SESSION 6A AGROCHEMICAL OPTIMISATION: LAND MANAGEMENT OPTIONS

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PESTICIDE MANAGEMENT IN UK WHEAT PRODUCTION - OPTIMISATION AND PRAGMATISM

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ABSTRACT

The adoption of effective pesticides over the last thirty years or so has, along with other advances in technology, resulted in UK wheat production becoming more competitive internationally. However, the consequent changes to arable farming have led to concerns about the environmental value of the countryside. This, along with the prospect of more exposure of UK arable farmers to international markets, suggests that additional improvements in competitiveness are required through adopting approaches to crop management which also enhance the environmental value of the countryside. It is only the further adoption of existing and new technologies which will achieve these two aims. The factors which influence the adoption of pesticide optimisation in practice are identified and future prospects discussed.

INTRODUCTION

The demand for wheat is rising faster than the growth in world population. This is due to increasing industrialisation, particularly in China, South East Asia and South America. The consequent exodus from the countryside to towns and the increase in real incomes is resulting in diets which are based on what the consumer wants rather than on what a subsistence or 'balanced' agriculture can produce. Such major structural changes were already occurring in the UK in the mid-19th century, at a time when the area devoted to wheat was hampered by the need for rotations. The additional demand for wheat, along with the introduction of steam engines, resulted in the opening up of the prairies in North America and imports of cheap grain. Farmers in the UK could not compete and there followed a long agricultural depression.

However, the unit cost of production of wheat in the UK is now comparable with some of the major production areas in the USA and Canada, where yields are limited by drought. In comparison, the last thirty years have seen a tremendous increase in wheat yields in the UK (Figure 1). This has been due largely to improvements in soil management, plant breeding, nutrition and pesticides, enabling a fuller exploitation of a climate and soils which can sustain high yields. These increases in yield, coupled with increased mechanisation, have resulted in the greater competitiveness of not only UK but of Northern European wheat production.

Effective herbicides and chemical fertilisers have largely removed the need for rotations, hence avoiding crops which are only grown to 'weed and feed' crops such as wheat. This has resulted in the concentration of crops onto land which is most suited to their production. Continuous autumnsown wheat is now possible on heavy soils in the UK but complete reliance on herbicides has led to herbicide resistance in annual grass weeds, particularly black-grass (*Alopecurus myosuroides*). Hence, some cultural control measures are still essential to achieve sustainable production.

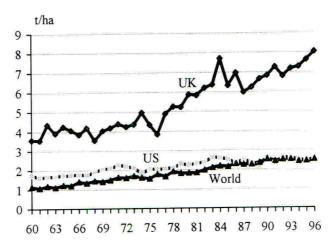


Figure 1. Average yields of wheat (t/ha) in the UK, US and World from 1960 to 1996. Source - USDA (1997)

However, the concentration of crops onto the land most suited to their production has led to habitat destruction and fragmentation. In addition, there are concerns expressed over biodiversity of arable land, pesticide use in general and their presence in water in particular. This has led to the current calls for a return to more 'balanced' farming systems with reductions in the reliance on new technologies, including the use of pesticides. However, at the same time the EU is projecting a further increase in the exposure of its farmers to world markets as a result of the next round of talks organised by the World Trade Organisation.

Therefore, it is important that UK wheat production maintains or increases its competitiveness in world markets, whilst at the same time trying to minimise the impact of its production on the environment. The return to more 'balanced' rotations is not an option unless tax-payers are willing to increase their support of agriculture and/or consumers are willing to adapt their diets to eat what is produced by such rotations. The only way that this dilemma can be resolved is through the further adoption of new and existing technologies, which will result in a reduction of the environmental impact of arable production and an increase in economic optimum yields. In this context, the aim must be to minimise the overall environmental impact of arable production, not to minimise the usage of pesticides or other inputs *per se*.

WHAT IS PESTICIDE OPTIMISATION?

The use of pesticides is said to be optimised when the financial margin from their use is maximised, either in the current crop or over the whole cropping cycle. This is the context within which optimisation is discussed in this paper. However, it has to be recognised that this is rather a simplistic view because pesticides are one of many management options that can be taken by a farmer, all of which have an environmental as well as a financial impact on crop production.

Increasingly, optimisation has been taking on an environmental perspective. Pesticide legislation in the UK has resulted in compliance with approved product labels which have the aim of protecting human beings, creatures and plants, safeguarding the environment and ensuring safe and effective and humane methods of controlling pests. In addition, arable farmers are responding to the concerns of consumers by trying, where possible, to maintain biodiversity on cropped margins and non-cropped land.

Therefore, in the future, optimisation needs to aim for acceptable financial margins whilst minimising the impact of crop production, including pesticides, on the environment. Hence, pesticide optimisation should ideally be defined as the management of pesticides within arable systems whose environmental impact is minimised whilst achieving acceptable financial margins. To help ensure sustainability, it is essential to encourage biodiversity, particularly on non-cropped land and cropped field margins, and also to avoid pesticide resistance and to maintain soil health.

This definition inevitably results in complexity. Minimising the use of one type of input may increase or decrease the requirement for another type of input. For instance, it can be postulated that managing the crop canopy of wheat to optimise the interception of radiation will result in a reduction in the use of nitrogen and perhaps fungicide but may increase the use of herbicides, due to less crop competition with weeds. There are also complex relationships between crop management decisions and pesticide use. Sowing winter wheat early in the autumn will reduce nitrate leaching over the winter but increase the use of insecticides, to control the aphid vectors of cereal viruses, and of herbicides to control the resulting higher populations of weeds. Hence, it is clear that farmers will not only need to have clear environmental as well as financial objectives but also realise that optimising pesticide use should ideally be carried out in the context of the whole farm system rather than as a series of individual inputs to individual crops.

Achieving the aim of minimising the impact on the environment of systems which produce acceptable financial margins will not only involve complex decision making but also, it will be a dynamic process. New technological developments, such as biotechnology (Salamino & Motto, 1993) and new advances in existing technologies, will be introduced as and when they offer robust advantages. This places huge demands on the transfer of technology from research into practice.

There is enthusiasm amongst many farmers and advisers to receive more information on the environmental impact of individual pesticides. Great care will have to be taken as to how such information is presented because pesticides vary in their impact on different aspects of the environment. While setting specific environmental targets for an individual farm, in itself a complex exercise, may help to clarify pesticide selection, it is also essential that decision makers have sufficient knowledge to ensure that such information is used correctly. Pesticide labels now contain more information on environmental impact and this particular approach may be preferable to a more complete disclosure of information.

PROGRESS TOWARDS OPTIMISING PESTICIDE USAGE

The success of previous efforts to transfer into practice the optimum economic use of pesticides is difficult to measure because insufficient recording hampers analysis. Some simple concepts can be rapidly transferred because they can easily be assessed in the field. An example of this is the need to apply herbicides in the autumn for the control of annual grass weeds in winter wheat. Cereal

growers soon recognised from their own experience that this was the correct approach to take. It is those approaches, whose success is not easily assessed in the field, which are more difficult to transfer into practice. An example of this is disease control in winter wheat, which has been dominated by the triazole group of fungicides over the last 10-15 years. By the mid-1980s, it had been clearly demonstrated that the optimum time for a single application of these fungicides to reduce yield loss is when the flag leaf of the wheat is emerging or fully emerged (GS 39 - see Table 1). Lower yield losses to disease occur from single applications at the other main timings, when the crop has one-two nodes detectable (GS 31/32) or when the ear is fully emerged (GS 59). Even in sequences, an application to the emerging or fully emerged flag leaf is the key timing. However, despite the best efforts of many researchers and advisers, it took until the mid-1990s for farmers to start to, if not fully, recognise this approach (Table 1).

Table 1. Time of spraying of fungicides in winter wheat according to growth stage (GS; Tottman, 1987) - % crops treated in England and Wales.

Source: CSL/ADAS Cereal Disease Surveys (described in Polley & Thomas, 1991)

	No. of crops surveyed	% crops treated with fungicide			
) -	GS 29-35	GS 36-45	GS 49-71	Unsprayed
1986	373	73.2	41.8	50.4	7.8
1991	367	71.9	32.7	70.8	6.8
1992	380	74.7	36.3	64.5	5.3
1993	369	75.0	39.0	69.1	4.3
1994	392	55.6	64.6	49.7	4.8
1995	376	75.8	67.1	47.9	4.5
1996	368	77.3	69.5	43.7	2.2

The explanation for this slow adoption of a simple message from research is fairly clear. Yield losses are minimised by protecting the three youngest leaves against disease. The major yield threatening disease in the UK is due to the fungus Septoria tritici. Applications of a triazole fungicide at full flag leaf emergence protects the youngest leaf from infection from this fungus and controls the infection in the second youngest leaf. However, at the flag leaf stage, the symptoms of this disease are frequently not visible on these leaves and other diseases are often at a low level due to high and perhaps unnecessary fungicide usage at earlier growth stages. This results in farmers tending to delay treatment until a time when application provides a reduced level of control.

Another well proven approach to the more economic use of fungicides in wheat has not yet been generally adopted. There is overriding scientific evidence that the optimum requirement for fungicides varies considerably between cultivars and reflects their genetic resistance. Despite this, surveys show little differential in fungicide use between cultivars (Stevens *et al.*, 1997).

Overall, it is difficult to assess whether or not there is a trend towards farmers adopting optimisation techniques. Surveys of pesticide use can produce misleading information. Reductions in the weight of active ingredient used can be due to the adoption of new pesticides which have lower recommended doses. The number of products used can be misleading due to the widespread adoption of tank-mixtures and often, the more specific nature (i.e. narrower range of target species) of some of the recently introduced pesticides. The adoption of doses more appropriate to crop, site and season may increase the number of spray passes. Recent surveys in winter wheat suggest that whilst the rate of active ingredient applied to wheat continues to fall, the number of products used and the number of spray passes continue to rise.

The University of Cambridge has provided some indirect evidence that leading wheat producers are now adopting a more discriminating approach to pesticide use. During the 1980s, it was often quoted that the most profitable wheat growers used more than average amounts of pesticides on their crops. However, since 1990, the 25% of farmers producing the highest gross margins in wheat in East Anglia have spent less than average on pesticides (Table 2). There are alternative explanations for this data, such as the more varied rotations on the better and higher yielding soils reducing the cost of weed control in wheat and the adeptness of some farmers to buy pesticides more cheaply.

Recently completed research has proved that there is still considerable potential to reduce pesticide usage further. Therefore, it is clearly time for new initiatives to ensure that the whole industry more closely matches pesticide inputs with crop requirement. The easy to adopt approaches have already been put into practice by the leading farmers. The newer approaches being researched to reduce further pesticide usage further are inevitably more complex. Hence, based on previous experience, their transfer into practice will present an enormous challenge.

Table 2. Percentage of the mean pesticides costs/ha of the upper quartile and lower quartile of wheat producers in Eastern England, ranked according to gross margin/ha, 1989-1995. Source: Murphy (1997).

	1989/90	1990/91	1991/92	1992/93	1993/94	1994/95	1995/96
Upper quartile	112%	97%	93%	93%	96%	98%	94%
Lower quartile	88%	112%	104%	98%	103%	102%	107%

PESTICIDE OPTIMISATION VERSUS PRAGMATISM

Optimisation, in the simplest sense of maximising the benefit of pest, disease or weed control in a specific crop, involves an assessment of the impact of the target organisms on crop yield and quality, identifying the pesticide product, the rate to apply and defining the time of application. Farmers are averse to risk, having learnt from experience that failure to take action sooner rather than later can result in severe repercussions. The risk assessment, usually based on previous average experimental results, may prove difficult to estimate or even prove to be

wrong in individual circumstances. Also, there is the constant uncertainty over future weather conditions, which may influence the threat from the target organism and/or the ability of the farmer to apply a pesticide.

Hence, for these and other reasons, optimisation is currently compromised by a large degree of pragmatism. There is no better example than in weed control. Delaying treatment of winter wheat for annual grass weed control in the autumn until all or sufficient of the weeds have emerged to assess a treatment threshold, i.e. the infestation of weeds estimated to be necessary to make a herbicide application worthwhile, may result in an application which is too late for the level of control required, leading to re-treatment. In addition, weeds are generally patchy in nature causing great difficulty in assessing their true threat to the cropping system. A major field investigation recently concluded that an insurance approach using lower than recommended doses was more cost-effective than using thresholds (Proven et al., 1991).

Farmers are very suspicious of weed thresholds. Failure to control weeds in one crop can result in harvesting difficulties, leading to additional machinery costs and also result in an increase in weed numbers in future crops, where difficulties in their control may occur. These factors should be taken more into account by weed scientists when identifying realistic thresholds (Orson, 1997). However, technological advance in the form of broad-leaved crops genetically modified to be tolerant of non-selective herbicides may, in some circumstances, provide farmers with more confidence to adopt weed thresholds. Currently, farmers are overzealous in controlling annual broad-leaved weeds in winter wheat because they are either difficult or expensive to control in the broad-leaved crops which share the same rotation. On heavy soils, where there are many farms with only oilseed rape and wheat in the rotation, the introduction of herbicide-tolerant rape could result in effective control of broad-leaved weeds in this crop. This may lead to farmers being more willing to leave untreated low populations of broad-leaved weeds in their winter wheat, not only resulting in lower herbicide usage but also the surviving weeds providing a food source for farmland birds.

Examples of pragmatism influencing the optimal use of pesticides can also be cited with fungicide usage in winter wheat. In recent experiments, the more disease resistant cultivars often require only one triazole-based fungicide application to optimise use. However, a significant delay in application due to weather would negate any advantage from this approach when compared to a prophylactic approach. Hence, those farmers tailoring their disease control programme according to the genetic resistance of the crop are adjusting their approach in these cultivars by applying a reduced rate of the fungicide a few days prior to the optimal timing and then 'topping-up' the rate a couple of weeks later.

UK wheat growers, recognising the concern over the impact of insecticides on non-target species, are most successfully adopting threshold management in the control of summer aphids. Prophylactic treatment with an aphicide with the fungicide 'ear' spray used to be common. This fungicide application timing is now less commonly used (Table 1), particularly in hot and dry conditions which are conducive to an increase in aphid populations but not to diseases. In these same conditions, application is also more assured. Hence, a significant majority of producers apply insecticides according to simple and easy to assess thresholds and many select a more expensive but specific aphicide in order to increase the number of surviving predators, thus helping to avoid the need for re-treatment. The opposite applies to the autumn application of insecticides to control the aphid vectors of virus diseases in wheat. In this

situation, a more insurance-based approach is used. This is because the potential yield reduction is very significant, the cost of the treatment is low, the cost of application is minimal as the insecticide is usually mixed with an autumn herbicide and the treatment threshold is difficult to assess.

These examples emphasise the fact that the assessment of the likely impact of a pest, weed or disease is a major consideration in the adoption of practices which match more closely crop inputs with crop requirement. The additional management time to assess properly the challenge to the crop may outweigh the cost-saving achieved through lower pesticide use. This issue cannot easily be resolved, particularly at a time when the average size of arable farms in the UK is increasing.

FUTURE PROSPECTS FOR PESTICIDE OPTIMISATION

Farmers are anxious to remain competitive and hence will reduce all their costs, including pesticides, to a minimum, provided that they think that this will not result in an unnecessary increase in risk to their livelihood. Hence, pesticide optimisation techniques need to offer a robust approach to enhancing the income of the whole farm. In addition, they should be flexible enough to allow farmers to minimise the risk of weather conditions preventing application. Many of the recently introduced pesticides provide control of target organisms over a greater time scale, allowing farmers to use them in a more discriminating manner.

Much of the current research into pesticide optimisation is aimed at predicting yield loss on a field by field basis rather than on the average response to a pesticide input on which current thresholds are based. This suggests that further progress in optimisation is going to be more difficult to manage successfully in the field. Hence, there will be an increased need for effective technology transfer and it is envisaged that this will be in the form of inter-active computer programmes and electronic forms of information transfer in addition to face to face meetings between farmers and researchers.

It is also envisaged that technology will overcome a major problem threatening the implementation of optimisation techniques; that of assessing the likely impact of the target organism on the cropping system. It will eventually provide assistance in the physical assessment of risk through the use of tractor or satellite borne sensors. Coupled with the ability to apply pesticides spatially, this would particularly help in the adoption of treatment thresholds by being able to take into account the patchy nature of the challenge to the crop. The patchy nature of weed populations is one of the major factors which currently prevent the adoption of optimisation techniques with herbicides.

CONCLUSIONS

There is little doubt that the most successful farmers have reduced their use of pesticides in recent years to more realistic levels; in some cases by using simple treatment thresholds but modified by a considerable degree of pragmatism to ensure that risk is reduced to an acceptable level.

It is clear from current experience that the adoption of more optimal pesticide practices for both financial and environmental advantage are more likely if:

- farmers and their advisers have confidence in the decision making process
- the results can easily be judged on the farm
- the requirement for pesticides is easy to assess
- · the risk to the business is low
- there is sufficient flexibility in the time of application to overcome uncertain weather conditions
- unnecessary pesticide use will have an impact on non-target species and/or the environment.

Experience suggests that it can be difficult for farmers to accept even simple optimisation methods if they cannot easily assess the benefits. It is also clear that further progress towards pesticide optimisation requires the adoption of more complex approaches. Therefore, considerable effort and innovation will be necessary to put such approaches into practice, although developments in yield mapping to help measure their benefit, computerised decision support systems, crop assessment and spatial application techniques may provide a conduit for their adoption. In the longer term, other technologies, such as biotechnology, may have a profound impact on pesticide use. Eventually, information needs to be generated to enable the true optimisation of all inputs, not just pesticides, in order to achieve acceptable and sustainable financial margins and to reduce the environmental impact of individual arable farms. Hence, it is advances in knowledge and technology which will reduce the level of pragmatism currently involved in on-farm decision making and make true optimisation more achievable. Whilst in the past technology has been accused of being the cause of environmental damage, it should now be recognised as the means of enhancing the current environmental value of the countryside and of providing the ability to feed a rapidly increasing world population.

