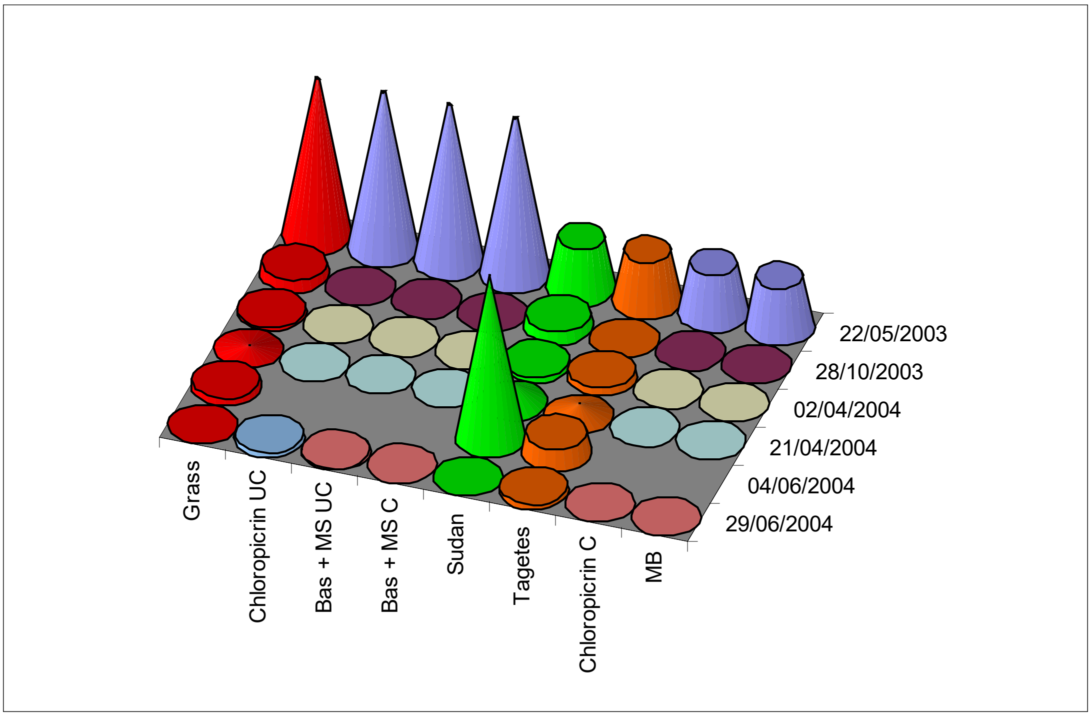


Figure 2: *Verticillium* wilt levels

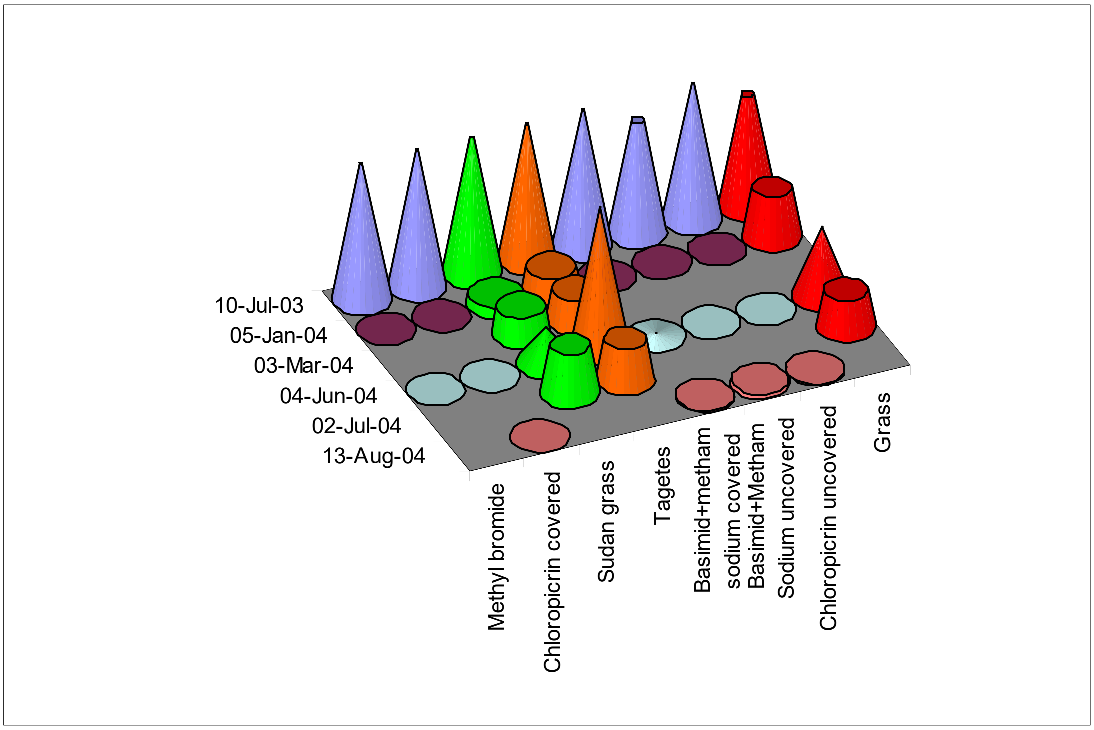


Figure 3: *Tagetes* before flowering

Figure 4: Sudan grass before first cut

Other benefits that have not been fully quantified can be found in the Discussion section (3.3.2).