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ECOtillage: A SUSTAINABLE MANAGEMENT PROGRAMME WHICH REDUCES THE COSTS OF CROP ESTABLISHMENT AND WEED CONTROL, WHILST PROVIDING ENVIRONMENTAL RENEFITS

C D STRIDE

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ABSTRACT

ECOtillage is a sustainable reduced tillage system that provides solutions to the need to reduce the costs of crop production and improve the level of grass weed control in a lower value market. It is flexible in terms of cropping and weather, it allows rotational ploughing, and it suits progressive adoption. Key elements are the creation of a consolidated medium quality stale seedbed using discs and press/roll to facilitate weed control, straw breakdown, minimise slug damage and ensure a weather-proof seedbed. This is then drilled with a cultivator drill, and a good herbicide programme used in crop.

INTRODUCTION

Falling prices for cereals and other crops, as well as the lack of value in cereal straw, has had a major impact on the profitability of arable farming in 1997 and onwards, which has brought about a need to examine farming systems with the aim of reducing the costs of production. Most arable farms still rely on high cost and low output systems based on ploughing and power harrowing to establish crops.

Grant Thornton (Markham, 1997) figures show that 44% of the labour and equipment costs of crop production are purely to establish the crop. In total, these costs equate to an average of £118/ha, and some 71% of these are just to establish a seedbed.

Many farms are also challenged by a need to control an increasing diversity and infestation of annual grass weeds such as Blackgrass (Alopecurus myosuroides), Wild Oats (Avena spp), and Brome (Bromus) grass species, and also resistant strains, which are not always well controlled currently. These weeds now appear in all tillage systems, driven by the predominance of winter crops, often early drilled. There is a strong perception that ploughing aids weed management, and whilst this is true in terms of rotationally ploughing to bury a weed problem, repeated annual ploughing is a costly and poor way to control weeds and our work suggests a better alternative exists.

We believe ECOtillage, a form of Conservation Tillage, provides solutions to these problems, as well as overcoming the barriers / pitfalls often associated with reduced tillage both past and present. This paper describes the system and it's development, and shows how it meets the immediate needs as well as looking forward.

Technological advances in both cultivation and drilling technologies, as well as herbicides and management understanding, we believe bring new hope to sustainable agriculture for the future.

METHODS AND MATERIALS

ECOtillage brings together two areas of technology, Simba's tillage systems and Monsanto's experience in herbicide programmes to develop a combined systems approach which utilises many proven individual components.

Simba systems provide a reliable and proven way to establish crops, while reducing costs, and Monsanto herbicide programmes provide more effective control of a range of invasive grass weeds including resistant strains. It was not just a case of combining two systems, we have also have some synergy (elements of the systems come together to both reduce costs and improve weed control). We have developed the concept of making a firm stale-seedbed to actually encourage weeds to germinate and then kill them prior to planting the crop, this is critical to sustainable use of reduced tillage systems in the presence of grass weeds.

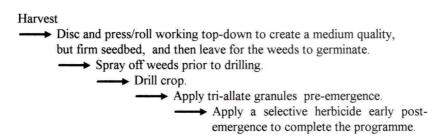
Studies were conducted on a large plot/ commercial scale, with an unreplicated strip or lattice design. To develop the cultural, tillage and chemical components of ECOtillage we conducted a range of individual and system trials in conjunction with various contract companies and farmer co-operators. Assessments were made of weed germination, crop establishment, weed control, slug numbers and harvest yield. In the absence of plot replication, assessments were replicated to improve consistency of results The trials reported are mostly from the 1996 programme and were drilled in the period 15th October to 5th November to allow weed germination in a dry autumn. Four similar studies from the period 1992-95 are also reported. The conventional programme was ploughed, power harrowed and drilled with an airflow drill. ECOtillage was disced and double pressed one or twice, rolled if need, and drilled with the Freeflow drill. The full herbicide programme was Sting CT (120g/l enhanced glyphosate formulation) applied in the stale seedbed, Avadex Excel 15G (15% w/w triallate) applied pre-emergence of the crop and full rate isoproturon mix herbicide applied early post-emergence of the crop. The low input comparison was either untreated, glyphosate in a stale-seedbed, or had isoproturon alone to show cultivation comparisons more clearly.

The programme has been tested and used by many farmers who have proven its use or the use of its components over many seasons.

The system of ECOtillage

The combination of many key components brings success in ECOtillage. Schematically the system is as depicted below.

The ECOtillage system/ programme



Critical areas

To ensure success of practising ECOtillage it is important to start early by planning prior to harvest. Cut the crop low, and if incorporating straw ensure a good chop and even spread of straw and chaff. Following as soon as possible after harvest, cultivation's should be made working top down to create a firm stale seedbed to encourage both straw breakdown and weed germination. By starting early and leaving sufficient time, weeds will germinate before drilling. It is important, as always, to review the need to subsoil to correct compaction problems. Cultivation's are best performed with heavy discs set to cultivate at two depths 1-3" and 2-5" for straw incorporation, shallower if not. Soil should be consolidated with a double press on medium to heavy land, or Cambridge roll on light land. The overall aim is to till the soil without soil inversion, and reduce number of passes, conserve moisture, not create clods, and always leave the profile in a consolidated state. Effective drilling is crucial to optimise establishment. The drill must be capable of accurate placement, and cope with both trash and a consolidated seedbed. A herbicide programme should always be used, starting with controlling those weeds that emerge in the stale seedbed, and following with a programme in keeping with the weed problem and the WRAG guidelines (HGCA, Moss and Clarke 1993) to control weeds emerging in the crop.

RESULTS AND DISCUSSION

Overcoming barriers

The main perceived barriers to widespread adoption of reduced tillage now are the memory of the failed direct and minimum tillage systems in the 1970's, problems of extreme weather conditions delaying drilling, inadequate weed control compared to a plough based system, and the reduction in plough usage, it traditionally being the main cultivation machine used on the farm. ECOtillage addresses all these areas to provide a solutions approach as indicated below.

a) Drilling for optimum crop establishment

Heavy, disc based direct drills often smeared the seed into a slot, generally in direct contact with straw. This lead to poor seed to soil contact, toxicity, slug and compaction problems in wet years, together with disease carryover at the surface due

to high volumes of straw. The problems resulting can limit the yield of the next crop.

ECOtillage provides a good consolidated soil/straw mix which leads to rapid straw breakdown and the lack of trash concentration in the profile. The Freeflow drill can establish all seeds into trash without compromise to seed placement and emergence (ADAS, 1992). Twin rollers give adequate consolidation for optimum seed to soil contact, and reduced slug problems (ADAS 1992). Work at Long Ashton has also shown that slug damage to cereals can be reduced by cultural means designed to prevent slugs reaching seeds even in presence of straw trash in a minimum tillage system. These are fine, well consolidated seedbeds, and deeper (4cm) even seed placement (Glen et al, 1990, 1993).

b) Minimum tillage to produce seedbeds

Traditional light (<100kg/disc) discs required multiple passes to be effective. The lack of pressing in combination with discing reduced soil to seed/straw contact and left a loose profile. This resulted in over worked soil, loss of moisture, panning at depth, slug problems, and poor germination of weed seeds prior to drilling.

'C' series discs progressively cultivate and mix in one pass for an optimum mix of straw, soil, and weed seeds. Panning and blockages are minimised due to progressive working depth. The combination with a double press, and or Cambridge roll depending on soil type, ensures soil is consolidated through to depth for good seed/straw contact giving effective breakdown and early weed germination.

c) Extreme weather conditions

The lack of effective discing and pressing led to the creation of a loose seedbed which in wet years proved difficult to traffic, and was prone to water logging, delaying drilling. Ineffective subsoiling exacerbated these problems. In dry years, the overworked loose profile lost moisture which failed to germinate weeds, and lead to poor germination of the next crop.

The effectiveness of the 'C' series and press/ roll combination with selective subsoiling is to provide a profile which is trafficable in the wet, drains readily, and allows good root growth. It also retains moisture in the dry for effective germination of weeds in the stale seedbed, and creates optimum conditions for establishment of the next crop.

d) Ploughing

The perceived benefits of ploughing are to bury weed seeds and straw, providing a clean soil surface. Once cultivated to form a consistent seedbed, this is capable of being drilled easily, in a wide range of conditions, with all soils and crops.

Regular inversion, however, returns a proportion of buried, often dormant, seeds to the surface to infest the next crop. The need to create a fine seedbed for a conventional coulter drill to operate properly implies multiple cultivation passes after the plough which loose moisture in the dry, destroy soil structure in the wet, increase costs, reduce output and may result in sub-optimal crop emergence. The low output and high

skill requirement of a plough, combined with high fuel and wearing parts usage, all increase associated costs of establishment with this traditional method

The stale seedbed approach of ECOtillage produces the seedbed progressively from the surface, minimising clods, whilst at the same time effectively controlling weeds. Surface tilth is retained, reducing cultivation requirements which minimises susceptibility to extremes of weather. The robust design of equipment is capable of high output with increases in output of 100% (ADAS, 1992) at low cost, with unskilled operators. Being a flexible system can be tailored to conditions (wet or dry) and crop requirements. This then enables the use of the plough rotationally if required, for example to suit certain crops and conditions.

The use of the high output ECOtillage system allows operations to be made on the days when soils should be driven on, cultivated or drilled to achieve intended drilling dates and crop yields. Even if conditions are adverse the system is stable.

e) Weed control

One of the principle barriers to adoption of reduced tillage is that grass weeds are known to increase more quickly in such systems. The old minimum tillage systems of the 1970's were overcome by grass weeds that could not be controlled. These systems, however, were also defeated by a limited selection and reliance on selective grass weed herbicides, and use of contact non-selective herbicides. Burning of straw and stubble created ash which bound residual herbicides and reduced their performance.

ECOtillage relies not only on the ability to create a stale seedbed to encourage weeds to germinate outside the crop which are then sprayed out with a translocated non-selective herbicide based on glyphosate and formulated specifically to control annual weeds, but on a programme of herbicides. The key is to reduce the weed seed bank. This is done by targeting weed control prior to drilling and in the crop, to minimise weed levels and thus seeding in the crop.

To encourage germination in the stale seedbed consolidation is key. Table 1 shows data from four trials in the dry autumn of 1996 and clearly shows the benefit of increased consolidation on weed germination on a overall germination of weeds (Blackgrass, Wild-oat, Volunteer wheat and annual broad-leaved weeds).

Table 1. Effect of consolidation on weed germination in a stale-seedbed in four trials on medium-heavy land (weeds plants/m²).

Disc	Disc + Roll	Disc + Double Press	Disc + Double press + Roll
63	87	133	191

The value of the herbicide programme is illustrated in Table 2, where it can be seen that the control of emerging Blackgrass in the stale-seedbed alone reduced germination in the crop by 56%, the entire programme by 99.6%. Table 4 shows recent results.

Table 2. Control of Blackgrass assessed in wheat with components of the herbicide programme (4 trials 1992-1995, average 150 heads/m² in untreated).

Glyphosate	glyphosate + tri-allate	glyphosate + tri-allate + isoproturon	isoproturon alone
57	70	99.6	74

ECOtillage working on farm

Recent trials results show higher crop plant counts, from equal or lower seed rates, gave slightly higher yields in dry year of 1996/7 (Table 3). These trials also show that by encouraging weed germination in a stale-seedbed and controlling those weeds prior to drilling then lower weed numbers occur in crop (Table 4).

Table 3. Crop establishment and yield results from 6 studies in winter wheat in 1996/7 with full herbicide programme.

System	Seed rate (kg/ha)	Plant counts per m ²	Yield (t/ha)
ECOtillage	184	451	8.24
Plough/ conventional	209	410	7.90

Table 4. Weed control results from 6 studies in winter wheat in 1996/7

Average weeds levels were as follows in untreated plots; Blackgrass 213 heads per m², and Wild-Oats 93 heads per m²

System	Blackgrass germination in stale seedbed plants/m ² (6 studies)	Blackgrass heads/m2 in June/July (6 studies)		Wild-oat heads/m2 in June/July (3 studies)	
Herbicide programme	None	Low	Full	Low	Full
ECOtillage	144	49	1	16	4
Plough/ conventional	1	124	1	56	8

Comparing the costs of just establishing the crop (tillage, costs of weed control in stale seedbed, and costs of drilling and rolling) there are substantial savings in both cost and time as calculated for an average 700 acre (283 hectare) farm (Table 5). Savings in fixed costs are an important area not addressed on most farms. Saving in number of days means tillage and drilling can be performed speedily on the best days to ensure optimum crop establishment, and prevent delays in drilling due to adverse weather.

Table 5. Average costs and labour need to establish 700 acres (283 hectares) on medium to heavy land (Nix 1996, 1997).

System	Farm cost (£/ha)	Days
ECOtillage	79.6	43
Plough/ conventional	106.3	92
Saving	26.7	49

Many environmental benefits can be seen; reduced erosion due to wind and rain, reduced leaching of fertilisers, and less requirement for herbicides and insecticides, compared to traditional plough-based systems (Jordan et al. 1996, 1997).

Future developments

a) Cultivation aspects

Current developments include the combination of working elements to reduce costs and number of passes further, with refinements to further improve performance. The Mono is a combined disc, press and selective depth subsoiler machine that reduces passes to minimise costs and maximise output, reduces moisture loss, while limiting loosening to the areas needing it. The Cultipress is a press cultivator which leaves a level consolidated profile for weathering, and allows the tilth produced to be retained evenly on the surface when drilling for optimal emergence.

b) Alternative crops

ECOtillage has been successfully applied ahead of the following crops, but further developments are in progress to facilitate widespread adoption.

In Sugar Beet, on medium soils, the previous crop stubble is disced and pressed once, and subsoiled to ensure a full shatter at depth and a level consolidated surface. This is over wintered, and weeds sprayed off prior to drilling in the normal manner. On light soils, research continues into the optimum technique to establish the crop and reduce wind erosion. Some 25% of soils on which Sugar beet are grown are subject to wind erosion it is thought (British Sugar, personal communication, 1997).

In Vining peas the use of the Freeflow drill directly into a sprayed off over-wintered stubble gives the benefits of minimal inputs, minimal moisture loss at drilling, combined with above average yield potential. Additionally, retaining a surface stubble minimises the risk of wind and water erosion on light soils. Subsoiling should be considered if stubble's are compacted or unlevel.

In potatoes the system is to use the Mono to prepare a bedformed seedbed, which is then over-wintered, weed growth sprayed off, and planted conventionally in spring. Alternatively, stubble's can be disced and pressed twice in autumn, over wintered, weeds sprayed off, and then beds formed and planted.

c) Weed control technologies

At this conference a new molecule was revealed for broad spectrum weed control in Wheat. JV485 is a pre-emergence herbicide and is ideally suited to the ECOtillage system in that it controls a wide range of annuals grass weeds including Blackgrass, even that resistant to other herbicides. Its performance is not affected by the presence of incorporated chopped straw. At Brighton conference 1995, sulfosulfuron was launched, and is now used commercially in parts of eastern Europe for the control of brome grasses and couch, two weeds which can become a problem in reduced tillage systems. The incorporation of crops tolerant to herbicides like Roundup Biactive, such as Oilseed Rape and Sugar Beet, will also provide a fresh opportunity in the rotation to control annual grass weeds, even those resistant to other herbicides like Blackgrass.

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ADOPTING INTEGRATED CROP MANAGEMENT - A PRACTICAL WHOLE FARM APPROACH FOR THE OPTIMISATION OF AGROCHEMICAL USE

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ABSTRACT

The need to find practical, achievable and realistic options to optimise agrochemical use for farmers is becoming increasingly important. Integrated Crop Management (ICM) offers such an option that meets the business needs of farmers and the environmental and food hygiene, health and safety concerns of the general public.

Focusing on a whole farm approach that is site specific LEAF (Linking Environment And Farming) has set up demonstration farms throughout the UK and developed guidelines and the LEAF Audit. The LEAF Audit is a self assessment approach for farmers to prepare them for future challenges, focus their thoughts on their farm practices and take steps to adopt ICM - a practical way to manage resources effectively and optimise agrochemical use.

INTRODUCTION

What are the challenges?

For the past two years we have seen continued food scares with BSE, E Coli, organophosphates in carrots, to name a few, and these concerns are not decreasing. Quoting from an article in Vogue magazine (Robson-Scott, 1997) 'Wholewheat bread pesticide residues lie on the outside of the grain, so non-organic wholewheat bread contains more pesticides than non-organic white bread.' Furthermore, Graham Harvey's book states 'Down below the ripening ears, on the bare earth, no bugs or insects are visible among the forest of stems. Nothing lives here; the pesticides have seen to that. Those that don't kill the insect predators directly destroy the smaller invertebrates on which they feed.' (Harvey, 1997). The result has been for calls for concerted action to be taken by addressing the genuine concern of the general public as to how their food is produced.

Although we often deal with perceptions in some specific cases these statements can be justified. This is where Integrated Crop Management (ICM) has a vital role to play. While not being prescriptive in its approach, it allows farmers to make their own choices through informed management decisions. It also encourages farmers to be far more disciplined in their record keeping and communication channels, both on the farm and beyond the farm gate. Indeed the development of the LEAF Audit (LEAF, 1997a) has acted as a 'conscience' for farmers leading to improvement in farm practices and giving credit where they have got it right.

This paper will focus on ICM and the approach LEAF is adopting to encourage farmers to develop and practice ICM on their farms including some examples that have been adopted by farmers to reduce agrochemical use which hopefully will reduce the risk of environmental pollution.

THE LEAF APPROACH

Over the last 6 years LEAF has been actively promoting ICM through the setting up of demonstration farms and the development of the LEAF Audit. There are now 24 LEAF Demonstration Farms in England and Scotland and a growing membership base of over 800 farmers, 94 Corporate members and 24 agricultural colleges. LEAF believes that attention to detail demanded of ICM can result in better utilisation of resources, optimisation of inputs and reduced costs.

Why ICM?

ICM is a whole farm policy which aims to provide the basis for efficient and profitable production which is economically viable and environmentally responsible.

The key for success in the development of agricultural systems is that they are self supporting in the marketplace, respond to consumer demands and are realistically achievable by the majority of farmers. As such the approach that LEAF has adopted in the development of ICM is that it must:

- ensure commercial reality is achievable
- be acceptable to the customer/consumer
- provide environmental benefit
- be developed without the need for legislation
- be market orientated
- be developed with a long term vision in mind
- be self-supporting in the long run
- be of high probity and ensure high standards of production are profitable and traceable.

Interpretation

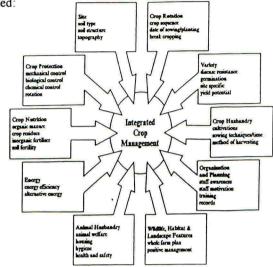
Many growers have adopted ICM on a voluntary basis as a natural progression of their current practices. To these growers, the rewards are obvious: improved safety, environmental protection, improved staff motivation, addressing public concerns, gaining market opportunity and enhanced economic returns. The continued development of a successful, long term ICM programme is critical if the UK is to meet future needs for high quality, good value food while creating environmental and economic harmony between producers and consumers. Technological advances, such as precision farming will provide the keys to sustaining successful ICM programs (LEAF, 1997b).

However to be successful, any criteria used to judge the effectiveness of a given ICM program must be practical (science based), agronomically sound (allow for variation within and among crops and soil types), economically viable (cost effective) and have achievable and measurable

objectives. Specifically ICM is not formula driven, but is broad in its interpretation and takes into account differences among commodities, as well as the areas in which they are grown.

The principle of ICM

The principle of ICM is illustrated by the following diagram which shows the different elements and how they are interrelated:



The goals of ICM

- To ensure the production of high quality food and fibre in a sustainable, environmentally sensitive and economical manner
- To minimise the risks to human health and to the environment
- To enhance farmland resources
- To restore public confidence through good stewardship

In one form or another ICM has been around since the advent of agriculture, but scientifically based programmes focused in this area have only been around for a few decades. The development of ICM is a dynamic process that adopts the findings of research and the introduction of new technologies. It is a culture of continuous improvement and involves participants such as university and extension researchers, production agriculture, environmentalists, advisors, industry scientists and most importantly, farmers. Depending upon the site and the problem, programmes may differ for the same crop in different areas. However, the underlying procedures and management approaches are the same and should thus be encouraged. ICM is a management systems approach and the LEAF Audit really does focus the mind on all these areas.

THE NECESSARY ELEMENTS OF A SUCCESSFUL ICM POLICY

Practical

ICM programmes must be based on sound science. This process involves the coupling of solid field biology research with workable delivery systems.

Realistic

ICM programmes, while being guided by a well defined philosophy, must be broad enough to allow for variation from area to area and from time to time throughout the growing season. ICM policy elements must allow for enough flexibility and options to accommodate differences due to geography, temperature, climate and other variables that exist within any given commodity. Rigid, specific formulas will not provide the necessary flexibility.

Economical

The bottom line is that in order for ICM programmes to be successful they must work economically for the grower. Rigid, formula driven ICM programmes will be likely to result in increased grower costs ie. exposure to economic loss, because growers may not have the flexibility to adapt procedures to problems that are specific to a particular crop or region.

Achievable

ICM programmes must include measurable objectives. Measurements must be based on sound science rather than philosophy. Criteria that are too narrowly focused may invite manipulation rather than interpretation needed for meaningful change. For example a narrowly focused measurement, such as a fixed pesticide use reduction. Indeed the application rate of pesticides per hectare has decreased but this must be considered with the fact that some active substances are highly effective at low rates - grammes as opposed to kilograms.

Dynamic

It is important to encourage the adoption of farming systems that take into account new technologies and developments in line with the best of traditional methods, such as rotations, soil management etc. For example rotations are decreasing on many farms, especially on heavy land, due to the present regime often comprising wheat and oilseed rape. This however does not increase or maintain the diversity of wildlife.

A PRACTICAL APPROACH FOR FARMERS

Along with the perceptions about the farming industry, farmers often perceive that the risks associated with ICM are both costly and can create problems. Our work has proved different, from experiences of farmers who do the LEAF Audit and our LEAF Demonstration Farmers.

When LEAF Demonstration Farms were first set up there were many farmers who said 'I farm like them' and others who said 'I would like to farm like them'. This in mind, together with the development of environmental standards in other industries, led to the first LEAF Environmental Audit, launched in 1994. March 1997 saw the launch of the new LEAF Audit building on the experiences of the first three years of the original audit. In effect it encourages farmers to question their current practices and consider other options.

The new LEAF Audit comprises seven parts, namely detailed examination of crop protection, soil management and crop nutrition, organisation management and record keeping, energy

efficiency, pollution control and waste management, wildlife and landscape management and animal husbandry. Farmers may conduct the audit as a whole or in individual modules to provide a stepwise approach. Taking the crop protection section as an example, it focuses on a detailed series of questions on documentation, training, decision making, record keeping, monitoring, planning, equipment and facilities and targets for action.

Farmers complete the LEAF Audit annually and return the completed forms to LEAF. It is not a pass/fail document - it is a management tool. We then send details of the general trends of all those who have responded, together with indicators for improvement and a personalised action plan. Although the LEAF Audit is currently only a self assessment approach, the benefits on setting people on the right track are enormous, it really does help 'focus the mind' and 'make you think'.

Early 1998 will see the LEAF Audit on computer disc, together with the development of an eco-rating system and hypertext link for information and advice. This will provide a starting point for farmers to gauge their progress not only qualitatively, as with the current audit, but also quantitatively.

PRACTICAL SOLUTIONS TO REAL PROBLEMS

Integrated Crop Management not only is a whole farm approach it is also a whole industry approach. Carried out with the right attitude, advice specific to the site and training not only will it save unnecessary applications of pesticides, it will also save money. ICM gives farmers the incentive to move away from a total reliance of pesticides to a more informed approach in conjunction with other options. Although the adoption of ICM does require commitment and may involve a higher risk and management input in the early stages, the approach is highly challenging and motivating for all involved. It can also be more profitable. The rewards far outweigh the disadvantages.

Seed rates

Experience on one of the LEAF Demonstration Farms in Norfolk has shown that seed rates can be reduced, especially on light lands resulting in reduced diseases pressure and improved stand ability, the final samples being identical at 2.4% screenings, 1.43 nitrogen at 7.41 tonne/hectare (Purslow). Indeed increased drilling densities have resulted in a reduction in root mass, as well as a hindered uptake of potash from thick stands. This in turn leads to both decreased winter hardiness and drought tolerance.

Seed rates can also be used to counter other problems on the farm. Decisions are based on the whole farm with a view to avoid any problems in the first instance and identifying risk areas susceptible to certain problems. For example with wheat bulb fly (*Delia coarctata*) seed rates should be held high enough to cope with the attacks. The use of organophosphates can be very damaging to the environment, particularly natural predators as these chemicals are actually targeted to the soil surfaces (Frampton et al. 1996).

As a further example before assessing the need for application of pesticides in early spring one should count the plants and assess the numbers of tillers and the potential ear placing. A full

yield comes from 600-800 ears per square metre and if there are any more the wheat bulb fly can be left. At the onset of attack, the number attacking, should be taken into account and subtracted from the potential ear placing. So much spraying for bulb fly often is tantamount to revenge.

Drilling date is also important at the Manydown Company, the LEAF Demonstration Farm in Hampshire and one of the LINK-IFS research sites. Delayed drilling, to avoid spraying for aphids in the autumn has been successful in reducing broad-leaved weed emergence but can be more costly due to more expensive products being used the following spring (Holland 1997). This has to be balanced with the fact that late drilling can often reduce yields and poorer seedbeds can increase slug problems (monitoring is essential). This again emphasises the importance of recognising that ICM is not a single issue or prescriptive system and that there is a strong interaction between many practices and problems that have to be considered before action is taken.

Fungicides

Each disease has its own threshold level and weather conditions to become an epidemic and requires good monitoring and the ability to respond (ie with a sprayer if the situation changes). It is essential to know these before applications are made. Thus Septoria (Leptosphaeria nodorum) will not pose a threat until GS32 at a time when rain splash drives the disease. Brown rust (Puccinia recondita) will not be a problem until we have dew and warmish nights. Take-all (Gaeumannomyces graminis) is reduced in severity if one uses green manures, such as mustard and to a lesser extent poultry manures. These factors should be then cross referenced with the plants natural ability to defend itself. Only if these indicate high risk should chemicals be applied. Resistant varieties are also important (Parry, 1990).

As with all issues in farming there is no single answer and it should be noted that some diseases can be exacerbated by low nutrient supply and vice versa, for example botrytis (Botrytis fabae) in peas is made worse by low levels of potash. Furthermore the development of habitats and wildlife areas is again important. With ICM the whole farm has to be taken into account to ensure this balance results in the best decisions to be made on the farm. It brings together both the best of modern technology and the best of traditional methods. For example, yield mapping shows us that the headlands are often the lowest yielding areas of the farm and these areas can be exploited to encourage increased natural predators and thus optimise the level of pesticide use to combat various pest species.

Findings on the Manydown Company Farm have shown how threshold levels are often lower than need be and we could get away with a higher tolerance of insect population especially in oilseed rape. Observations have shown that the right selection of chemical, at the right time and place results in lower costs and more effective control.

Weeds

Rotations are essential to ensure a balanced approach. The most recent results from the LINK IFS project has shown an average of 33% less pesticides in IFS fields. On the Manydown Company farm, black grass (Alopecurus myosuroides) is one of the main agronomic threats on most fields together with a wide range of broadleaved weeds, including cleavers (Galium

aparine). Since seed crops are produced, weed management is a high priority but the creation of Conservation Headlands has allowed many rare arable weeds to survive. Mechanical weeding has not been adopted because there are few opportunities when the crop, weeds and soil conditions are suitable. On heavy lands it is often better to use break crops to reduce the effects of blackgrass.

The crops most threatened by pests are oilseed rape and vining peas, the latter having a zero tolerance. To reduce environmental impact, more selective insecticides have been used in the integrated system. Cereal aphids, although often present, rarely exceed threshold levels until late in the season, when there is no benefit from spraying. Cereal aphids can be reduced by increasing predator numbers, which in turn increases game/wildbird food.

Applications

When pesticides are used and justified in the economic and environmental context, farmers should make sure they are effective. If it needs a full dose then give it such. Blackgrass is often better treated with the full recommended rates once in the autumn using a mixture of chemicals, such as Triallate/IPU/Trifluralin, than small doses which may need a number of applications and could create resistance. Smaller doses repeated are often useful in other situations to give the environment time to break them down and reduce any detrimental toxic effects. If there are enough natural predators about one can reduce the level of insecticides to take out the first damaging flush of insects and then allow the predators to take over.

In the future it is likely that seed treatments may offer a valuable and targeted option. For the application of pesticides for example, in sugar beet, these treatments have the advantage of reducing the toxicity to the predatory species which can create a natural balance. On top of all this it is essential that application techniques are accurate. Farmers who complete the LEAF Audit judge calibration of machinery a high level of priority and findings from the Private Costs and Benefits of Pesticide Minimization (Risk & Policy Analysts Ltd et al, 1997) reinforce the need to make sure all application equipment is checked and as near-perfect as possible. Findings from this report have estimated that losses from this alone amount to £175 million per year. Indeed in some parts of Germany and in the Netherlands there is an annual MOT test for sprayers and in the UK AEA are developing a sprayer test.

Timeliness is next to Godliness. The difference between a good and a bad farmer they say is a week. Rates can be significantly reduced if timeliness is correct, if one is late the rates have to be increased to cope with the increased pressure (Whale, 1992). This means an improved management system on farms should decrease costs. The choice of chemical use boils down to attention to detail together with monitoring, planning, training, good communication and good independent advice.

CONCLUSION

ICM does offer some real opportunities to be more focused on an informed management approach, the following are some of the key principles. LEAF adopts an approach that:

fosters cooperation between regulators and growers

- involves collaboration across the community, ie growers, advisors and farm suppliers
- considers geographical, seasonal, climatic, biological and cropping differences
- adopts evaluation criteria that are

practical (science based)

realistic (allow for variation within and among crops)

economically viable

and have measurable and achievable objectives.

- encourages the extension and promotion of ICM research, development and demonstration programmes at all levels
- recognises ICM as a mature concept which improves the environmental and economic consequences of crop and livestock management through the better use of information and technology

There is a willingness among growers and consultants to use innovative production techniques as long as those techniques can demonstrate equal or greater economic benefit. More than ever growers and the agricultural industry as a whole are aware that to stay in business they must ensure favourable public perception of their practices. ICM can truly be a solution to address both the needs of the farmers and the public.

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THE USE OF IMPACT RANKING INDICES IN THE REGULATION OF PESTICIDE USE: THE CASE OF TAXES.

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ABSTRACT

Increasingly, governments are considering the use of models that rank pesticides by their environmental impact. The purpose of such models are twofold; firstly they seek to influence agricultural practices, either at an individual farm or regional level, and secondly they seek to influence policy decisions at a national level. It is widely accepted that such models fall far short of formal quantitative risk assessment, and yet it has been argued that quantitative risk assessment falls far short of the requirements of policy makers. Pesticide ranking indices, therefore, seek to fill an informational gap at both the farm and policy levels. This paper introduces pesticide ranking indices, and discusses policy areas where such models might make a useful contribution. In particular the paper addresses the issue of pesticide taxes and the use of ranking indices in their design.

INTRODUCTION

As no single parameter can fully describe the environmental impacts associated with pesticide use, it is argued that multi-parameter environmental impact models are required (Levitan.et al. 1995). Pesticide ranking indices are models of environmental impact that report the potential environmental impacts of pesticides. Their purpose is to simplify complex relationships about the hazard posed by pesticide use and provide this information to decision-makers (farmers, policy makers, consumers) in an easily understandable manner. As such, ranking indices can be used in a variety of ways; for example, they can assist farmers to move toward Integrated Pest Management (IPM) production, form the basis of IPM accreditation schemes, help monitor and evaluate policy measures to protect the environment, target R&D efforts and form the basis of a tax, charge or subsidy programme.

Available indices vary greatly in methodology, input variables and output. They range from simple reporting of sales of pesticides; toxicological registers that categorise pesticides into hazard groups (e.g. World Health Organisation), complex equations and cryptograms that seek to place a single numerical value to environmental impact (Kovach et al., 1992), and economic analyses that seek to place a monetary value on the hazards associated with pesticide use (Higley and Wintersteen, 1992). This paper examines the potential utility of one available ranking index, the Environmental Impact Quotient (EIQ) model of Kovach et al., (1992) to design pesticide taxes. The use of this particular model does not imply that it is preferred over all others, it merely serves as a suitable example.

THE ENVIRONMENTAL IMPACT QUOTIENT MODEL

The purpose of the model is to allow farmers wishing to adopt Integrated Pest Management (IPM) strategies to choose between more or less harmful pesticides, and thus more or less harmful pest management strategies. The EIQ model has two outputs. One is an impact figure, the EIQ, assigned to individual pesticides, the other is the Field Use Rating (FUR) assigned to pest management strategies. The FUR is equal to the EIQ of the individual compound multiplied by the % active ingredient (a.i.) multiplied by the use rate. The formula for determining the EIQ value of an individual pesticide is given below, and an example of the outputs of the EIQ models are presented in Tables 1 and 2.

$$EIQ = \{ [C(DT*5) + (DT*P)] + C*(S+P)/2*SY) + (L)] + [(F*R) + (D*(S+P)/2*3) + (Z*P*3) + (B*P*5)] \}/3$$

where:

DT = dermal toxicity

C = chronic toxicity

SY = systemicity

F = fish toxicity

L= leaching potential

R = surface loss potential

D = bird toxicity

S = soil half life

Z = bee toxicity

B = beneficial arthropod toxicity

P = plant surface half life

Table 1. An excerpt from the EIQ tables of Kovach et al., 1992.

Compound	EIQ
copper sulphate + lime	54
thiram	37
metiram	30
copper sulphate	27
mancozeb	24
maneb	24
zineb	20

The EIQ value is calculated by taking the average impacts from the farm worker, consumer and ecological component of the equation, each of which in turn is made up of multiple variables (Table 3.). Each of the variables included in the EIQ equation are both weighted and rated. The weight (1, 3 and 5 representing low, medium and high respectively) is assigned to each of the attributes displayed in Table 3 and reflects the relative importance of that attribute. The rating (the values applied to the toxicity of the compound, 1, 3 or 5) is multiplied with the weighting to give a value that forms part of the overall EIQ equation.

The EIQ provides the decision maker with additional information not provided by current pesticide legislation (91/414/EEC) or quantitative risk assessment. Decision makers can explicitly choose to avoid pesticides with a high EIQ value and opt for those pesticides at the lower end of the EIQ scale. Together with the FUR this provides potential impact data for

individual compounds and entire pest management strategies at the farm, sectoral, regional or national levels, potentially over multiple time horizons.

Table 2. A theoretical EIQ FUR for apple production in the UK.

Compound	EIQ	%A.I.	Rate	EIQ FUR
dithianon	35.9	0.75	2.2	59.20
captan	28.6	0.80	9.8	224.20
pyrifenox	34.9	0.20	1.8	12.56
bupirimate	41.2	0.25	4.4	45.30
lorsban	52.8	0.48	3.0	76.00
pomex	22.6	0.50	8.2	92.70
dicamba	38.7	0.21	5.0	41.60
amitrole	20.5	0.22	5.0	23.10
simazine	15.7	0.50	1.7	133.30
diuron	20.5	0.50	1.0	10.20
TOTALS			42.1	718.16

Table 3. EIQ for captan.