Financial benefits

- Costs were significantly reduced when comparing the growth of a green crop to the use of chemical soil sterilisation (see Table 1).
- Soil fertility and structural benefits should reduce land work, fertiliser applications and related labour and machinery costs.
- No significant capital expenditure required.

- Cutting the Sudan grass when growth is at a height of 3-4 ft can increase the mass of the root system by up to eight times (McGuire, 2003). Larger root systems can encourage a better soil structure without the need for further mechanical manipulation.
- Competitive advantage can be achieved as far as some customers are concerned. Having an environmental approach to pests and disease control may be to the grower's benefit.

	Sudan grass (£)	***Tagetes* (£)**		**methyl bromide (£)**	**dazomet+metam sodium (£)**	**chloropicrin (£)**
Land preparation	71.75	71.75	Land preparation	71.25	71.25	71.25
Fertilizer	22.50	22.50	Rate	1,000g/ha	400kg/ha	200l/ha
Seed	1225.00	210.00			900l/ha	
Herbicide		13.60	Product	7,500.00	5,750.00	5,000.00
Incorporation	30.75	30.75	Polythene cover	(included)	1,297.00	(included)
12 months land replacement rental	988.00	988.00				
Cost per hectare	**£ 2,338.00**	**£1,336.60**		**£7,571.25**	**£7,118.25**	**£5,071.25**

Table 1: Costs per hectare

Risks

There have been numerous nursery trials using green manures in the UK, especially with *Brassicas* and *Tagetes* but Sudan grass is still quite a novel crop to use to control various soil-borne pests and pathogens in this country. This project has highlighted the need for further work with this green manure to try and maximise its beneficial effect in a UK soil environment, especially when the majority of research has been carried out in much warmer and more stable climates such as Italy and the USA (see Appendix 1).

Environmental conditions are the most important factor when deciding sowing, cutting and incorporation timing, to try and get maximum growth from these crops. The following risks must also be taken onboard.

1. Seed sources

None of these sources have been fully explored with regard to continuous availability, varieties available (and their efficacy/achievable growth rates in the UK) or the quality of seed.

2. Repetition of crop

Green manures may be required for another season on very infested land as the pest/disease levels seem to reduce slowly, over a period of time.

3. Livestock

Growing a crop of Sudan Grass near livestock or where livestock could graze is not recommended because of the risk of prussic acid poisoning if consumed.

4. Soil management techniques

The way a field is managed may affect the ability of any green manure to improve the soil quality and control its pests and diseases. Variation of these techniques was not explored within this project.

5. Re-planting after treatment

Some delay may be required prior to replanting arable crops as seed germination can be affected for up to 10 days after green manure incorporation.

6. Pests and diseases

Using a new crop may introduce new pests and diseases onto the nursery. There may also be some cross-over from related crops which can occur when using oilseed rape and other *Brassicas* for green manuring.

7. Fertiliser application

The amount of fertiliser applied has a direct effect on the amount and quality of organic matter (OM) produced. However, making sure that the crop is cost effective may mean a slight reduction in the amount of OM achieved prior to the incorporation period.

Action points for growers

- You will require 60-45kg per hectare of seed for Sudan Grass.
- You will require 3-4kg per hectare of seed for *Tagetes*.
- You will require a base dressing of 16:0:32 fertiliser at 150kg/ha for both crops.
- You will require a top dressing of ammonium nitrate fertilizer at 150kg/ha, 4 weeks after sowing to both crops.
- Magnesium is only added if the index rate is lower than 2 on both crops.
- The varieties that have been used and have an effect are *Sorghum bicolour 'Sudanense'* and, in the case of French marigold, a combination of *Tagetes patula* and *Tagetes erecta*.
- The combination of two types of marigold gives a broader range of nematode species control through the release of a greater amount of biocidal agents from the roots.
- Make one application of Linuron herbicide after the *Tagetes* have established (approx. 75mm high).
- The Sudan Grass needs to be flail cut to cause tissue damage that allows the hydrogen cyanide to release.
- See Appendix 1 for bibliography on global research carried out using green manures as 'biofumigants'.

Science Section

1.0 Introduction

Using green manures not only as source of organic matter but also as an environmental approach to combating pests and diseases is now considered to be a practical option in many countries worldwide. There are a lot of different types of green manures that seem to have a similarly beneficial effect on removing various soil-borne pests and diseases.

This project looks at Sudan grass (*Sorghum bicolor* 'Sudanense') because of its ability to grow in adverse conditions; the extensive root system that can be produced during its lifetime; its weed suppressive root exudate and the release of dhurrin. This is a harmless glucoside produced by the crop when it is finally incorporated into the soil and breaks down to release hydrogen cyanide (HCN). The sudden disruption of growth such as frost, extreme drought or cutting causes HCN to be released from inside the plant at a more rapid rate. The literature study suggested that younger tissues are thought to have a greater amount of HCN present.

The project also included a mix of *Tagetes patula* and *Tagetes erecta*, which both have a nematicidal activity via the roots. This has been proven through global research over a number of decades. Its effect covers a wide range of nematodes that cause a variety of destructive symptoms and reduce overall crop quality. It does depend on the intra-specific differences in the plant varieties and the types of nematodes present in the soil on their overall effectiveness. They contain and excrete butenylbithiophene (BBT) and acetoxybutenylbithiophene (BBTOAc). These thiophenes are natural broad-spectrum biocides, which act as fungicides, bactericides and nematocides.

1.1 *Verticillium* wilt

There are more than 300 woody and herbaceous species that are affected by *Verticillium* wilt and include various species of *Acer*, *Aesculus*, *Azalea*, *Buxus*, *Fraxinus*, *Magnolia*, *Syringa* and *Viburnum*. Cultural practices and adverse environmental conditions (resulting in high levels of plant stress) can affect the rate of infection of susceptible plants. Crop rotation with non-host plants or planting a cover crop are ways in which infected fields can be used after high levels of infection have been identified (see Appendix 1). Plants with low susceptibility to wilt include species of *Betula*, *Crataegus*, *Cornus*, *Juglans*, *Malus*, *Prunus*, *Populus*, *Pyracantha*, *Quercus*, *Robinia*, *Salix*, *Sorbus* and *Taxus*.

1.2 Nematodes

The nematodes that were identified in the trial field include the following species.

Nematode Name	**Nematode Type**
Pratylenchus penetrans	Root lesion
Trichodorus sp.	Stubby root
Macroposthonia sp.	Ring
Paratrichodorus sp.	Stubby root
Rotylenchus sp.	Spiral
Criconemoides sp.	Ring
Paratylenchus sp.	Pin
Merlinius sp.	Stunt

Table 2: Nematode species identified

Pratylenchus penetrans are commonly known as lesion nematodes because of the necrotic lesions that are left after their entry into the plants' roots. They are endoparasites that are a troublesome pest on a wide range or ornamentals and agricultural crops. They can facilitate diseases that include *Verticillium dahliae* (CSL, 2003). *Paratylenchus* nematodes are commonly known as pin nematodes and although they are of minor economic importance, have a wide host range. *Trichodorus* and *Paratrichodorus* species are commonly known as stubby root nematodes and can affect the root system of crops. They can be vectors of diseases such as Pepper Ringspot, Tobacco Rattle Virus and Pea Early-browning Virus (CSL, 2003). *Macroposthonia* and *Criconemoides* species are commonly known as ring nematodes. Higher numbers can cause symptoms of direct feeding damage (CSL, 2003). Spiral nematodes such as *Rotylenchus robustus* can affect crops such as lettuce, carrots and nursery trees and produce early yellowing in peas (CSL, 2003). *Merlinius* species are stunt nematodes and ca be seen in soils across the UK and their feeding may contribute to the damage done by other pathogens (CSL, 2004).

Foliage symptoms include wilting with quality becoming progressively poor and lacking sheen. New foliage is stunted and weak with fewer leaves being produced. Prolonged root stress can lead to yellowing of the plant and during dry conditions; the plants react by wilting quickly compared to uninfected plants in the same situation. Root symptoms are varied depending on the type of nematode. Symptoms include galls, stunting, root decay and deformation. Other problems such as viruses and fungal infections take hold much quickly because of previous nematode damage.

Results with summer cover crops, which include Sudan grass, have been positive. Some varieties have a 'trap crop' effect on nematodes (Winslow, 1955) and others have also identified that compounds are secreted through the Sudan grass root systems to remove unwanted predators (Luna, 1993, MacGuidwin and Layne, 1995) and weeds (Einhellig and Souza, 1992, Einhellig *et al*., 1993, Einhellig and Rasmussen, 1989). Some inhibition of weed growth in the following season has been noted (Putnam *et al*., 1983).

According to some recent German research carried out in 2004, Sudan grass can control nematodes during its growing phase too (pers. comm., H Lösing).

The removal of root knot nematodes with *Tagetes* sp. has been researched and recorded for many years in several different environments on various crops (Abawi and Vogel, 1999, Abivardi, 1971, Bruce-Reynolds *et al*., 2000, de Wael *et al*., 2003, Nirula and Bassi, 1965, Motsinger *et al*., 1977, Phloeg, 2002, Reynolds *et al*., 2000, Siddiqui and Alam, 1987/1988/1989, Supratoyo, 1993, Tompsett, 2003, Topp *et al*., 1998 and Wang *et al*., 2003).

The amount of research that has been carried out worldwide on the benefits of various crops is vast and now seems to be increasing because of the pressure to reduce chemical inputs in food and ornamental production.

2.0 Materials and methods

The site chosen was a field section (consisting of just over 3 hectares) at Hillier's Andlers Ash broadleaved tree nursery in Liss, Hampshire. The soil type was sandy loam and the trial layout can be seen in Figure 5.

The initial sampling was over the whole area and all subsequent sampling was on each individual treatment plot. In the process of crop clearing, the land was considerably disturbed and left with large holes some several meters wide and up to one meter deep. The initial chisel ploughing of the area was carried out to attempt levelling of the soil. Tree remnants that remained were gathered into large heaps from the whole area and burned in a series of fires. There was considerable soil disturbance and large-scale movement of tree debris over the whole trial area.

Soil samples were taken after the previous crop removal, to identify the levels of free-living nematode populations and the number and viability of any *Verticillium* wilt propagules. The protocol adopted was issued by EMT based on a maximum sampling area of 2 hectares and samples taken on a 10-metre by 20-metre grid. Samples were kept in cool conditions during storage and posted to the laboratory during weekdays to avoid excessive postal delays caused by weekends. At the time of initial sampling no trial boundaries had been marked out. Trial boundaries were marked out after final soil preparations. All the plots were ploughed up in May 2004 as part of a normal crop preparation procedure. It was requested that the machinery was cleaned free from all soil particles before tillage work commenced and that the methyl bromide plots were worked first to avoid cross contamination. No confirmation has been possible that this was carried out.