Attributes	EIQ
Applicator effects	10.0
Picker effects	6.2
Total farm worker component	16.2
Consumer effects	4.1
Groundwater effects	1
Total consumer component	5.1
Aquatic effects	5
Bird effects	6.2
Bee effects	9.3
Beneficial effects	38.3
Total ecological component	58.7
Total EIQ	26.7

TAXES, CHARGES, STANDARDS AND SUBSIDIES

Over the last decade the use of economic instruments, such as taxes, charges and subsidies, has grown in importance (Barde and Opschoor, 1994). A tax has two main purposes, to raise revenue for the Government and to alter behaviour. A charge (or product tax) is an administrative measure which aims to change behaviour but not raise revenue, and a subsidy aims to encourage a change in behaviour through rewarding voluntary change and innovation. Standards represent levels of pollution deemed permissible by society, and may be enforced by the legislature, or form part of an economic instrument. In general taxation and charges

make organisations less profitable, whilst subsidies make organisations more profitable (Baumol and Oates, 1988).

Ideally, the amount of tax (or charge) placed on a product should reflect the amount of damage it causes (Barde, 1997). Similarly the amount of subsidy offered should reflect the amount of damage avoided. In the past, however, a lack of environmental impact data has hindered the use of economic instruments generally, but especially so with taxes (Cropper and Oates, 1992). If pesticide ranking models could be used with some confidence, then it is clear that the figures of environmental impact assigned to pest management strategies could reflect the potential hazard to the environment, and therefore, in some respects provide the data on environmental damage required to design pesticide taxes. Whilst the identification of the true marginal external cost of pesticides hinders the adoption of Pigovian Taxes, product taxes and industry standards may be a convenient solution (Barde, 1997). Also, subsidies, even when not truly representing damage avoided by alterations in behaviour, could be a powerful incentive to adopt less harmful pest management strategies.

Product taxes (or charges) are taxes placed on "damaging" products, or inputs, the purpose being to either force the user to reduce the quantity of that product or input used, or to encourage the user to switch to an alternative (less damaging) product or input (Cropper and Oates, 1992). One of the main problems with this strategy is often the lack of information on existing substitutes (Barde, 1997). The EIQ model, however, explicitly provides the farmer with information regarding the environmental impacts of various pesticides. For example, referring to Figure 1, assuming that each of three randomly selected pesticides, sevin, guthion and thiodan, are at least as efficacious as each other and target the same pests, then from a purely environmental point of view thiodan is preferable to sevin and guthion is preferable to both thiodan and sevin (data from Kovach et al., 1992). Whilst, at one level, it is important to inform farmers of the environmental impacts of individual compounds so that voluntary changes can be made, by making alterations to the market price of individual compounds, in the form of a pesticide tax based on EIQ value, it may be possible to bring about an enforced change in behaviour (if that is deemed desirable). Figure 2 indicates how an increase in pesticide price due to taxation might affect use rates.

One of the criteria that has to be met when designing a tax is that there should be a direct linkage between the tax and damage, with the region specific emission tax being the most efficient economic instrument available to policy makers. The data requirements for designing such instruments however, are both immense and complex. If there were complete confidence in models such as the EIQ then the FUR could be used as a representation of potential environmental hazard. This could be used as a proxy for environmental damage and could thus form the basis of a tax - standard amalgam as proposed by Baumol in 1977 (Baumol and Oates, 1988).

EIO's, TAXES AND SUBSIDIES

From an economic perspective the optimal level of pollution is where Marginal Net Private Benefit (MNPB) is equal to Marginal External Cost (MEC) (Figure 2.). Baumol and Oates (1988) argue, however, that the most realistic goal of policy makers should be to seek efficient, rather than optimal outcomes. They argue that setting a desired standard of environmental quality (or degradation) and enforcing or encouraging this standard with a tax or subsidy, is the most realistic mechanism for protecting the environment.

Figure 1. The comparison of three pesticides' FUR (EIO x % a.i x Use Rate).

EIO FUR

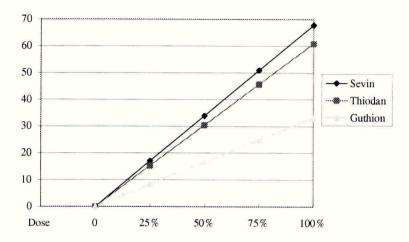


Figure 2. Pollution taxes and external damage from differing EIO rated pesticides.

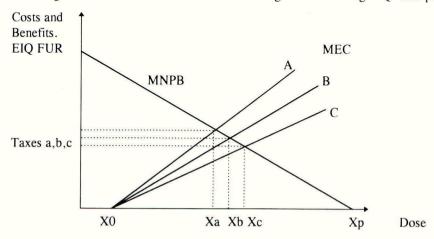


Figure 2 represents a simplified model of the costs and benefits associated with pesticide use. MEC is assumed to be proportional to the inputs used, MNPB represents the benefits from

spraying and for any input below X0 the waste is assimilated by the environment. The MNPB is the extra benefit farmers receive from changing their level of pesticide use by one unit. Thus, excluding the MEC the rational farmer will operate at emission level Xp, where MNPB = 0. The MEC is the extra cost imposed on society by using one extra unit of pesticide. The economic optimal level of emissions is where MNPB = MEC, or where the two curves bisect each other. Imposing a tax (Tax a,b,c) on farmers will force them to move from point Xp to one of Xa, Xb or Xc thus reducing EIQ FUR values, emissions and damage. The three MEC curves actually represent either three different pesticides, A, B, C, or three different pest management strategies, based on EIQ values from the Kovach et al., (1992) model. Pesticide A is the most harmful, pesticide B the next and pesticide C the least harmful. In order that the EIQ FUR values be used as MEC curves then the monetary value of the damage caused by the individual pesticide would have to be calculated. This is perhaps why the standard is the most realistic option at present.

Assuming that each of these pesticides target the same pests and are at least as efficacious as each other Tax a, will either force the user to move to a less harmful pesticide or reduce inputs. Even if pesticide B were less efficacious than A, but the tax was greater than the expected losses due to greater pest or disease susceptibility, then the farmer would still change to pesticide B. If, however, the value of losses were expected to exceed the tax level, then there would be no incentive to change. It is possible to see from Figure 2 that of the three pesticides (or pest management strategies) A, B, C, because potential damage from pesticide A is the greatest, the tax on this pesticide must be higher, and the reduction in emission, or EIQ FUR, greater than B and C to be optimal. Similarly, because pesticide C is potentially the least harmful the tax is lower (or indeed may be zero) than for B and A, and the required reduction in emission less than for B and A to be optimal (Figure 2.). Thus for farmers and society as a whole B is preferable to A, and C is preferable to both A and B. Again, however, this is assuming that the three pesticides are at least as efficacious as each other.

Referring to Figure 2 once again, a standard could be set that was equal to Xb, this also represents a given EIQ FUR, of say 200, at a use rate of 5 kg per hectare, for example. Thus, following the path from Xb to Tax b (or now EIQ FUR b) pesticide B satisfies the desired standard. At use rate Xb pesticide C also satisfies the standard, whereas pesticide A exceeds the standard. Thus, the farmer can either operate with pesticide A at a level to the left of Xa, or switch to pesticides B or C. Tax b indicates the taxation level required to ensure that use of pesticide B is at the rate Xb. The EIQ model of Kovach et al., (1992) is potentially well suited to supporting a programme of environmental protection based on standards, indeed as a possible IPM accreditation tool standards would need to be explicitly stated.

EIQ's AND SUBSIDIES

The outcome for the individual organisation with regards to taxes and subsidies should be identical. If a firm expands output it foregoes a subsidy which could be obtained through pollution reduction, which is the same as incurring a tax. So the incentive is there to either reduce the use of a particular input, or switch to an alternative. The EIQ supports this solution in two ways. Firstly it informs policy makers, and hence farmers, of the acceptable FUR (thus use rate), and secondly it informs farmers of less harmful alternatives.

In the long run, however, subsidies cause additional problems with regard to pollution and the environment. Subsidies generally make firms more profitable, whereas taxes make firms less profitable. This alters the exit-entry conditions of the industry. Taxes reduce emission for individuals, and force individuals out of the industry. Subsidies reduce emissions for individuals but encourage additional firms to enter the industry thus increasing the overall number of polluters. Potentially, therefore, subsidies will result in increased environmental degradation.

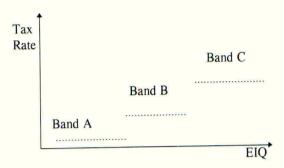
STRENGTHS AND WEAKNESSES OF PESTICIDE INDICES IN THE APPLICATION OF TAXES TO PESTICIDES

The potential weaknesses associated with the whole range of the available pesticide ranking indices are manifold and include mathematical problems associated with weighting and manipulating variables, data gaps in the input requirements and possibly unrealistic linear dose response curves. On the positive side pesticide ranking indices may fill a gap in the information requirements of decision makers at all levels and could be useful monitoring and evaluation tools for policy initiatives. In addition they have the potential to form the basis of IPM accreditation schemes, and to operate at a range of spatial scales and across sectors. Finally, and importantly from the perspective of the current analysis, if the models become acceptable to all involved in pesticide use, their outputs do lend themselves to tax and charge design.

Although the idea of taxing pesticides is not new several specific problems associated with the adoption of ranking models to set taxes do need to be considered. For example, it is unclear whether or not the tax should be applied linearly with the EIQ (as in Figures 1 and 2) or should a step threshold be utilised (see Figure 3), with pesticides split into "impact bands". The latter is easy to imagine as three or four bands of pesticides could be defined with the lowest EIQ pesticides attracting zero tax and pesticides in the other bands attracting increasing amounts of tax. Setting these threshold, or standard levels, however, would be a sensitive policy issue, and much thought would have to be given to this area. Also, the instrument to enforce such standards would also be controversial. Should such standards be voluntary, enforced by taxes, or enforced by subsidies? Further, if no equivalent pesticide was available, should any tax be levied at all? One of the rationales of imposing pesticide taxes would be to encourage users to move to a low damage option, but if none exist, then the idea of a tax may be invalid.

In addition to this there are other concerns with the use of economic instruments in environmental protection. Who will pay for, administer and collect such a taxation scheme? Should groups of compounds be treated within one tax scheme? If this is the case then generally insecticides will carry a higher EIQ FUR, and thus a higher tax, than herbicides. Is this desirable? Finally it is unclear if a tax should differentiate between low doses of harmful pesticides and high doses of relatively harmless pesticides. These are just some of the important issues that would need to be addressed prior to the implementation of a taxation scheme for pesticides.

Figure 3. A stepped function for the taxation of pesticides.



CONCLUSIONS

Models which aim to rank pesticides by environmental impact are still in their infancy. There is currently, however, a policy drive towards the development of such models at an international level (OECD, 1997). If policy makers, farmers, consumers and pesticide industry representatives agree on one particular methodology, the integrity of the data input, and form of the output, then such models provide much of the information required by environmental economists in the design of emission tax, product tax and subsidy programmes.

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SESSION 6B WEED ECOPHYSIOLOGY

Session Organiser DR R P FINCH

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Poster Papers 6B-1 to 6B-6

PHENOLOGY AND REPRODUCTIVE ALLOCATION OF BROMUS STERILIS, B. DIANDRUS, B. HORDEACEUS AND B. COMMUTATUS

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ABSTRACT

Time to anthesis and reproductive allocation were assessed in a spaced plant trial for 25 populations of *Bromus sterilis*, *B. diandrus*, *B. hordeaceus* and *B. commutatus*. Difference in time to anthesis between slowest and fastest populations was 30 days. Southern populations allocated more resources to reproductive weight than northern populations. Seed number per plant ranged from 1196 to 9377, seed number per panicle from 60 to 393 and TSW from 2.57 to 15.16g. Multivariate analysis distinguished between populations, based on seeds per panicle, seeds per plant and time to anthesis. Canonical Variate Analysis also separated northern and southern populations.

INTRODUCTION

Differences in morphology and phenology may be due to local adaptations through genetic changes and plastic responses to different environmental conditions. *Bromus* spp. have been introduced to a range of climates and habitats from their native range and occur as weeds of cereal crops in many countries. *B. diandrus* shows genetic variation in seed dormancy and rate of plant development at different sites in southern Australia (Gill & Blacklow, 1985). *B. sterilis* shows a high degree of phenotypic plasticity which might have prevented genetic differentiation (Theaker *et al.*, 1995). Growing plants in a common environment and assessing morphological and phenological attributes in a spaced plant trial provides a useful indication of the genetic basis for these characters. The aim of this experiment was to investigate differences in time to anthesis, seed output and seed weight in *B. sterilis*, *B. diandrus*, *B. hordeaceus* and *B. commutatus* from a range of geographic locations.

MATERIALS AND METHODS

A spaced plant trial was set up within the experimental grounds of the Plant Sciences Laboratories at the University of Reading on 28/10/93. Seeds from 25 populations of four *Bromus* spp. (Table 1) were sown in three fully randomised blocks at a spacing of 1.5m between individual sowing points within the blocks and 2m between blocks. Each of the blocks measured 9x10 m and contained one replicate for each population. Initially, excess seeds were sown but subsequently thinned to one plant per sowing point. At various stages during development, plants were assessed for particular morphological and phenological attributes. Shoot dry weight per plant (Ws) and reproductive dry weight per plant (Wr) were determined and expressed as a Wr/Ws ratio. This paper reports results on time to anthesis, resource allocation, seed number and thousand seed weight (TSW). Univariate analysis was carried out for different attributes, and provenance

(northern or southern) was included as an additional factor. For the multivariate analysis, Canonical Variate Analysis (CVA) was used.

Table 1. List of *Bromus* accessions (ⁿ=northern accession, ^s=southern accession, ^{*}=ruderal, ^o=arable)

Number	Population	Site information
1	B. sterilis	England, Cambridgeshire, arable ^{n*}
2	B. sterilis	England, Cambridgeshire, hedgerow
3	B. sterilis	England, Cambridgeshire, untreated plot ^{no}
4	B. sterilis	England, Cambridgeshire, field I
5	B. sterilis	England, Essex, IPU treated plotn*
6	B. sterilis	England, Essex, untreated plot no
7	B. sterilis	England, Cambridgeshire, field II n*
8	B. sterilis	Italy, Legnago-Padova, ruderal so
9	B. sterilis	Germany, Braunschweig, ruderal no
10	B. sterilis	Greece, Macedonia, sprayed s*
11	B. sterilis	France, Rennes, sprayed "
12	B. sterilis	Israel, Rehovot, non-cultivated field so
13	B. sterilis	Hungary, Keszthely, ruderal n*
14	B. diandrus	S-Australia, Adelaide, arable s
15	B. diandrus	Spain, Madrid, arable s*
16	B. diandrus	Australia, Victoria, ruderal so
17	B. diandrus	New Zealand, Canterbury, ruderal so
18	B. diandrus	France, Montpellier, arable n*
19	B. diandrus	Israel, Rehovot, non-cultivated field so
20	B. diandrus	W-Australia, Chapman Res. Stn., ruderal ^{so}
21	B. commutatus	Sweden, Brinklosta, ruderal no
22	B. commutatus	Germany, Göttingen, ruderal no
23	B. hordeaceus	New Zealand, Canterbury, ruderal so
24	B. hordeaceus	Sweden, Öland, ruderal no
25	B. hordeaceus	France, Rennes, arable n*

RESULTS

Time to anthesis

The majority of accessions required 190-200 days from seedling emergence to anthesis. The fastest time to anthesis was observed for *B. diandrus* from France and Australia (populations 18, 16), and *B. sterilis* from Israel and Germany (populations 9, 12). These required 30 days less to reach anthesis compared with the slowest accessions (Fig. 1). B. hordeaceus from New

Zealand (population 23) and *B. commutatus* from Sweden (population 21) took the longest time to reach anthesis (210 days after seedling emergence).

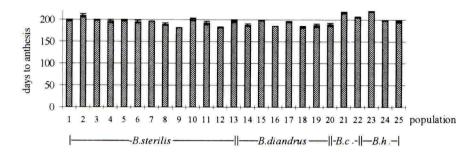


Fig. 1. Days from seedling emergence to anthesis for 25 *Bromus* accessions (vertical bars are \pm standard error; *B.c.=B. commutatus*; *B.h.=B. hordeaceus*).

Resource allocation

B. sterilis from Israel (population 12) exhibited the highest Wr/Ws ratio, followed by B. hordeaceus from France (population 25) and B. diandrus from Israel and Australia (population 16), with similar resource allocation. The Wr/Ws ranged from 10.03 for population 12 to 0.85 for population 9. The latter population was the only one where vegetative weight exceeded reproductive weight (Table 2). Southern populations tended to show a higher allocation to the reproductive weight (Wr:Ws(g), $8.14\pm1.0:4.83\pm1.2$) whereas northern populations allocated their resources more towards vegetative weight (Wr:Ws(g), $4.95\pm0.7:9.60\pm0.9$).

Table 2. Vegetative/reproductive ratio of 25 *Bromus* accessions (populations with the same letter are not significantly different).

Population	Wr/Ws	Location	Habitat	Population	Wr/Ws	Location	Habitat
12	10.03 ^a	southern	arable	5	2.30 ^{bcd}	northern	arable
25	5.66 ^b	northern	ruderal	22	2.29 ^{bcd}	northern	ruderal
19	5.64 ^b	southern	arable	6	2.25 ^{bcd}	northern	ruderal
16	5.36 ^b	southern	ruderal	1	2.24 ^{bcd}	northern	arable
20	4.83 ^{bc}	southern	ruderal	3	2.21 ^{bcd}	northern	arable
24	4.23 ^{bcd}	northern	ruderal	21	2.13 ^{bcd}	northern	ruderal
11	4.11 ^{bcd}	northern	ruderal	4	1.95 ^{bcd}	northern	arable
15	3.27 ^{bcd}	southern	ruderal	10	1.88 ^{bcd}	southern	arable
2	3.19 ^{bcd}	northern	ruderal	13	1.49 ^{cd}	northern	arable
14	2.96 ^{bcd}	southern	arable	23	1.38 ^{cd}	southern	ruderal
8	2.84 ^{bcd}	southern	ruderal	18	1.24 ^{cd}	northern	arable
7	2.75 ^{bcd}	northern	arable	9	0.85 ^d	northern	ruderal
17	2.40 ^{bcd}	southern	arable				

Seed output and TSW

Seed weight was greatest for *B. diandrus* accessions with a TSW above 10g. For *B. sterilis* accessions from England, the TSW averaged approximately 8g. Particularly low seed weights occurred in *B. commutatus* and *B. hordeaceus* and in two accessions of *B. sterilis* (populations 12 and 13).