Date sampled	Area name	Area Ha	No. of wilt propagules/200g soil	No. of free-living nematodes
15 May 2003	Upper field	1.77	11.5	117
15 May 2003	Lower field	1.51	9.5	38

Table 3: *Verticillium* wilt and nematode analyses

2.1 Treatments

2.1.1 Chemical fumigants

All fumigants were applied during September 2003.

Product Name	Active Ingredient	Application rate	Application time
Sobrom BM100	methyl bromide	144g/cu m	Sept 2003
Discovery+Basamid	metam-sodium+dazomet	1.8l/20m^2+220kg/ha	Sept 2003
K&S Chlorofume	Chloropicrin	400l/ha	Sept 2003

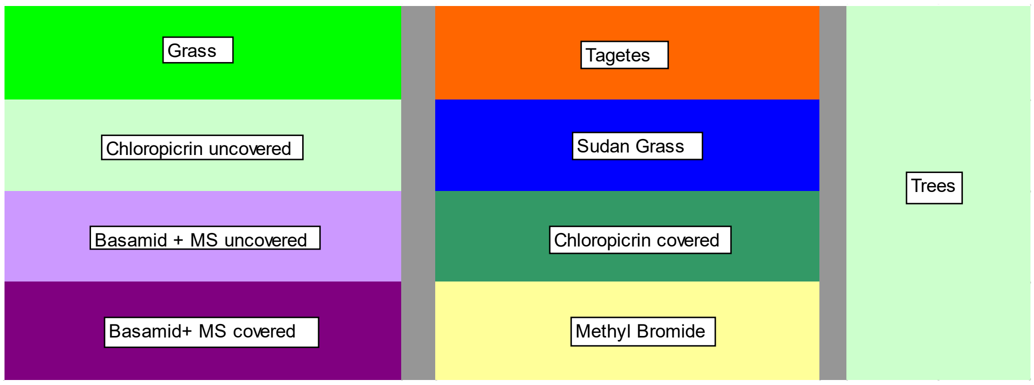
Table 4: Breakdown of application rates and timings.

2.1.2 Green manures

Sudan grass (*Sorghum bicolour* var. *Sudanense*)
French marigold (a combination of *Tagetes erecta* + *Tagetes patula*)

The plots were set into an area measuring just over 3 hectares (32,880m^2). Assessments were carried out at at various intervals to monitor the levels of *Verticillium* wilt (*V. dahliae*) in colony forming units (cfus) and numbers of free-living nematodes per 200g of soil. The weather in 2003 was particularly conducive to the growth of the Sudan grass. Its height reached about 1 metre prior to the first cut in July on part of the area with the remaining plot reaching 2 metres before incorporation. The final chop and integration occurred in September. Analyses were achieved when the field conditions allowed (see Figure 6).

Figure 5: Trial layout



Plot identification	Area	Total Area	pH
methyl bromide	126m x 30m	3,750m^2	5.9
chloropicrin (covered)	126m x 15m	1,890m^2	5.9
chloropicrin (uncovered)	148m x 15m	2,220m^2	5.5
dazomet+metam-sodium (covered)	148m x 20m	2,960m^2	5.5
dazomet+metam-sodium (uncovered)	148m x 20m	2,960m^2	5.5
Tagetes	126m x 32m	4,332m^2	5.9
Sudan grass	126m x 32m	4,332m^2	5.9
Grass control	148m x 65m	9,620m^2	5.5
Total Area		**32,064m^2**	

Table 5: Plot areas

2.1.3 Sampling techniques

(See also introduction to 2.0)

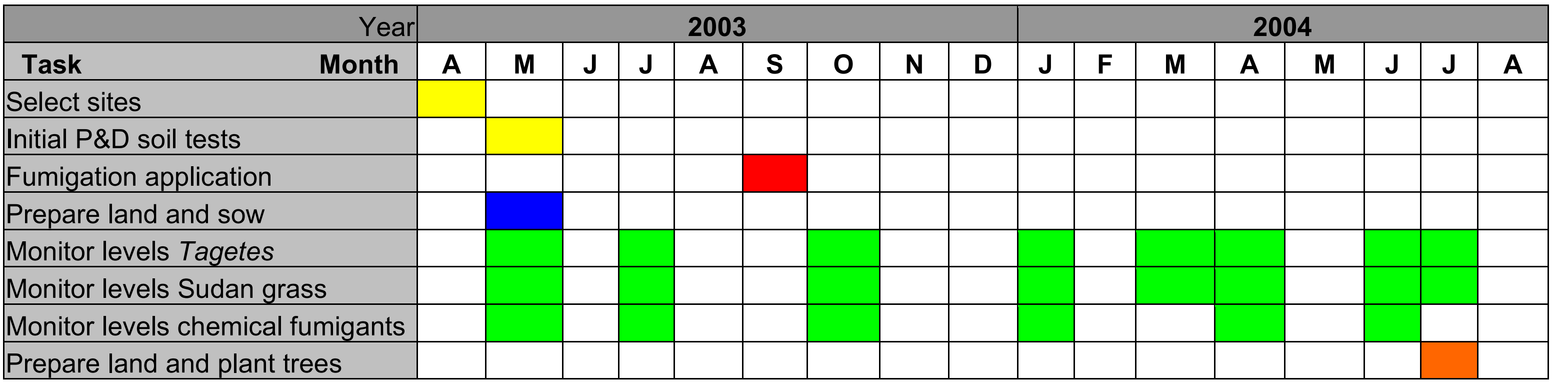
The sampling regime was reduced during the wet and continuously poor soil conditions on the nursery at the end of 2003 and the beginning of 2004. However, the results that were collected give a good indication that there was a reduction in the amount of wilt infection and nematode populations in all non-chemical plots.

There were fewer samples taken of the chemical fumigant plots because of the constantly negative results that were still being achieved some months after treatment had occurred.

In the larger plots (Sudan grass, Tagetes, methyl bromide, grass control and dazomet+metam sodium covered), approximately 100 cores were taken from throughout the area to ensure that a good sample was taken each time. The depth of the core was 150mm. The smaller plots (chloropicrin covered and uncovered, dazomet+metam sodium uncovered) had approximately 60 cores taken from them.

Throughout the trial, the chemically treated plots were always sampled first to prevent cross contamination from the coring tools.

Figure 6: Breakdown of analyses taken in 2003/2004.



3.0 Results

3.1 *Verticillium* wilt test results

Viable propagules per 200g soil	Verticillium wilt sampling dates					
	10/07/2003	05/01/2004	03/03/2004	04/06/2004	02/07/2004	13/08/2004
methyl bromide	11.5	0				
chloropicrin covered	11.5	0				0
chloropicrin uncovered	11.5	0				0.1
dazomet+metam-sodium covered	11.5	0				0.1
dazomet+metam-sodium uncovered	9.5	0				0.3
Tagetes	11.5	2	2.6	12	3.1	further
Sudan grass	11.5	0.8	2.3	3	4.1	results
Grass control	9.5	4.1		5.8	3.5	to come

Table 6: *Verticillium* wilt test results

All the treatments achieved a reduction in the levels of *Verticillium* over the period of sampling. All of the chemically treated areas had no cfus present after treatment, which remained basically at that level throughout the trial. The cfus levels of the *Tagetes* showed a reduction after incorporation, as did the control grass area. As time progressed, the grass and *Tagetes* had higher wilt levels than the Sudan grass. It was not expected that the *Tagetes* area would reduce much and it appears that the levels do rise after incorporation. *Tagetes* have no direct effect on *Verticillium* wilt levels, only on the reduction in nematode populations that can act as a vector of the disease. The grass area had no treatment at all and also showed a reduction in wilt levels.

The variation in the results could be due to several factors including:

1. Contamination during ploughing in May 2004.
2. The dry soil conditions during some sampling times.
3. The lack of food for the nematodes after incorporation and breakdown.
4. The unforeseen ability of grass to act as a successful non-host plant.

The samples may have been affected by the ploughing that occurred in May 2004 (see graph spikes in Figures 1 and 2). The whole area was ploughed with no reference to the original plots, even though this was requested. No information was available as to which area the ploughing was started. This may go some way to explaining the variation in results from the ploughing period onwards, especially the numbers of propagules in the grass control compared to the Sudan grass plot. The grass control's ability to act as a non-host must be acknowledged as a potential method of reducing nematode populations. However, it will not improve soil structure, significantly increase organic matter levels and encourage beneficial predators (*Tagetes*) like a green manure.

3.2 Nematode test results

The total figures presented suggest a general decline in the number of nematodes present in each of the green manure crop areas.

The variation in the figures received from this date onwards may not necessarily provide a clear set of data because of the ploughing regime adopted (see comments in 3.1). This probably brought some nematodes to the soil surface and exposed them to adverse conditions, therefore reducing the populations.

A significant amount of microbivorous nematodes (>2000 per 200g of soil) were present after the *Tagetes* were incorporated (tested in January 2004). This appears to be due to the high levels of organic matter present in the soil. No reference could be found in the literature search as to the lack of effect or otherwise by *Tagetes* on this nematode group. Competition for food would have been significant and may have contributed to a decline in the number of parasitic nematodes. The microbivorous nematodes feed on micro-organisms and bacteria in the soil as well as organic matter and may contribute to a reduction in other soil-borne problems. They can play a significant role in the decomposition of soil organic matter, mineralization of plant nutrients and nutrient cycling (Ferris, 1998, Ingham *et al*., 1985, Hunt *et al*., 1987 and Griffiths, 1990). The lack of these nematode types in the Sudan grass may have been due to the breakdown of dhurrin to hydrogen cyanide, therefore probably removing the majority of both beneficial and parasitic soil micro-organisms.

The overall picture shows a significant amount of reduction during the growth of both the French marigold and Sudan grass.

Nematode types seen per 200g soil	Nematode populations sampling dates						
	2003		**2004**				
	22 May	28 Oct	26 Jan	2 Apr	21 Apr	4 Jun	29 Jun
methyl bromide	38	0		0	0		0
chloropicrin covered	38	0		0	0		0
chloropicrin uncovered	117	0		0	0		3
dazomet+metam-sodium covered	117	0		0	0		0
dazomet+metam-sodium uncovered	117	0		0	0		1
Tagetes	38	0	1	5	8	15	3
Sudan grass	38	9	17	3	13	122	0
Grass control	117	8		2	2	5	0

Table 7: Nematode population data

3.3 Weed control

Weed suppression and control was achieved with a single application of Linuron to the *Tagetes* and none to the Sudan grass. The Sudan grass was able to grow, even during drought periods, when most of our native weed species' growth became severely affected. The plant releases a compound called sorgoleone, which is exuded through the living root hairs and is an effective weed suppressant. Some work has shown that low concentrations of this compound can still have significant effects. The amount produced can depend on environmental factors as well as the genetic differences between each Sorghum variety (Nimbal *et al*., 1996). The thick canopy layer in both green manure crops contributes to the smothering of germinating weed seedlings.