The highest numbers of seeds per plant and seeds per panicle were recorded for *B. hordeaceus* (population 23) and *B. commutatus* (populations 21, 22) and these populations also had a low TSW. In spite of similar TSWs between British accessions, one ruderal accession (population 2) exhibited a higher number of seeds per plant. Population 2 also had the highest seed number per panicle for the British accessions, whereas the average was 100 seeds per panicle for other British accessions. Some *B. diandrus* accessions had a lower seed number per plant but not a lower seed number per panicle. A higher TSW in *B. diandrus* accessions corresponded with a lower seed number per plant (Table 3).

Table 3. Mean seed number per plant, mean seed number per panicle and mean seed weight of 25 *Bromus* accessions (figures are means \pm standard error).

Population	No. seeds per plant	No. seeds per panicle	TSW (1000 seed wt. g)
1	2210±239	108±08	8.24
2	5303±365	179±83	7.21
3	2550±462	84±16	8.20
	2467±421	84±10	7.12
4 5	3126±520	128±25	7.52
6	2938±453	104±26	7.96
7	3139±308	96±04	8.24
8	1535±322	105±57	9.07
9	1762±152	60±10	8.85
10	3102±332	107±10	7.32
11	2753±275	102±23	14.87
12	3415±149	196±36	4.81
13	2760±135	123±34	3.08
14	1196±138	109±27	11.13
15	1774±152	100±31	14.87
16	2729± 96	134±09	12.85
17	1582±241	88±36	15.12
18	2203±444	59±34	12.00
19	1592±183	81±09	15.16
20	1841±256	96±11	11.76
21	8337±707	261±59	3.56
22	8340±191	358±82	3.68
23	9901±744	190±04	3.00
24	3403±267	320±83	3.04
25	9377±629	393±46	2.57

Multivariate Analysis

The first canonical variate (CV I) accounted for 53% of the total variation, and the first three canonical variates together accounted for 79% of the variation. CV II and CV III clearly separated the *B. commutatus* accessions, *B. hordeaceus* from France and *B. hordeaceus* from New Zealand. The populations from Britain were fairly close together (1, 3, 4, 7), along with *B. sterilis* from France and *B. diandrus* from New Zealand. CV III separated the *B. sterilis* populations 2, 5 and 6 from the remaining British accessions. *B. diandrus* accessions were fairly close together, except for population 16 from Australia. Differences in CV I were largely due to differences in seed number per plant, days to flowering and seed number per panicle whereas differences in CV II were mainly attributable to differences in panicle weight. Differences in CV III were largely due to differences in days to seedling emergence. Canonical Variate Analysis clearly separated northern and southern accessions. Differences in CV I - IV were largely due to differences in number of tillers (vegetative, reproductive and total) at the preliminary harvest and also to seed and panicle number per plant.

DISCUSSION

Southern populations tended to have a greater allocation to reproductive weight and also reached anthesis earlier. This is considered an adaptation to a shorter growing season and a less predictable environment. Northern populations of slender wheatgrass tended to have a higher proportion of vegetative growth than populations from southern areas (Pringle et al., 1975). B. hordeaceus, and B. commutatus needed longer to reach anthesis than the two other brome species. Hulbert (1955) reported that Bromus accessions from various locations varied by about three weeks in time to maturity, in the order B. sterilis, B.tectorum, B.hordeaceus, B. commutatus. B. commutatus and B. hordeaceus produced a large number of small seeds whereas B. diandrus produced a smaller number of large seeds. The potential of Bromus spp. to produce large seed numbers may confer a competitive advantage during crop establishment (Kon & Blacklow, 1988).

Using multivariate analysis it was possible to discriminate between populations, mainly based on differences in reproductive and phenological attributes, including time to anthesis and seed number per plant. Genetic differentiation exists if plants of the same species from different origins have different phenotypes in the same environment. *Bromus* spp. exhibit genetically determined variation but are also characterised by phenotypic plasticity.

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GERMINATION ECOLOGY OF STELLARIA MEDIA

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ABSTRACT

Seeds from 25 UK populations of *Stellaria media* were collected from arable field sites and grown for a generation in a common environment. Using second generation seeds, significant differences were recorded in germination and seedling growth statistics between populations.

INTRODUCTION

A major objective in ecological research is to explain and predict the distribution and abundance of organisms. A major objective in agronomic research is to control the distribution and abundance of pest organisms, such as weeds, in farming systems. In order to control weeds more effectively, it is clear that agronomists would benefit from a better understanding of weed biology and ecology. For many weed species the greatest uncertainty is associated with the factors that affect germination and seedling establishment. A criticism of most studies of germination ecology is their failure to study more than one population, ignoring the differences that can exist between populations. For example, van der Vegte (1978) showed that populations of *Stellaria media* from the same field differed markedly in the temperature range over which freshly collected seeds would germinate and whilst one population built up a relatively uniform short-lived seedbank, the other built up a persistent seedbank of phenotypically diverse seeds.

This paper addresses the germination ecology of *S. media*, studying inter-population differences and controlling for the effects of the maternal environment. The results presented were derived from a large scale screening of 25 UK populations. The aim was to assess the magnitude of differences between populations in both germination and seedling establishment in order to select contrasting populations for more detailed study.

MATERIALS AND METHODS

Over the summer of 1995, 25 seed populations were collected from arable sites in the UK. Seeds were collected from a large number of plants at each site and were allowed to dry at room temperature for ca. 2 weeks prior to storage in paper envelopes in a dry incubator maintained at 10 °C. In October 1995, seed populations were sown in replicated pots in a

greenhouse for seed multiplication in a common environment (in order to control for maternal effects related to climatic differences between sites and differences related to time of collection). On first flowering, pots were covered in a layer of muslin to prevent cross fertilisation between populations. Mature seeds were collected over a period of 6-8 months, dried and stored as described above. Seeds collected at different times were stored separately and only those collected in August 1996 were used in the screening experiment.

Screening for differences in germination characteristics

Starting in November 1996, the germination characteristics of the 25 populations were assessed in 2 FISONS controlled environment cabinets, each maintained with a 14 hr day coinciding with the higher of the alternating temperatures (W = 20/15 °C; C = 10/5 °C). The irradiance during the day varied between a photosynthetic photon flux of 160 to 205 µmol m² s¹ (Campbell CR10). To counter the lack of replication of the controlled environments, replicates were repeated at ca. 10 day intervals. One hundred seeds were counted, weighed and placed on two layers of Whatman Grade 181 paper (9cm discs) placed in a 9 cm Petri-dish. The papers were then moistened with 10 ml distilled water and placed in a randomised block on one shelf in the cabinet. Three replicates were used in each of the two cabinets. The seeds were checked after 24 h, then three times daily for the next four days, (coinciding with peak of germination) followed by further daily checks for another seven days. The Petri-dishes were inspected in the same order on each occasion and seeds that had germinated (as defined by radical emergence) were counted and removed. The cumulated count data from each Petri-dish were fitted to Gompertz distributions and the parameters were tested for differences between population x environment by analysis of variance.

Screening for differences in seedling growth characteristics

A subset of the seeds germinated in the warm cabinet were transferred to a growth room maintained with the same temperature and day length conditions (14 hr day; 20/15°C). Irradiance varied between 75 to 90 µmol⁻¹ m⁻² s⁻¹. A minimum of 40 germinated seeds per population (except Boxworth) were transferred and sown individually into 30 mm diameter cells in 10 x 15 cell-trays filled with John Innes no. 2 compost. Seed diameter was measured to the nearest 0.05 mm at sowing. Seedling height was measured at emergence (cotyledons expanded) and at two leaves, and the time taken from sowing to each stage for each individual was recorded. For each population, random samples of a third of the established seedlings were harvested at emergence, two leaves and four leaves. The dry weights of these harvested seedlings were recorded and relative growth rates calculated by subtracting the mean population seed weight from each seedling dry weight and dividing by the difference in days to emergence and days to reach a particular growth stage. In order to select contrasting populations for further study the combined germination and seedling growth data was entered into a furthest neighbour cluster analysis.

RESULTS

Table 1 shows the significant differences between populations for mean 100-seed-weight and seed diameter. The largest seeds were from Caithness (population 25); the smallest from Rosemaund (population 10).

Table 1. Mean population 100-seed-weight (100wt) and seed diameter (sd) for 25 UK populations of *S. media* (seeds harvested from a common environment).

Population	sd	100wt	Pop	oulation	sd	100wt
•	(mm)	(g)			(mm)	(g)
1. Bridgets (1), Hamps.	1.27	0.0486	14.	High Mowthorpe, N. Yorks.	1.19	0.0473
2. Bridgets (2), Hamps.	1.19	0.0449	15.	St Boswells, Borders	1.31	0.0545
Wye, Kent	1.19	0.0488	16.	Lanark, Lanarkshire	1.22	0.0464
 Rothamsted, Herts. 	1.16	0.0419	17.	Balerno, Lothian	1.31	0.0561
Boxworth, Cambs.	1.17	0.0459	18.	Eaglescarnie, Lothian	1.30	0.0531
Arthur Rickwood, Cambs.	1.19	0.0438	19.	Bush, Lothian	1.23	0.0466
7. Loddington, Leics.	1.12	0.0456	20.	Balmalcolm, Fife	1.21	0.0443
Morley, Norfolk	1.22	0.0473	21.	Carnoustie, Angus	1.18	0.0534
9. Rosemaund (1), Herefords.	1.14	0.0419	22.	Crieff, Stirlingshire	1.22	0.0500
10. Rosemaund (2), Herefords.	1.10	0.0383	23.	Tofthill, Perthshire	1.14	0.0392
11. Wellesbourne, Warwicks.	1.16	0.0427	24.	Griminish, Benbecula	1.31	0.0573
12. Gleadthorpe, Notts.	1.21	0.0420	25.	Stanstill, Caithness	1.43	0.0818
13. Thelwell, Cheshire	1.14	0.0515				
LSD $(p > 0.01)$	0.09	0.0025			0.09	0,0025
[df]	[985]	[125]			[985]	[125]

Table 2 shows the population parameters derived from fitting the Gompertz distribution to the cumulated counts of germinated seeds. The parameter b is related to the slope of the curve and larger values of b are associated with greater synchronicity of germination. The parameter mequals the time from the start of the experiment (X) at the point of inflexion in the curve, and the sum of parameters c and a is the asymptote of the fitted curve, representing the total number of seeds germinated for each population. Also presented is the mean time to 50% germination (150), calculated numerically from the Gompertz equation. The test temperature had significant effects on all four parameters and the value of t_{50} . Germination was less synchronous (except for Gleadthorpe) and a smaller proportion of seeds germinated (except for Boxworth) in the colder regime. In both temperature regimes there were significant differences between populations for the parameters b, m, c, (c+a) and for the calculated value for t_{50} . With the exception of parameter b, there were no significant interactions between population and temperature. In the warm temperature regime, germination was most synchronous for Loddington and Crieff and least for Caithness and Gleadthorpe. In the colder temperature regime, germination was most synchronous for Bridgets (population 1) and least for the population from Thelwell. This synchronicity of germination was not significantly correlated with the proportion of seeds that germinated in each population in each of the two -0.09; r_[10/50C, 73] = -0.05). Similar patterns in the temperature regimes (r 120/150c 731 = proportion of seed germinated occurred in both temperature regimes with the most seeds germinated from Perthshire, the least from Boxworth, Wye and Thelwell.

Table 3 shows the mean population growth statistics. With the exception of time from sowing (germination) to emergence (t1) and to two leaves (t2), there were significant differences between populations for each statistic. The population from Caithness produced seedlings with a consistently higher dry weight at each growth stage. These were also the tallest seedlings at two leaves and exhibited the fastest growth rate between emergence and two leaves. At the other extreme, the population from Bridgets (population 2) tended to produce small seedlings with low dry weight and slow growth rates.

Table 2. Mean parameters from fitting the Gompertz $(Y=a+c(exp(-exp(-b(X-m))))+\epsilon)$ distribution to cumulated counts of germinated seeds for 25 UK populations of S. media (populations coded as for Table 1), n=3.

- 2			10/5 °C	.50					
				1318.					P
									1
									2
					2 200	34.03	52.95	0.1300	3
				0 E 19 E		25.26	58.82	0.1000	4
				69.30	-0.113	8.07	64.74	0.0900	5
		121.74	0.0500	58.93	-0.955	66.52	55.83	0.1250	6
		126.81	0.0533	59.76	-0.420	34.12	56.82	0.1333	7
	30.27	111.14	0.0600	58.69	-0.873	47.03	54.58	0.0967	8
0.127	45.48	104.01	0.0767	58.42	-0.870	51.34	54.92	0.1133	9
-0.003	28.14	123.97	0.0467	66.71	-0.377	33.51	61.20	0.0700	10
-0.113	21.18	116.16	0.0500	60.76	-0.700	37.36	57.42	0.1200	11
-0.010	35.91	117.45	0.0767	64.60	-0.630	53.31	60.39	0.0667	12
-0.167	14.35	121.75	0.0300	58.75	-0.467	24.11	54.84	0.1067	13
-0.140	22.49	121.32	0.0433	62.09	-0.363	27.57	57.90	0.0967	14
0.110	53.59	111.88	0.0467	58.09	-1.450	58.21	52.76	0.0767	15
0.413	36.25	128.31	0.0400	68.02	-0.227	50.77	63.41	0.0833	16
0.023	50.00	105.86	0.0533	58.72	-1.507	57.96	53.60	0.0800	17
0.047	49.65	117.56	0.0567	59.20	-1.577	70.46	54.54	0.0867	18
-0.140	44.66	98.56	0.0533	52.54	-0.210	48.36	48.88	0.1067	19
0.223	36.04	111.62	0.0600	59.37	-0.893	52.55	55.51	0.1067	20
-0.003	44.45	113.34	0.0600	60.67	-0.750	54.03	57.08		21
0.137	28.87	99.73	0.0733	62.59	2.597	39.28	60.53	0.1333	22
0.247	79.62	137.14	0.0333	70.90	1.437	81.00	66.58	0.0867	23
0.187	23.54	114.27	0.0567	58.51	-0.757	29.53	54.87	0.1167	24
0.043	22.64	115.92	0.0367	71.07	-0.163	30.33	65.45	0.0667	25
	-0.113 -0.010 -0.167 -0.140 0.110 0.413 0.023 0.047 -0.140 0.223 -0.003 0.137 0.247 0.187	17.70 -0.020 29.29 -0.067 17.48 0.017 24.19 0.083 11.29 -0.027 51.08 -0.203 31.29 -0.087 45.48 0.127 28.14 -0.003 21.18 -0.113 35.91 -0.010 14.35 -0.167 22.49 -0.140 53.59 0.110 36.25 0.413 50.00 0.023 49.65 0.047 44.66 -0.140 36.04 0.223 44.45 -0.003 28.87 0.137 79.62 0.247 23.54 0.187	97.15 17.70 -0.020 111.06 29.29 -0.067 126.30 17.48 0.017 117.08 24.19 0.083 137.43 11.29 -0.027 121.74 51.08 -0.203 126.81 31.29 -0.083 111.14 30.27 0.087 104.01 45.48 0.127 123.97 28.14 -0.003 116.16 21.18 -0.113 117.45 35.91 -0.010 121.75 14.35 -0.167 121.32 22.49 -0.140 111.88 53.59 0.110 128.31 36.25 0.413 105.86 50.00 0.023 117.56 49.65 0.047 98.56 44.66 -0.140 111.62 36.04 0.223 113.34 44.45 -0.003 99.73 28.87 0.137 137.14 79.62 0.247 <td>$\begin{array}{c ccccccccccccccccccccccccccccccccccc$</td> <td>t50 b m c a 61.25 0.0867 97.15 17.70 -0.020 55.13 0.0700 111.06 29.29 -0.067 56.10 0.0467 126.30 17.48 0.017 63.03 0.0733 117.08 24.19 0.083 69.30 0.0433 137.43 11.29 -0.027 58.93 0.0500 121.74 51.08 -0.203 59.76 0.0533 126.81 31.29 -0.083 58.69 0.0600 111.14 30.27 0.087 58.42 0.0767 104.01 45.48 0.127 66.71 0.0467 123.97 28.14 -0.003 60.76 0.0500 116.16 21.18 -0.113 64.60 0.0767 117.45 35.91 -0.010 58.75 0.0300 121.75 14.35 -0.167 62.09 0.0443 121.32 22.49 -0.140</td> <td>a 150 b m c a -0.073 61.25 0.0867 97.15 17.70 -0.020 -1.183 55.13 0.0700 111.06 29.29 -0.067 -0.943 56.10 0.0467 126.30 17.48 0.017 -0.463 63.03 0.0733 117.08 24.19 0.083 -0.113 69.30 0.0433 137.43 11.29 -0.027 -0.955 58.93 0.0500 121.74 51.08 -0.203 -0.420 59.76 0.0533 126.81 31.29 -0.083 -0.873 58.69 0.0600 111.14 30.27 0.087 -0.870 58.42 0.0767 104.01 45.48 0.127 -0.377 66.71 0.0467 123.97 28.14 -0.003 -0.467 58.75 0.0300 121.75 14.35 -0.167 -0.363 62.09 0.0433 121.32 22.49 -0.1</td> <td>c a t50 b m c a 29.91 -0.073 61.25 0.0867 97.15 17.70 -0.020 41.61 -1.183 55.13 0.0700 111.06 29.29 -0.067 34.03 -0.943 56.10 0.0467 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	b	m	C	а	(c+a)	t50
SED [population]	0.011	6.487	4.03	0.57	4.07	6.88
SED [population]	0.003	1.84	1.14	0.16	1.15	1.95
SED [temperature] SED [population x temperature]	0.016 -	9.17	5.69	0.81	5.75	9.72

Cluster analysis (results not presented) of the combined germination and seedling growth statistics identified Caithness and Perthshire as distinctive populations and suggested that Boxworth and Bridgets would merit further study. Average characteristics were represented by Balerno, Carnoustie and Loddington.