4.0 Discussion

There are many areas that need to be assessed by the grower prior to planting a green manure crop for the first time. These include the weather, soil type, field conditions, the characteristics of the crop to be planted and the level and type of infection present. These all contribute to whether the green manure crop can achieve a good enough effect in the same year of planting and incorporation. Further studies, looking at varieties and cultural techniques are required to establish how the maximum effect can be achieved.

A heavy infection of either nematodes or wilt may require more than one sowing of a green manure crop to combat the problem. The consequences of insufficient control for a five-year tree crop can be significant. Section 4.2 lists the further benefits of using green manures.

It must be noted that comparing the holistic and chemical approach to pest and disease management is difficult because each method has a different way of controlling the pest/disease. Green manuring is a fairly new way of managing soil-borne problems in the UK whereas chemical fumigation is known to work because of the large amount of quantified research behind it. Where nothing survives in a chemically treated soil, with some green manures e.g. *Tagetes*, there can be a greater competitive environment created through OM breakdown.

Extensive research on green manures and their benefits within the soil environment (see Appendix 1) has been confirmed and demonstrated by many other countries. Using them ultimately contributes towards a more friendly approach to farming. However, further work based in the UK is now required to identify the relationship of our environment and the efficacy of the crops (section 4.3).

4.1 Issues with sampling

Maintaining a stable field situation is not always feasible on a working nursery and some samples were taken when parts of the trial area had been ploughed or re-planted. The soil conditions may have contributed to the reduction of nematodes because of the lack of soil moisture for long periods during the beginning of 2004. These dry and bright conditions were still present after ploughing had occurred.

The number of nematodes found in the uncovered chemical fumigation plots was seen to start slightly increasing in the summer of 2004. The efficacy of the fumigants may have been further affected with just a smear roller being used after application. Success of this roll does depend on the weather and 2003 was conducive to maintaining the hardened layer of soil on top of the chemicals applied.

Green manure crops were re-sown in late June 2004 in exactly the same plot areas by the nursery to try and see if even greater reductions could be achieved. The subsequent reduction seen in the levels of wilt and nematodes in late June 2004 probably indicates that the manures may now be having some effect on the nematodes as they grow and develop. This initial establishment (75mm tall) may have been enough for the plants to start excreting their control compounds into the soil environment (pers. comm., H. Lösing).

4.2 Further benefits

Additional benefits that have been identified through global research by the use of green manures crops in place of chemical methods include:

- Higher organic matter re-introduced back into the soil environment
- Energy source is provided for beneficial micro-organisms that go on to out-compete pathogens for a place on the rhizosphere surface. This exclusion ultimately prevents infection.
- Root penetration into the soil environment and the potential in reducing soil works such as ploughing.
- Improvements in water-holding capacity, nutrient-holding capacity, cation exchange capacity, crop productivity, aggregation and infiltration, water and air quality and wind erosion reduction.
- Dense planting and rapid growth can out-compete weed population, thus reducing the need for herbicides.
- Reduction in general chemical applications can lead to further arable benefits such as lower input growing.
- Resistance strategies can be reinforced when using systems that do not rely on chemical inputs.
- Changes in the species of green manures may be able to accomplish a reduction in other soil-borne problems.
- No harvest intervals.
- Encouragement of beneficial predators (particularly *Tagetes*) into the field to attack pests.
- The use of *Tagetes* as an intercrop is successful because of the limited height the plant gets to and the abundance of flowers produced.

4.3 Future work

1. Plant profiling of each green manure to identify glucosinolate levels. No work could be found that measures the levels of glucosinolate produced by the many varieties of Sudan grass available in the world. Limited varieties appear to grow successfully in the UK but determination of the soil sterilizing effect is needed for each of them.
2. Continued analysis of Sudan crop during growth cycle to identify optimal glucosinolate and therefore the best time to cut and incorporate. Work suggests that young soft growth produces a greater level of glucosinolate.
3. Effect of irrigation on matching release of isothiocyanates throughout the test area. Rainfall levels may also affect the retention of the product in the soil. The leaching potential requires determination.
4. Effect of various methods of damaging tissue to start the release. Work shows that frost can cause release, but no work has been done on flailing, rolling or mowing as being the most suitable method to achieve maximum release.
5. Work on the effect of Sudan grass on Apple Replant Disease should be investigated.
6. Analysis of the nutrient levels of soils prior, during and after green manure incorporation to identify any potential fertiliser savings.
7. Use of trap crops compared with biofumigation crops.
8. Methods of seeding affecting growth. Drilling or broadcasting could have an effect on the seed rate. Work is needed to establish which method is better and the plant population per square metre actually required.

9. Timing of the intermediate cut to encourage root penetration in particularly compacted soils. Root penetration of *Tagetes* and Sudan grass improves the control potential.
10. Amount of biomass produced by each crop, during a particular weather season.
11. Crossover of pests from locally grown crops from the same family.
12. Using other different types of green manures e.g. *Brassica* types to control nematode populations.
13. Identification of the best green manure for different UK environmental conditions to gain maximum effect.
14. Recording the levels of beneficial organisms removed from the soil during fumigation compared to green manure crops.
15. The types and levels of beneficial organisms that Sudan grass can remove during its growth and after incorporation.
16. The appropriateness of using *Brassica* as a potential follow-on crop from Sudan grass.
17. Confirmation that there is negligible impact on carbon dioxide atmospheric levels compared to chemical fumigants.