DISCUSSION

The effect of temperature on the proportion of seeds germinating and the synchronicity of germination mirrors the pattern of autumn and spring flushes of *S. media* emergence in the field. Colder winter temperatures do not completely inhibit seedling emergence, but fewer seedlings establish with little synchronicity in emergence. Boxworth was the only population for which more seeds germinated in the colder temperature regime and Gleadthorpe was the only population for which the synchronicity of germination was greater in the cold. The total Boxworth germination was low and as such the significantly greater cold germination is

Table 3. Mean population seedling statistics for height at emergence (h1) and at two leaves (h2); for dry wt. at emergence (d1), at two leaves (d2) and at four leaves (d4); for time from sowing (germination) to emergence (t1), to 2 leaves (t2) and to four leaves (t4) and for mean seed wt. for population x) to two leaves (d2gr) and to four leaves (d4gr) for 25 UK populations of S. media (populations coded as relative growth rates calculated for height gain from emergence to two leaves (hgr) and for dry wt. gain from emergence (assuming d1 = for Table 1), n > 10. * = missing value.

	f																								1	
d4gr (mg d ⁻¹)	0.2406	0.2043	0.3836	0.3207	*	0.3654	0.3285	0.3574	0.3450	0.3429	0.3509	0.2643	0.3052	0.2424	0.2958	0.3799	0.3178	0.3163	0.3691	0.2974	0.2988	0.2939	0.2863	0.3467	0.3468	*** [24, 245]
d2gr (mg d ⁻¹)	0.1978	0.1257	0.1320	0.1614	*	0.1331	0.1268	0.1966	0.1640	0.1924	0.1994	0.1770	0.1618	0.0713	0.1535	0.1907	0.1490	0.1846	0.1721	0.1764	0.1839	0.1471	0.1451	0.1625	0.2232	*** [23, 242]
hgr (mm d ⁻¹)	2.37	2.46	2.33	2.29	2.45	2.11	2.09	2.30	2.27	2.01	3.14	1.78	1.98	2.20	1.72	2.71	2.49	2.71	1.97	2.32	2.38	2.71	2.00	2.18	2.97	*** [24, 357]
t4 (days)	17.50	18.71	16.87	15.00	14.00	15.83	15.00	15.50	14.25	14.33	13.67	16.00	13.84	16.37	14.74	15.27	14.31	16.63	15.23	16.71	15.71	16.00	14.14	14.82	15.11	*** [24, 253]
t2 (days)	13.39	13.79	13.25	11.67	11.78	12.55	11.96	11.21	12.20	11.50	11.71	12.48	11.42	12.32	11.96	12.21	11.87	12.38	12,30	13.70	12.55	12.43	12.58	12.29	11.25	[24, 530]
t1 (days)	82.9	7.18	7.21	5.10	4.67	5.48	5.57	4.88	5.90	4.45	6.14	5.80	6.70	6.26	5.67	5.71	5.84	00.9	6.18	6.87	5.97	6.92	6.21	6.93	5.47	[24, 789]
d4 (mg)	3.0040	2.7070	3.9900	3.4580	3.6290	3.9400	3.5090	3.8780	3.8630	3.6650	3.7160	3.0130	3.4520	2.8080	3.4320	3.9120	3.3690	3.5450	3,8910	3.2450	3,4140	3.7680	3.0990	3.8610	4.1470	*** [24, 254]
d2 (mg)	1.8540	1.3980	1.3600	1,4450	*	1.2730	1.5190	1.7390	1.5160	1.6630	1.6890	1.4600	1.4130	1.4860	1.5140	1.6590	1.5290	1.6500	1.6750	1,6980	1,7240	1,4910	1.3520	1.6110	2.2430	*** [23, 243]
d1 (mg)	0.4644	0.3959	0.4570	0.5400	*	0.5333	0.4035	0.4975	0.5640	0.4026	0.3418	0.4215	0.4562	0.5100	0.5614	0.6289	0.5064	0.5663	0.6216	0.5225	0.6200	0.5471	0.3886	0.4904	0.7380	*** [23, 192]
h2 (mm)	35.39	32.40	37.02	33.21	37.19	29.75	34.37	36.82	36.10	31.01	36.82	30.98	31.54	34.85	28.40	37.66	35.74	36.65	30.89	33.74	35.98	37.67	28.74	29.59	42.62	*** [24, 429]
h1 (mm)	20.89	18.97	24.48	19.23	19.23	17.02	19.84	20.80	20.07	18.63	17.94	19.81	19.85	21.42	18.29	25.31	19.61	21.25	19.57	19.61	22.51	22.03	16.38	17.69	21.96	*** [24, 680]
Pop'n	_	7	3	4	S	9	7	∞	6	10	Ξ	12	13	=	15	91	17	81	16	20	21	22	23	24	25	$F_{ m [aff]}$

unlikely to result in greater weed problems in the field. The low Gleadthorpe synchronicity in the warm temperature regime is likely explained by a tendency for a double peak in germination. This characteristic could lead to greater weed problems in the field as a result of weed control measures targeting the initial population of seedlings germinated, allowing the later seedlings to escape. However over the timescale involved (< 14 days) this is unlikely to cause a problem. Of greater concern is the large number of seeds left ungerminated in many of the populations surveyed. Tetrazolium tests (methods described in IBPGR, 1985) showed that these remaining seeds were > 95% viable and as such the factors required to promote germination in these seeds requires further investigation.

Previously quoted mean seed weights have varied from 0.35 mg (Grime et al., 1988) to 0.67 ± 0.04 mg (Sobey, 1981). This range encompasses all of the mean seed weights recorded in the study, except for the population from Caithness which was clearly larger than those previously recorded. The Caithness population also produced the largest and heaviest seedlings and some of the highest growth rates. For other populations the relationship between mean seed size, weight and seedling growth statistics was less pronounced, although (except Perthshire) there was perhaps a tendency for the Scottish populations to produce larger heavier seeds and seedlings at emergence. A reciprocal transplant experiment would test whether this difference was the result of local adaptation to the Scottish common environment. Grime et al. (1988) quote seedling growth rates ranging between 0.246 and 0.343 mg day⁻¹ for the five weeks from emergence. These values are comparable to the values calculated for seedling growth from emergence to four-leaves. The values calculated for seedling growth from emergence to two leaves tend to be smaller than this and further investigation is required to ascertain whether these growth rates are actually slower or whether this is a result of greater inaccuracy in the smaller measurements made at this growth stage.

The significant variation between the different populations of *S. media* in germination and seedling growth can be attributed to genetic differences between populations. This is likely to result from complex interactions between the maternal and test environments and the genetic structure of the populations. Different responses would be expected if the seeds had been produced in a different maternal environment or if the seeds had been tested under different conditions. The magnitude of differences in germination and seedling growth characteristics observed in this study suggest that future studies aimed at describing the ecology of *S. media* need to consider the extent of inter-population variability.

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A MOLECULAR STUDY OF SPECIES-RICH GRASSLAND ECOSYSTEMS

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ABSTRACT

The underlying processes of vegetation dynamics are poorly understood in scientific terms mainly due to methodological limitations. The increased resolution of molecular technologies, however, can allow these processes to be dissected and, specifically, the genetic component of vegetation dynamics to be investigated at the population level. By utilising the polymerase chain reaction to study random amplified polymorphic DNA (RAPDs), we report here the initial results in a study to assess the inter- and intra-population levels of genetic diversity in species-rich grassland ecosystems.

INTRODUCTION

Vegetation management is a key component of land husbandry, utilising weed control strategies to control the dominance of selected species at the expense of unwanted species, and grazing control strategies to maintain the range and productivity of valuable species. Such management strategies attempt to modify vegetation dynamics, the continuous patterns of change which occur in a mixed plant community in response to the changing environment.

A greater understanding of the processes of vegetation dynamics, which can take a range of forms, particularly in species-rich vegetation where the options for change are especially great, would lead to the improved management strategies essential for increased productivity and conservation of biological diversity (Burrows, 1990; Hebren *et al.*, 1993; Randall, 1996). These goals are ecologically very important since such mixed plant communities constitute a habitat essential to a wide range of fauna.

Most studies of vegetation dynamics have been limited by shortcomings in the methodology available. The predominant technique used to examine levels of genetic diversity in weed populations has been isozyme analysis, but the levels of polymorphism disclosed by this approach can be too low for comprehensive analyses of diversity within and between populations (e.g. Warwick, 1987; Holt, 1994). However, over the last few years, molecular techniques have become increasingly important in complementing the more traditional biochemical or physiological approaches. They are now routinely used to investigate and characterise plant genetic diversity, plant population history and evolution and the classification of cultivars and varieties. Molecular approaches have been adopted to examine hybridisation, introgression and phylogentics of weed species such as *Helianthus annuus* and *Brassica nigra* L. (Palmer *et al.*, 1983; Quiros *et al.*, 1991; Reisberg, 1988). Indeed, there is an increasing number of examples of where molecular approaches have been applied to questions

of interest to weed scientists (Colosi and Schaal, 1992, 1994, 1997; Nissen et al., 1992; Richard et al., 1995; Meikle et al., 1995; Moodie et al., 1997).

In studies of genetic diversity, one of the most widely used molecular techniques has been Restriction Fragment Length Polymorphism (RFLP) analysis. However, this technique has a number of constraints when studying diversity at the population level since it requires relatively large amounts of DNA and is laborious and expensive when dealing with large sample numbers. An alternative technique which is now more widely used is that of Random Amplified Polymorphic DNA analysis (Welsh and McClelland, 1990; Williams *et al.*, 1990). This approach requires far less DNA than RFLP analysis, it requires no previous knowledge of the genome being analysed and is less laborious and less expensive for population studies.

To investigate the nature of the genotype-phenotype-environment interactions of vegetation dynamics in semi-natural habitats, a project has been initiated which aims to assess intraspecific genetic variation of three grassland species (*Agrostis capillaris*, *Festuca rubra* and *Rumex acetosa*). Using plants sampled from two distinct sites in Scotland, spatial and temporal genetic variation is being monitored over a five year period. In this paper we present initial results from *A. capillaris* plants sampled in 1995 although the materials and methods apply to all three species under study.

MATERIALS AND METHODS

Plant Material

A. capillaris plants were sampled from two distinct geographical locations in Scotland: Cleish in Fife, and Kirkton in West Perthshire. At each location, 11 loci were identified from each of five 50m transects spaced at 10m intervals in a single 40m x 50m plot. The plant closest to each sampling locus was removed and its position (distance and angle from the locus) recorded. Samples were then grown on in individual pots and retained for analysis.

DNA Extraction

DNA was extracted from leaf tissue using the Nucleon Phytopure DNA extraction kit (Nucleon Biosciences) following the manufacturers instructions. Approximately 0.1g of leaf material was ground to a fine powder in liquid nitrogen. The powder was then transferred to an ice-chilled Eppendorf tube and 700µl of extraction buffer added. After thorough mixing, the mixture was incubated at 65°C for 10min with regular agitation. The tube was placed on ice for 20min and 500µl of chloroform (-20°C) added. Nucleon Phytopure DNA Extraction Resin (100µl) was added and the tube repeatedly inverted at room temperature for 10min before centrifugation (1,300g, 10min). The upper, aqueous layer was transferred to a fresh Eppendorf tube and 450µl isopropanol (-20°C) added. After repeated invertion the tube was centrifuged (4,000g, 5min) to pellet the DNA. Following a brief wash in 70% ethanol and further centrifugation (4,000g, 5min) the pellet was allowed to dry thoroughly before being redissolved in sterile distilled water. DNA solutions were stored at -80°C.

RAPD Amplification

RAPD amplification was carried out in a 25µl reaction mixture containing 2ng genomic DNA, 0.5 Units DNA Polymerase (Amplitaq, Perkin Elmer), 2mM magnesium chloride, 2.5µl 10X Taq buffer (Perkin Elmer), 100mM of each dNTP (Perkin Elmer), 5pmol primer (Advanced Biotechnologies) and sterile molecular biology grade water. The final reaction mixture was overlayed with 35µl mineral oil. Amplification was carried out in a Perkin Elmer Cetus 480 thermocycler under the following conditions: 5min @ 94°C, followed by 45 cycles of 1min @ 94°C, 1min @ 41°C and 2min @ 72°C, then a final extension period of 5min @ 72°C. Amplified reaction products were resolved by agarose gel electrophoresis in 1.5% (w/v) agarose gels (Seakem LE agarose), stained with ethidium bromide and visualised at 300nm.

Data Analysis

Presence/absence data were recorded only for primers giving reproducible, scorable markers. A matrix of pairwise genetic distance were calculated between all possible pairs of plants using Jaccard's coefficient. A matrix of pairwise physical distance between plants was similarly calculated. The correlation between genetic and physical distance matrices was calculated using the Mantel test.

RESULTS AND DISCUSSION

RAPD profiles were derived by screening individual *A. capillaris* plants with a range of 10-mer random primers. The data shown were obtained using Advanced Biotechnologies primer AB4-07 (CAGCACTGAC). This primer disclosed a total of 16 scorable polymorphic loci. Figure 1 shows examples of the highly polymorphic banding patterns that were obtained. No population-specific molecular markers were immediately detectable with this or any other primer tested so far. These results illustrate the high levels of genetic diversity present in this species.

Analysis of the A. capillaris RAPD data using the Mantel test revealed no correlation between genetic distance and physical distance (statistically insignificant correlation; r=0.0326). Figure 2 shows a plot of genetic distance versus physical distance. The high levels of inter- and the intrapopulation genetic variation apparent in Figure 1 are clearly demonstrated by the analysed data.

The high level of genetic variability exhibited by *A. capillaris* at the two collection sites is somewhat unexpected given that its reproduction strategy is thought to be vegetative rather than sexual. The highly polymorphic profiles obtained with a large number of RAPD primers suggests this may not be the case, although polymorphism may also be due to a dynamic polyploid genome undergoing numerous recombination events.

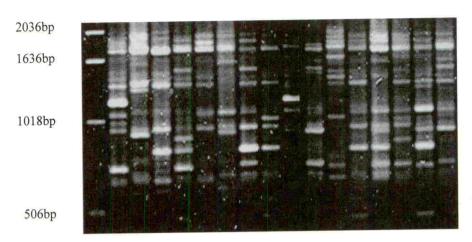


Figure 1. Agarose gel of *Agrostis capillaris* RAPD profiles generated using primer AB4-07. Numbers to the left indicate molecular weights of the DNA standards. Numbers along the top refer to the samples listed below: 1: 1Kb ladder DNA standards 2: Cleish, transect 1, locus 4, plant 1 (c1.4.1), 3: c1.10.1, 4: c3.2.1, 5: c3.9.1, 6: c5.5.1, 7: c5.9.1, 8: c2.4.1, 9: c2.9.1, 10: c4.2.1, 11: c4.8.1, 12: Kirkton, transect 1, locus 1, plant1 (k1.1.1), 13: k1.7.1, 14: k2.5.1, 15: k2.10.1 16: k3.9.1, 17: k3.10.1. Note the apparent polymorphism between samples.

Similarly high levels of genetic diversity have also been observed in the analysed samples of *R. acetosa* and *F. rubra* (data not shown). *R. acetosa* is an obligate outbreeder and so significant variability is expected, representing the flow of genes between individuals of the same and other populations. *F. rubra*, like *A. capillaris*, is generally thought to reproduce asexually and so lower levels of intraspecific variation would be expected. Again, however, *F. rubra* is polyploid which allows potential for recombination.

It is not unusual to detect high levels of polymorphism when screening natural/semi-natural plant populations using RAPDs. For example, Moodie *et al.* (1997) detected extensive polymorphism in RAPD profles of *Sinapis arvensis* and were not able to discriminate populations sampled from herbicide-treated sites from those derived from untreated organic sites.

Further extensive genetic screening is being undertaken using additional RAPD primers. Alternative DNA fingerprinting assays employing minisatellite and microsatellite sequences are also being employed. The resulting genetic data will be correlated with parallel physiological data (root growth and resource remobilisation studies) concerning the ability of the species under study to regenerate in response to grazing pressure.

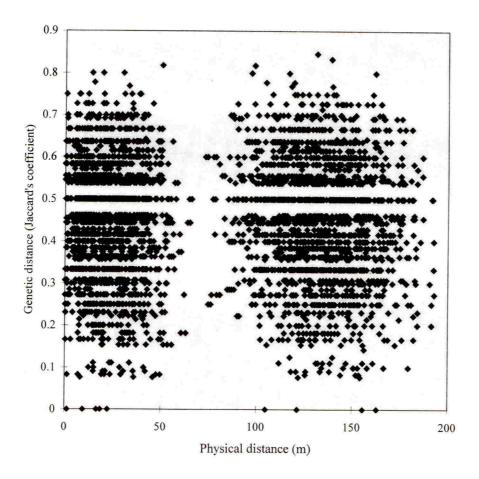


Figure 2. Pairwise genetic distance versus pairwise physical distance between 106 samples of *A. capillaris*. Note that physical distances greater than 70m are shortened by 78.8km (inter-site distance) for the purpose of graphical representation. The cluster to the left represents pairwise relationships within sites. The cluster to the right represents pairwise relationships between the two sites. This plot illustrates the significant intraspecific variability of *A. capillaris* and the lack of correlation between physical and genetic distance, e.g. genetic variability between adjacent plants can be as great as, or greater than, that between adjacent plants.

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COMPETITIVE EFFECTS OF MULTIPLE WEED SPECIES ON WEED BIOMASS AND WHEAT VIELD

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ABSTRACT

An experiment was established in autumn 1992 to study the interactive effects of multiple weed species competition in winter wheat. *Galium aparine*, *Matricaria perforata* and *Papaver rhoeas* were grown singly and as pairs of species in wheat. *P. rhoeas* produced most biomass both singly and in mixture resulting in greatest crop biomass and yield reductions. In mixture, *P. rhoeas* had the greatest influence in depressing the biomass of the associated species. The effects of weeds in mixture on crop biomass were additive in May, but at harvest, yield reductions from weeds in mixture were only slightly greater than from single species.

INTRODUCTION

Competition between crops and weeds has been studied by numerous authors with most studies concentrating on the effects of single weed species. However, natural weed infestations usually contain many different species and the combined competitive effects of these infestations are important when considering weed management strategies. Little information is available for crop-weed relationships when more than one weed species is present.

The crop equivalent system (Wilson, 1986) and standard weed units (Aarts & Visser, 1985) have been used to predict the effects of mixed species infestations by simply totalling the effects of the individual species. This approach may be useful at low weed densities, but takes no account of intra- and inter-specific competition between the weeds themselves. Research by Alex (1970) and Haizel & Harper (1973) suggests that the effects of a mixture of weeds cannot be predicted from the effects of the individual weed species grown alone. Alex (1970) found that the effects of one species tended to obscure the effects of the other, whereas Haizel & Harper (1973) found that mixed populations could produce either a lesser or greater effect than the sum of the weeds alone. In natural weed communities, Hume (1989) found that only one or two species are dominant, but if these are removed other species become increasingly important.

In this paper we describe a multi-species experiment undertaken at Long Ashton Research Station (LARS). The interactions between three broad-leaved weed species (*Galium aparine*, *Matricaria perforata* and *Papaver rhoeas*), grown singly and in mixture at a range of densities with winter wheat, were studied in an additive experiment. The effects on crop biomass, weed biomass and the yield of the crop are described.

MATERIALS AND METHODS

The field experiment was established in October 1992 on fine sandy loam soil at LARS. It consisted of one replicate of a fully randomised design. Three weed species at a range of densities (Table 1) were grown alone in winter wheat and in pairs of species, with every combination of species and density. The experiment was additive in design; the weeds sown to give the target seedling densities both alone and in mixture. Plots with wheat grown in the absence of weeds were also included.

The crop and weeds were sown on 8 October 1992. Weed seeds were sown on to the seedbed surface by diluting with sand and broadcasting from a 'pepperpot' (jar with perforated lid) into plots of 4m x 4m and were incorporated into the soil by the drill when the winter wheat (cv. Hereward) was sown shortly afterwards. A range of seed weights was sown to give five seedling densities.

The central 1m² of each plot was marked for later yield assessment. All crop and weed seedlings were counted in this area during the autumn. Crop and weed plants from outside this central area were used for growth assessments during the season. Naturally occurring weeds were removed by hand from all assessment areas. Crop and weed biomass were assessed at various dates during the spring and summer. Weed plants were removed from all plots and crop plants from only the weed-free plots, an additional crop assessment was made in May 1993, where crop plants were removed from three rows of 50 cm from all plots. Both crop and weeds were washed, oven-dried and weighed. The data were summarised as mean dry weights per plant and biomass m⁻². Weed biomass data are presented as means of all densities.

The experiment was harvested by hand in late July, each 1m² plot being cut to ground level. The weeds were separated from the crop, dried and weighed. Crop stem numbers were counted and the crop sheaf was weighed, threshed and fresh and dry weights of grain recorded. Yields of clean grain at 85 % dry matter were derived and 1000 grain weights obtained.

RESULTS

Weed populations

Weed seedlings emerged early, either with or soon after the crop, and a wide range of densities was established with high numbers of *P. rhoeas* (Table 1).

Table 1. Range of weed densities established

		seedlings m -2		
0	17	22	50	100
0	45	89	149	353
0	90	206	445	780
	0 0	0 17 0 45 0 90	0 17 22 0 45 89	0 17 22 50 0 45 89 149

Biomass of single weed species

From late-March to late-June, *P. rhoeas* produced the greatest biomass, followed by *M. perforata* then *G. aparine*, when grown as single species in wheat (Fig. 1). In late July, *M. perforata* biomass continued to increase and produced the most biomass, whereas both *P. rhoeas* and *G. aparine* biomass declined. Over all dates there was a significant difference between the biomass of the species with *P. rhoeas* the highest, followed by *M. perforata* and *G. aparine* (Table 2).

Weed biomass (g)

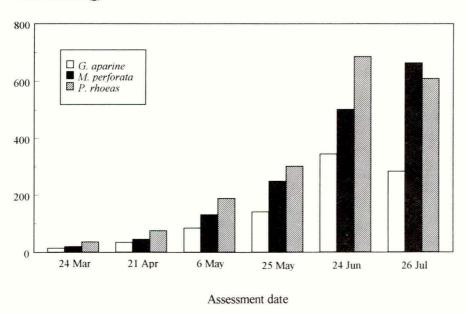


Figure 1. Weed biomass m⁻² for single species in wheat (mean of densities)

Biomass of weeds in mixture

The biomass of each weed species was significantly reduced when in combination with either of the other species compared to its biomass when grown as a single species (Table 2). For *G. aparine* biomass, competition from *P. rhoeas* caused a significantly lower biomass compared with competition from *M. perforata*.

Table 2. Loge weed biomass for single species and mixtures (mean of dates and densities)

		A	dditional species	s.e.d.			
	Single species	G. aparine	M. perforata	P. rhoeas	Single vs. mixture	Between mixtures	
G. aparine	4.32) =	4.01	3.73	0.146	0.120	
M. perforata	4.81	4.47		4.37	0.121	0.098	
P. rhoeas	5.17	4.76	4.83	-:-	0.102	0.084	
s.e.d.	0.089						
d.f.	48				2	.2	

Crop assessments

Crop biomass in May was significantly reduced by the presence of each weed species grown singly compared with weed-free (Table 3). P. P rhoeas caused the greatest decrease followed by P perforata and then P aparine, with reductions of 32%, 26% and 24% respectively. Larger reductions in crop biomass occurred when weed species were persent in combination. P rhoeas P aparine resulted in a 50% decrease, P rhoeas P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas caused a significantly larger crop biomass reduction than that due to P rhoeas caused a significantly larger than that caused by each of these species grown singly.

Table 3. Effect of weed species on crop biomass in May and harvest components

		Crop biomass in May (g m ⁻²)	Grain yield (t ha ⁻¹)	Crop stem number (m ⁻²)
Weed f	ree	675.8	5.01	394
G. apa	rine	513.2	1.33	236
M. per		500.7	1.07	244
P. rhoe		458.1	0.81	159
	rine + M. perforata	444.2	0.72	187
	rine + P. rhoeas	338.5	0.40	158
	forata + P. rhoeas	334.0	0.55	140
s e d	Single species	62.7	0.284	23.4
	Single vs. mixture	54.3	0.246	20.2
	Mixed species	44.3	0.201	16.5
d.f.	19			

Grain yield and stem numbers at harvest showed a similar pattern to crop biomass in May. However, reductions in grain yield and stem number were not significantly lower in the mixtures than those caused by P. rhoeas alone. Again, P. rhoeas alone and in mixture caused the greatest yield losses. However, in this case yield losses due to mixtures with P. rhoeas were not significantly greater than losses associated with G. aparine P. perforata. Grain yield losses were more pronounced than the earlier biomass reductions, the presence of weeds reducing yields by a minimum of 73%.

DISCUSSION

P. rhoeas produced the highest biomass both alone and in mixtures resulting in the largest crop biomass and yield reductions. In mixture, P. rhoeas had the greatest influence in depressing the biomass of the associated species. G. aparine and M. perforata are generally acknowledged to be more competitive than P. rhoeas (Wilson and Wright, 1990). However, in this case the dominance of P. rhoeas both singly and in mixture was most likely to be the consequence of the high plant numbers established. Another contributory factor was arguably the emergence of P. rhoeas at the same time as the crop. Weeds which emerge early with the crop are likely to be more competitive compared with those that emerge later when the crop is well established (Peters, 1984). This early emergence allowed P. rhoeas to compete from the early stages of crop growth until late in the growing season.

In May, effects of the mixed species on the crop were mostly additive, i.e. close to the sum of the effects of the single species, indicating that there was little interaction between the species before then. However, at harvest, reductions from the weed mixtures were only slightly greater than from the single species, thus species interactions appear to have developed from May onwards. This is supported by the work of Alex (1970) who found that the competitive effects of two weed species in a wheat crop were not fully additive because the effects of one species tended to obscure the effects of the other. Our results are based on mean weed densities; we would expect interactions to become greater and earlier with increasing density.

With later competing weed species such as *G. aparine*, *M. perforata* and *P. rhoeas*, predictions based on early spring biomass may produce underestimates in weed competitiveness and thus final yield losses. Further work with combinations of early and late competing weeds would aid our understanding of weed community interactions with crops.

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A SIMULATION APPROACH TO IDENTIFYING THE MECHANISMS OF MAIZE TOLERANCE TO VELVETLEAF COMPETITION FOR LIGHT

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ABSTRACT

An ecophysiological model of interplant competition for light (INTERCOM) was calibrated and tested for maize - velvetleaf competition in the north-central United States. Sensitivity analysis revealed that traits most important for conferring improved maize yield and tolerance to velvetleaf include extended vegetative development, distribution of leaf area over canopy height, fraction of new biomass partitioned to leaves, specific leaf weight, maximum height and time to 50% maximum height. Results are consistent with empirical research, suggesting that ecophysiological models may be useful tools for exploring the causes and effects of weed-crop competition.

INTRODUCTION

Optimizing the efficiency of an integrated weed management program requires an ability to predict the independent and interactive effects of management practices on crop and weed growth and competitive ability. Quantitative understanding is needed to improve predictive ability. Ecophysiological simulation models provide a quantitative structure for integrating the effects of management, the environment and competition on components of plant growth. INTERCOM was developed to simulate the effects of Chenopodium album competition for light and soil water on sugarbeet yield (Kropff & Spitters, 1992, Kropff & van Laar, 1993). The model has since been modified to simulate rice-Echinochloa competition in southeast Asia (Kropff et al., 1994, Lindquist & Kropff, 1996). INTERCOM simulates growth of each species on a daily time step using daily weather data and a number of genotype-specific parameters. Calculation of daily growth and the linkages between competing species were described by Kropff & van Laar (1993), INTERCOM for rice was modified to simulate competition for light between maize and velvetleaf (Abutilon theophrasti) in Nebraska (Lindquist, 1997). In this paper we evaluate INTERCOM performance in simulating maize - velvetleaf competition over two growing seasons and conduct a sensitivity analysis to identify the maize canopy traits most important for crop tolerance to velvetleaf competition for light.

MATERIALS AND METHODS

Estimates for required parameters were obtained for Pioneer 3379 maize and velvetleaf grown over three years (1994-1996) in monoculture under irrigated conditions in Nebraska (Lindquist & Mortensen, 1997a). Monoculture maize growth and velvetleaf growth were previously shown to be accurately simulated in all three years (Lindquist, 1997), so model performance was evaluated against mixture data using a quantitative measure of modeling efficiency (ME, Janssen & Heuberger, 1995) which is similar to an r^2 in regression analysis. Lindquist &

Mortensen (1997b) presented results of experiments in which Pioneer 3379 was grown in mixture with velvetleaf in 20 plots, each comprising six 0.76 m x 14 m rows, in 1995 and 1996. Maize and velvetleaf density was measured in each plot and maize grain yield was measured and yield loss determined from yield in mixture and mean weed-free yield. For the current study, maize and velvetleaf mixtures were simulated using the observed crop and weed density and planting dates from Lindquist & Mortensen (1997b) as input.

Table 1. Parameter name, description, and values used for maize in baseline simulations. LAI = leaf area index, GDD = growing degree days. Values of other parameters were presented in Lindquist (1997).

Parameter	Description	Value
DVRR	Rate of development, reproductive phase (GDD ⁻¹)	0.00136
DVRV	Rate of development, vegetative phase (GDD ⁻¹)	0.00134
SFA	Specific area of reproductive tissues (m ² g ⁻¹)	0.000015
SSA	Specific area of stems (m ² g ⁻¹)	0.00004
KF	Extinction coefficient for reproductive tissue	0.4
KS	Extinction coefficient for stems	0.4
KDF2	Extinction coefficient for leaves, LAI > 1.0	0.63
KDF1	Extinction coefficient for leaves, LAI < 1.0	0.4
HGTH1	Initial height (cm)	1.0
HS/HB	Time (GDD) to 50% maximum height	466
HMAX	Maximum height (cm)	281
LFDB	Shape coefficient for leaf area distribution	2.49
LFDA	Relative height of maximum leaf area density	0.541
RGRL	Relative growth rate of leaves, LAI < 0.6 (GDD ⁻¹)	0.0128
LAO	Initial leaf area per plant (cm ²)	17.773

Table 2. Fraction of new biomass partitioned to leaves (FLV) and reproductive tissue (FRP), and specific leaf weight (SLW, g m⁻²) parameter estimates as a function of development stage (DVS).

DVS	FLV	FRP	SLW
0.0	0.8500	0	350
0.1	0.7743	0	395
0.2	0.6986	O	441
0.3	0.6229	0	486
0.4	0.5471	0	532
0.5	0.4714	O	577
0.6	0.3957	0	623
0.7	0.3200	0	668
0.8	0.2525	0.1350	714
0.9	0.1850	0.2700	759
1.0	0.1175	0.4050	805
1.1	0.0500	0.5400	850
1.2	0.0333	0.6933	788
1.3	0.0000	0.8467	725
1.4	0	1	663
1.5	0	1	600
2.0	O	1	600

Sensitivity analysis was conducted to evaluate the relative importance of parameters to maize yield in monoculture and yield loss in mixture at a velvetleaf density of 3 plants m⁻². All parameters relating to plant morphology were tested in simulations using 1995 weather data. Parameters defining specific leaf weight and the partitioning of new biomass vary with development stage (DVS; 0=emergence, 1=anthesis/flowering, 2=physiological maturity) and values of these parameters were tested at each 0.1 increment of DVS to test their sensitivity during specific growth periods. Results are presented as the change in simulated yield (yieldi-yieldb, where i and b indicate yield calculated for the changed and baseline parameter value, respectively) or yield loss (Yli - Ylb) resulting from a 10% increase or reduction in each parameter. However, only the parameter change (+ or -) resulting in a desirable outcome (increase in yield, decrease in yield loss) was reported. Tables 1 and 2 list parameter abbreviations, descriptions and initial values.

RESULTS AND DISCUSSION

Average percent maize yield loss was accurately simulated for 1995 (ME = 0.73), but mean yield loss was over-predicted by 49% in 1996 (Figure 1). Simulated yield loss was similar at low velvetleaf densities in both years. The irregularity in the simulated yield loss:velvetleaf density relationship is due in part to the method of simulating early leaf area growth (Lindquist, 1997). Model performance in simulating 1995 mixtures was sufficient to continue with the sensitivity analysis. For reference, simulated yield loss at a velvetleaf density of 3 plants m⁻² was 25.2% in 1995.

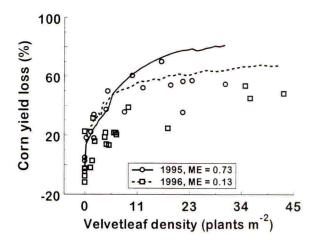


Figure 1. Observed (symbols) and simulated (lines) percent maize yield loss as a function of velvetleaf density for two growing seasons.

Parameters having the greatest influence on yield include rate of development during reproductive (DVRR) and vegetative (DVRV) stages, extinction coefficient for the attenuation of light (KDF2) and relative height of maximum leaf area density (LFDA). Parameters resulting

in the greatest reduction in yield loss include maximum height (HMAX), LFDA, DVRV, time (growing degree days, GDD) from emergence to 50% maximum height (HS/HB) and relative growth rate of leaves from emergence to LAI < 0.6 (RGRL) (Figures 2 & 3).

Increasing time to maturity has long been known to increase yields, thus it is not surprising that reducing the rate of development increases simulated yield. INTERCOM does not currently account for the potential damage of early frost, so it is not possible to evaluate whether a 10% decrease in development rate would genuinely benefit grain yields in Nebraska. Decreasing DVRV resulted in a 1860 kg ha⁻¹ increase in simulated yield and a 2.7% reduction in yield loss (from 25.2 to 22.5%, see Figure 2). Optimum benefit for both yield and yield loss reduction would therefore result from prolonging the vegetative phase of maize development.

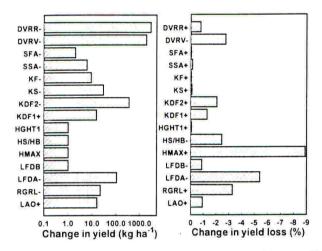


Figure 2. Change in yield or yield loss resulting from a 10% increase or decrease in each of 15 INTERCOM parameters. The + or - symbols following the parameter name indicate that the change shown occurred when the parameter was increased or decreased respectively.

Light attenuation within a crop canopy varies exponentially as a function of cumulative leaf area index. The extinction coefficient is commonly defined empirically as the shape coefficient of this relationship. Goudriaan (1988) showed that the extinction coefficient is dependent upon leaf angle distribution. Generally, erect leaves result in a lower extinction coefficient whereas planar leaves result in a higher extinction coefficient. Simulation results suggest a negative trade-off between yield and tolerance for this trait. Maize yield increased by 347 kg ha⁻¹ and yield loss increased 2.5% (from 25.2 to 27.7%) when KDF2 was reduced (Figure 2). Thus, a more erect leaf angle distribution may increase yield, but would also reduce crop tolerance.

Increasing the relative height at which maximum leaf area density occurs (smaller LFDA) increased yield by 106 kg ha⁻¹ and reduced yield loss 5.4% (Figure 2). Tollenaar & Aguilera (1992) showed that leaf area was distributed higher on maize plants when the population was increased, suggesting that cultural practices may be used to modify leaf area distribution to optimize both yield and crop tolerance.

Changing maximum height (HMAX) and time to 50% HMAX (HS/HB) had no impact on yield, but reduced yield loss by 8.9% and 2.4% respectively (Figure 2). This result may be expected under the assumption that there is competition for light only; velvetleaf does not respond to changes in maize height. A taller maize canopy has more leaf area above the velvetleaf, absorbs more radiation and produces more biomass. Reducing HS/HB confers a similar advantage on the maize prior to canopy closure.