5.0 Conclusions

1. Wilt and nematode reduction
The use of *Tagetes* can reduce the numbers of free-living nematodes in a soil environment and it also appears to have a limited effect on reducing *Verticillium* wilt levels either directly or indirectly. Sudan grass can effectively reduce the levels of *Verticillium* wilt propagules in a soil environment. It also has some ability to reduce the free-living nematode populations. Both green manures produced good control during the life of each crop and for a period after incorporation. The grass ley may have acted as a non-host plant for nematodes, reducing the levels and encouraged wilt reduction although not at levels seen in the Sudan grass.

2. Crop rotation
Green manures can be successfully incorporated into a crop rotation programme in nursery stock. There is no increase in the requirement for labour, contractors or specialist-growing knowledge.

3. Benefits
This type of soil 'fumigation' has the potential for application in many other related horticultural and agricultural field systems. Biodegradation of the material is guaranteed after incorporation and eco-toxicity is also avoided. Other benefits include increased soil fertility, renewed structure from root growth and an increase in organic matter levels. Green manures are also important when looking at an environmental approach to growing or using ICM on a farm or nursery. There is potential for competitive advantage in using alternative treatments for soil-borne pests and diseases.

4. Crop work requirements
Tagetes requires no other work after sowing until it is cut and incorporated. The Sudan grass efficacy appears to be enhanced from a cut to increase the root mass, root penetration and maintenance of young soft growth. No further cultural operations are necessary until the time of final incorporation. If soil conditions allow, another cut would benefit the soil structure during the crop's growth. Good soil moisture and a flail cut to damage the plant tissue will encourage the Sudan grass to produce and release the hydrogen cyanide successfully. When the crops are seeded and trying to establish, it is important that the *Tagetes* receives adequate moisture. Sudan grass is well known for its ability to grow in poor conditions but initial soil moisture will produce a quality crop.

5. Variation in results
Ploughing of the total trial area may have contributed to the varied results after May 2004. Wilt levels and nematodes may have been brought to the surface from the bottom of the tilled depth where they were not controlled. Further variation in the results may have been due to the low and high soil moisture levels experienced during the trial period. Nematodes need a film of water to 'swim' through the soil and without this would have had a limited ability to move and may have perished.

6. Weed suppression
Weed suppression during crop growth can be successfully achieved because of the large canopy layer produced by both crops. The root exudates from Sudan grass and *Tagetes* also contributed towards the control. There is anecdotal evidence to suggest that flail mowing Sudan grass mid season

reduces surface weed growth at that time, through both the biofumigation effect and smothering.

7. Choice of green manure varieties

The varieties chosen for this trial were identified through the desk study of current global research. However, the UK climate is quite different to many of the countries currently using green manures and this may have affected their efficacy. Current green manure effects are lower than chemical fumigants but further research is providing more refined and increasingly powerful varieties that will improve efficacy.

8. Polythene use for chemical fumigation

The efficacy of the chemicals used without polythene covers was unaffected during the first few months after application. However, the numbers of nematodes were starting to rise in June 2004. The polythene covers were left on after treatment and it was intended to remove them prior to planting. The strong winds during the winter period of 2004 destroyed most of the sheeting and it was removed. There appears to be no appreciable differences between the initial sheeting and surface smearing of the metham sodium/dazomet treatments. The advantages of not having to handle the removal and disposal of the sheeting was a positive advantage.

Glossary

EMT
East Malling Trust (formally East Malling Research Station)

Non-host plant
A plant that provides an environment where a pest or pathogen is unable to reproduce or feed successfully.

Organic matter
The fraction of soil that includes plant and animal residues at various stages of decomposition, cells and tissues of soil organisms, and substances synthesised by the soil population (NWCG, 2001).

ppm
A measurement which accounts for parts per million.

cfus
A measurement (in this case) used to determine the number of propagules of Verticillium wilt in soil as colony forming units.

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Appendix 1

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1.1 Sudan grass

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Appendix 2

2.1 *Verticillium* wilt data

Verticillium wilt (cfus)				
Area code	**m^2**	**Date sampled**	**Date analysis returned**	**Result**
Top	17,760	15/05/03	10/07/03	11.5
Lower	15,120	15/05/03	10/07/03	9.5
methyl bromide	3,750	14/10/03	05/01/04	0
		19/03/04	04/06/04	0
dazomet+metam-sodium covered	2,960	14/10/03	05/01/04	0
		19/03/04	04/06/04	0.2
		22/06/04	13/08/04	0.1
dazomet+ metam-sodium uncovered	2,960	14/10/03	05/01/04	0
		19/03/04	04/06/04	0
		22/06/04	13/08/04	0.3
chloropicrin uncovered	2,220	14/10/03	05/01/04	0
		19/03/04	04/06/04	0
		22/06/04	13/08/04	0.1
chloropicrin covered	1,890	14/10/03	05/01/04	0
		19/03/04	04/06/04	0
		22/06/04	13/08/04	0
Sudan grass	4,032	14/10/03	05/01/04	0.8
Uncut		15/01/04	27/02/04	2.1
Cut		15/01/04	27/02/04	2.5
		19/03/04	04/06/04	3
		21/05/04	02/07/04	4.1
Tagetes	4,032	14/10/03	05/01/04	2
		15/01/04	27/02/04	2.6
		19/03/04	04/06/04	12
		21/05/04	02/07/04	3.5
Grass	9,620	14/10/03	05/01/04	4.1
		19/03/04	04/06/04	5.8
		21/05/04	02/07/04	3.1

2.2 Nematode data

		Sample Dates					
Grass control	**Type**	22/05/2003	28/10/2003	02/04/2004	26/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion	5	1	2		2	0
Trichodorus sp.	Stubby root	15					0
Macroposthonia sp.	Ring	133					0
Paratrichodorus sp.	Stubby root	1					0
Rotylenchus sp.	Spiral	1	1		1		0
Criconemoides sp.	Ring				1	3	0
Paratylenchus sp.	Pin		5				0
Merlinius sp.	Stunt		1				0
Sudan grass	**Type**	22/05/2003	28/10/2003	02/04/2004	26/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		9	2	5	91	0
Trichodorus sp.	Stubby root					28	0
Macroposthonia sp.	Ring						0
Paratrichodorus sp.	Stubby root						0
Rotylenchus sp.	Spiral			1			0
Criconemoides sp.	Ring					3	0
Paratylenchus sp.	Pin				8		0
Merlinius sp.	Stunt						0
methyl bromide	**Type**	22/05/2003	28/10/2003	02/04/2004	26/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0	0	0		0
Trichodorus sp.	Stubby root		0	0	0		0
Macroposthonia sp.	Ring		0	0	0		0
Paratrichodorus sp.	Stubby root		0	0	0		0
Rotylenchus sp.	Spiral		0	0	0		0
Criconemoides sp.	Ring		0	0	0		0
Paratylenchus sp.	Pin		0	0	0		0
Merlinius sp.	Stunt		0	0	0		0
dazomet+metam-sodium C	**Type**	22/05/2003	28/10/2003	02/04/2004	26/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0	0	0		0
Trichodorus sp.	Stubby root		0	0	0		0
Macroposthonia sp.	Ring		0	0	0		0
Paratrichodorus sp.	Stubby root		0	0	0		0
Rotylenchus sp.	Spiral		0	0	0		0
Criconemoides sp.	Ring		0	0	0		0
Paratylenchus sp.	Pin		0	0	0		0
Merlinius sp.	Stunt		0	0	0		0
dazomet+metam-sodium UC	**Type**	22/05/2003	28/10/2003	02/04/2004	21/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0	0	0		0
Trichodorus sp.	Stubby root		0	0	0		0
Macroposthonia sp.	Ring		0	0	0		0
Paratrichodorus sp.	Stubby root		0	0	0		0
Rotylenchus sp.	Spiral		0	0	0		0
Criconemoides sp.	Ring		0	0	0		1
Paratylenchus sp.	Pin		0	0	0		0
Merlinius sp.	Stunt		0	0	0		0

Tagetes	Type	22/05/2003	28/10/2003	02/04/2004	21/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0		7		0
Trichodorus sp.	Stubby root	5	0	4		7	1
Macroposthonia sp.	Ring		0				0
Paratrichodorus sp.	Stubby root		0				0
Rotylenchus sp.	Spiral		0	1			1
Criconemoides sp.	Ring		0			8	0
Pratylenchus flakkensis	Root lesion		0		1		0
Paratylenchus sp.	Pin		0				1
Merlinius sp.	Stunt		0				0
chloropicrin uncovered	**Type**	22/05/2003	28/10/2003	02/04/2004	21/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0	0	0		3
Trichodorus sp.	Stubby root		0	0	0		0
Macroposthonia sp.	Ring		0	0	0		0
Paratrichodorus sp.	Stubby root		0	0	0		0
Rotylenchus sp.	Spiral		0	0	0		0
Criconemoides sp.	Ring		0	0	0		0
Paratylenchus sp.	Pin		0	0	0		0
Merlinius sp.	Stunt		0	0	0		0
chloropicrin covered	**Type**	22/05/2003	28/10/2003	02/04/2004	21/04/2004	04/06/2004	29/06/2004
Pratylenchus penetrans	Root lesion		0	0	0		0
Trichodorus sp.	Stubby root		0	0	0		0
Macroposthonia sp.	Ring		0	0	0		0
Paratrichodorus sp.	Stubby root		0	0	0		0
Rotylenchus sp.	Spiral		0	0	0		0
Criconemoides sp.	Ring		0	0	0		0
Paratylenchus sp.	Pin		0	0	0		0
Merlinius sp.	Stunt		0	0	0		0

Appendix 3

HNS 119 Seed sources

1. W.A. Church (Bures) Ltd., Bures, Suffolk. CO8 5JQ. Tel: (01787) 227654. Fax: (01787) 228325. Email: info@churchofbures.co.uk. Website: www.churchofbures.co.uk

Contact: Geoff Lakin

2. Rudy Raes Bloemzaden, Haenhoutstraat 204, 9070 Destelbergen. Tel: 0032 9 355 58 30. Fax: 0032 9 355 66 99. Email: info@raes.be Website: www.raes.be

Contacts:

Simon Crawford (UK)
Tel: (01608) 684548

Other potential seed sources

1. Plant Solutions Ltd., Pyports, Cobham, Surrey. KT11 3EH. Tel: (01932) 576699. Fax: (01932) 868973. Email: sales@plantsolutionsltd.com. Website: www.plantsolutionsltd.com