Increasing the fraction of new biomass partitioned to leaves, at the expense of stems, results in a yield increase of up to 620 kg ha⁻¹ and a yield loss reduction of up to 1.6% (Figure 3). Yield is maximized and yield loss minimized when this increase occurs at DVS = 0.5 to 0.6, which occurs during the most rapid phase of leaf area growth. Increasing biomass partitioning to reproduction, at the expense of stems, increased yield by up to 990 kg ha⁻¹, but had little impact on yield loss. A reduction in specific leaf weight at DVS = 0.4 to 0.6 increased simulated yield by 150 kg ha⁻¹ and reduced yield loss by 1.5% (Figure 3). Optimum yield increase occurred with changes in SLW during reproduction, but the benefit to crop tolerance was minimal.

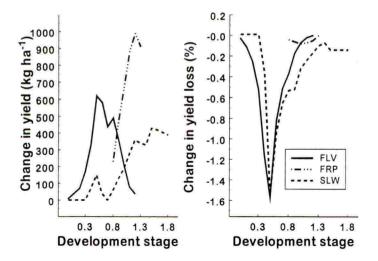


Figure 3. Change in yield or yield loss resulting from a 10% increase in the fraction of new biomass partitioned to leaves (FLV), reproductive organs (FRP), or a 10% reduction in specific leaf weight (SLW).

Lindquist & Mortensen (1997b) measured morphological characteristics of four maize hybrids in monoculture and in mixture with velvetleaf. Maize traits having the greatest correlation with yield loss included maximum leaf area index (LAI) and height, time (GDD) between emergence and 50% maximum LAI and height, and relative height of maximum leaf area density (LFDA). Definition of the parameters HMAX and LFDA discussed in the present paper are identical to those analyzed by Lindquist & Mortensen (1997b), and several parameters that were important in this sensitivity analysis are important determinants of LAI. Results of our analysis are consistent with their empirical findings, but provide more detailed information on specific traits (e.g. SLW rather than just LAI) that are important to both yield and crop tolerance, and on the

stage of development that these traits are most important. Results suggest that ecophysiological simulation models are useful tools for gaining improved understanding of the mechanisms of crop-weed competition and for identifying management practices useful in manipulating these interactions.

In the present analysis, an increase in relative growth rate of leaves while LAI < 0.6 (RGRL) increased yield by 220 kg ha⁻¹ and reduced yield loss by 3.3%. Rapid early growth may be critical for improving competitiveness of some crops (Jordan, 1993), but Lindquist & Mortensen (1997b) found no correlation between maize yield loss and RGRL. Moreover, Lindquist (1997) found that empirical estimates of RGRL vary with year of measurement. Further research is needed to improve the method of estimating RGRL to account for annual weather variation and to evaluate its importance in maize tolerance to weed competition.

ACKNOWLEDGMENTS

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ACTIVITY AND PERSISTENCE OF SORGOLEONE, A LONG-CHAIN HYDROQUINONE PRODUCED BY SORGHUM BICOLOR

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ABSTRACT

Sorghum bicolor (L.) Moensch and other related Sorghum species produce a long chain hydroquinone compound, MW=358, which is exuded by living root systems. This compound exhibits potent activity as a photosynthetic inhibitor. A diverse group of sorghum germplasm was evaluated for sorgoleone production. Production was quite variable, with certain accessions producing up to 15 mg sorgoleone/g fresh root weight. The root exudate composition among accessions was less variable, with sorgoleone the major constituent (76-99%) within the extract. The potential binding of sorgoleone to the D1 protein of the PSII complex was evaluated in triazine resistant and susceptible redroot pigweed (Amaranthus retroflexus (L).) thylakoids. Sorgoleone, metribuzin and diuron exhibited competitive binding with atrazine in susceptible thylakoids, while no competition was evident in resistant thylakoids. Sorgoleone has an intermediate affinity between that of diuron and metribuzin from estimated binding constants. Computer-aided design programs have proven useful to further evaluate structural activity relationships for sorgoleone, and PSII inhibitors. Sorgoleone at concentrations of over 40 ppmw reduced shoot fresh weight in several broadleaf weed species, when incorporated in soil, but had no effect on root development. Recovery of soil impregnated with sorgoleone was most effective (up to 85%) when acetonitrile:water (80:20 v/v) was used as the extractant, in comparison to methanol:water or water alone. Recovery declined over a 42 day period after incorporation.

INTRODUCTION

The use of allelopathic traits of crop species for weed suppression in agroecosystems is an idea first suggested by A.R. Putnam in the 1980's. More recently, the weed suppressive nature of cover crop species or green manures has been utilized to aid in weed management in both horticultural and agronomic cropping systems. Weed suppression provided by these covers can be variable, and is dependant upon factors such as inherent weed pressure encountered, climate, cover crop, amount and longevity of residue, soil microbial populations and water availability. The interactions of higher plants within an agroecosystem are often poorly understood and strong evidence for allelopathic interference is typically unavailable or poorly documented in the literature. However, the interference and weed suppressive potential exhibited by *Sorghum bicolor* (L.) Moensch in the field and by the natural product sorgoleone in the laboratory present a unique opportunity to assess the physiological basis of plant growth interference through allelopathy.

In the U.S., sorghum is cultivated upon more than 600,00 ha for grain, with greater than 350,000 ha used for forage, green manures and in the production of sorghum syrup (Heath et al. 1985).

Sorghum is often chosen as a summer annual cover crop becuase of its rapid growth and ability to suppress weeds (Forney et al, 1985). In Michigan sorghum covers are killed and used as weed suppressive residues in orchards. The allelopathic effect may last for several weeks after sorghum is killed (Putnam et al, 1983; Putnam and DeFrank, 1983). In Mississippi, Smeda has shown that spring-planted sorghum residues provide up to 90% reductions in weed biomass for 6 to 8 weeks in no-till summer-planted soybeans (personal communication). When sorghum was incorporated as a green manure, it strongly suppressed annual weeds in succeeding alfalfa crops (Forney et al, 1985). Einhellig and Rasmussen (1989) have shown that the inhibitory effects of grain sorghum on surrounding weed growth occurred through the following growth season.

The ability of sorghum residues or green manures to suppress weeds has been discussed in the literature for years, but until recently, the chemistry of the potential inhibitor (s) remained unknown. Several sorghum species including S. halpense, S. vulgare, S. sudanese, and S. bicolor have shown allelopathic interference with weed and crop growth (Einhellig and Souza, 1992). Sorghums produce and release cyanogenic glycosides, and a number of phenolic breakdown products of these glycosides contribute to short term plant growth suppression (generally less than 8 weeks) in field and greenhouse experiments (Einhellig and Rasmussen, 1989); Guenzi and McCalla, 1966; Nicollier et al 1983; Weston et al, 1989). The toxicity of sorghum to livestock has long been associated with the release of HCN from dhurrin (p-hydroxy-(S)-mandelonitrile b-Dglucopyranoside)(Figure 1), which is present in sorghum herbage. Although dhurrin was not phytotoxic in selected assays, two compounds identified from sudex (S. bicolor x S. sudanese) herbage by Weston et al (1989), p-hydroxybenzoic acid and aldehyde (Figure 1), are breakdown products of dhurrin. These simple phenolics possessed significant activity in bioassays of germinating seedlings and hydroponic systems (Weston, unpublished data). Using a modified Parker bioassay that simulated field conditions, Weston et al (1989) determined that herbage of young seedlings (2 to 4 weeks of age) was most phytotoxic and produced greater levels of these products on a per g tissue basis than older herbage (6 to 8 weeks of age).

Sorghum roots, herbage and germinating seeds all release phytoinhibitors. Panasuik (1986) established that germinating sorghum seeds inhibited germination and growth of grass and broadleaf species. In our own work, under closely controlled conditions for light and moisture availability, germinating sorghum was also inhibitory to growth of several weeds (Hoffman et al, 1996). Forney et al (1985) found that rhizosphere products of hydroponically grown sudex were

CHI,O

Sorgolone mw=358

more phytotoxic than compounds from other plant parts and toxicity increased with increasing plant age up to 6 weeks. They also noted a yellow pigmentation associated with the rhizosphere product. Our own work with ornamentals planted into living sudex root systems showed that seedlings developed chlorosis rapidly in the presence of sudex roots, and exhibited minimal growth over a 7

week period. This inhibition could not be overcome by the addition of water-soluble fertilizer. The living root system of sudex was most likely associated with the greatest phytotoxicity to ornamental seedlings (Geneve and Weston, 1988).

Netzley and Butler (1986) first reported on the presence of sorgoleone in the root exudates of *S. bicolor*. They isolated sorgoleone the oxidized quinone form of a hydrophobic p-benzoquinone from sorghum root exudates. Sorgoleone is the major constituent of sorghum root exudates (Table 1) and is easily extracted from roots in methylene chloride plus a small amount of acetic acid

(Netzley and Butler, 1986; Nimbal et al., 1996). Sorgoleone possesses remarkable phytotoxicity in numerous plant growth assays (Einhellig and Souza, 1992; Nimbal et al., 1996) and is primarily an inhibitor of plant growth, apparently through inhibition of photosynthesis (Einhellig and Souza, 1992; Nimbal et al., 1996b) and respiration (Rasmussen et al., 1992).

The mechanism of action of many herbicides inhibiting photosynthesis involves the blockage of electron transport in photosystem II (PS II) by binding to the Q_B electron acceptor at the D1 protein. This characteristic type of inhibition has been reported in triazines, phenylureas, ureas, uracils among others (Mets and Thiel, 1989). Binding studies have shown the existence of a common binding niche at Q_B to which these herbicides bind and a close correlation between electron transport inhibition and herbicide binding (Nimbal et al, 1996b). Plants resistant to triazines serve as a useful tool to examine the concept of different but overlapping binding sites at the 32 kDa protein of the PSII complex in diuron-type herbicide inhibitors. Studies in our laboratory with sorgoleone have shown that sorgoleone is a potent inhibitor of photosynthesis and its site of inhibition is within the PSII complex. The activity of sorgoleone compared well with that of diuron (Gonzalez, 1997; Nimbal et al, 1996). Further studies were conducted to evaluate the ability of diverse sorghum germplasm to produce sorgoleone, the specific mode of action and binding characteristics of sorgoleone within PSII, and soil persistence of sorgoleone over time, key questions related to the importance of sorgoleone as an allelochemic and its physiological role in the interference mechanism of *Sorghum bicolor*.

MATERIALS AND METHODS

Screening of sorghum germplasm for sorgoleone production. A diverse collection of grain sorghum germplasm (Table 1) was screened for sorgoleone production with 5-day-old seedling roots. Germination percentage and root fresh weight were recorded for each germplasm, and total quantity of sorgoleone produced was determined. The extraction procedure for sorgoleone was performed as described in Nimbal et al., 1996, where Pioneer 8333 seeds were germinated in a seed germination chamber at 29 C for 5 days in the dark. Seedling roots were excised and dipped in methylene chloride plus 1% glacial acetic acid. The crude extracts were filtered and then subjected to HPLC (reverse phase Nova-Pak C₁₈ column, 3.9 x 150 mm). The mobile phase was 75% acetonitrile/25% acidified water. Water was acidified with glacial acetic acid at 2.5%. Sorgoleone was detected as reported in Nimbal et al. 1996 using a Waters absorbance detector at 280 nm after 20 μL of the acetonitrile solubilized crude extract sample was injected. Flow rate was 2 ml/min with a 10 min total run time. For further experimentation with sorgoleone, Pioneer 8333 seeds were utilized and extracted. Sorgoleone (MW=358) constituted more than 90% of the extract.

Binding of sorgoleone and other PSII inhibitor herbicides. Experiments were conducted using labeled atrazine and unlabeled herbicide inhibitors (Nimbal et al, 1996b). The ability of sorgoleone to compete for a common binding site in PSII was evaluated in comparison to other inhibitors by varying the ^{14}C atrazine concentration from 0.01 to 0.4 μM in the presence or absence of 0.5 or 1.0 μM concentration of inhibitors. All binding experiments were conducted as reported in Nimbal et al, 1996b at room temperature and reduced light. Thylakoid membranes were isolated from resistant and susceptible redroot pigweed following the protocol of Smeda et al, 1993. Chloroplasts equivalent to 50 μg Chl/ml were suspended in 1 ml of binding medium containing ^{14}C atrazine. Inhibitors were then added to each tube and samples were incubated for 3 min after

vortexing. Finally, samples were centrifuged at 15,000 g for 5 min and an aliquot of 0.5 ml from the supernantant was used for radioactivity measurements to determine the amount of unbound atrazine. The amount of bound ¹⁴C atrazine was calculated by subtracting unbound atrazine in the supernatant from total added to the chloroplast suspension. Competitive binding was evaluated by plotting the total concentration of unlabeled inhibitor versus the concentration bound ¹⁴C atrazine per mg chlorophyll. The mechanism of competition was evaluated using double reciprocal plots. Competitive binding is indicated by a common intercept on the ordinate. The binding constant (K_b) for unlabeled inhibitors was computed by replotting the slopes of the double reciprocal plot versus the total concentration of unlabeled inhibitor. The absicissa intersection of the straight line fitted for this new data plot indicates the binding constant for that particular inhibitor (Nimbal et al, 1996b)...

The binding of sorgoleone to the Q_B binding site of the D1 protein was further analyzed using 3 dimensional computer aided graphics programs such as MOPAC 93 (Quantum Chemistry, Bloomington IN) and Chem-X (Chemical Design Limited, Oxfordshire, UK). Plastoquinone, sorgoleone, PSII inhibiting herbicides and 12 benzoquinones were evaluated using various software packages for bulk, electronic and energy properties. Electrostatic and lysophilic field potentials were evaluated, among other properties and correlated with biological activity of these compounds and their binding potential within the Q_B binding site of the D1 protein.

Soil activity and persistence of sorgoleone. Since sorgoleone is a major component of root exudates produced by sorghum which are released into soil by living sorghum root systems, we wanted to evaluate the soil activity of sorgoleone against weed species. Assays were performed by impregnating a sand soil mixture (50% sand/50% Maury sterilized Maury silt loam) with sorgoleone at concentrations ranging from 10 to 80 ppmw. Sorgoleone was applied by dissolving in a trace amount of acetone first, and final suspension in milli-Q water. Selected weed species were then grown for a 3 week period in plastic cones filled with treated soil. Roots and shoot fresh weights were evaluated at experimental termination. Weeds evaluated included common purslane (Portulaca oleracea), velvetleaf (Abutilon theophrasti), sicklepod (Cassia obtusifolia), redroot pigweed (Amaranthus retroflexus), large crabgrass (Digitaria sanguinalis) and green foxtail (Setaria viridis).

A stock solution of 10 mM sorgoleone was prepared in acetonitrile and added to triplicate 100 g soil samples in 500 ml glass bottles and mixed by shaking to achieve 25 ppmw concentration. Sorgoleone fortified soil was then incubated at room temperature. Sorgoleone persistence over time was evaluated by collecting samples of 10 g soil from each triplicate set at 0, 7, 14, 28 and 42 days after incubation. Extraction was performed using 20 ml of acetonitrile/water (80/20 v/v) and shaking on a rotary shaker at 180 rpm for 6 hours. Soil extracts were filtered using a .2 µm syringe and evaporated to dryness. Extracts were redissolved in acetonitrile (1 ml) and analyzed by HPLC using gradient analysis. HPLC was Waters system with a C₁₈ Nova-Pak column as described previously and mobile phase was acetonitrile/acidified water (2.5% glacial acetic acid), 80:20 v/v for 1 minute. 75:25 v/v 1-3 min and 60:40 v/v 3-6 min. The flow rate was 2 ml/min. Sorgoleone standard was obtained by extracting sorghum seedling roots and purfication by HPLC. Purified extract contained 98% sorgoleone or greater. Preliminary experiments to evaluate extractability of sorgoleone from soil with acetonitrile/water and methanol/water were performed, as well as extraction time for efficient recovery.

Screening of sorghum germplasm for sorgoleone production indicated that considerable variability

exists among genotypes with regard to the amount of sorgoleone produced (Table 1)(Nimbal et al. 1996). Rtx433 produced only 0.67 mg/g sorgoleone of root fresh weight, while B Redlan and IS 1318C produced 17.8 and 14.2 mg of sorgoleone, respectively. Most other genotypes ranged in between 1.5 and 10 mg/g. In another collection, Hess et al (1992) showed little difference among production among cultivars, but indicated production was sensitive to environmental conditions. Our studies, performed under controlled environmental conditions, showed that production and secretion of sorgoleone may also be dependent on inherent genetic differences among *Sorghum* genotypes. On average, sorgoleone constituted 85-90% of the root exudate composition of the germplasm evaluated, but the range varied from 76 to 99%. This range in purity suggests that differential allelopathic interference among genotypes may exist.

Table 1. Sorgoleone production by various sorghum genotypes^a

		3	G	
Sorghum	Root fresh wt	Sorgoleone	Sorgoleone	%
genotype	(g)	(mg)	mg/g root wt	Purity
Rtx433	0.15 ± 0.03	0.10 ± 0.00	0.67	91.8
R NB9040	0.13 ± 0.01	1.00 ± 0.25	7.70	89.5
IS 3723C	0.29 ± 0.01	1.23 ± 0.18	4.24	85.7
IS 8266C	0.22 ± 0.01	1.10 ± 0.10	5.00	87.2
B N122	0.35 ± 0.01	2.00 ± 0.10	5.71	75.9
Rtx7078	0.15 ± 0.01	1.80 ± 0.21	12.00	98.7
B Martin	0.32 ± 0.03	1.73 ± 0.13	5.40	78.2
IS 8160C	0.22 ± 0.03	2.00 ± 0.15	9.10	83.8
Rtx415	0.20 ± 0.02	1.87 ± 0.03	9.35	92.4
Rtx430	0.08 ± 0.04	0.30 ± 0.10	3.75	94.9
IS 1318C	0.17 ± 0.03	2.43 ± 0.47	14.20	87.0
Rtx7000	0.25 ± 0.02	2.30 ± 0.32	9.20	78.9
Greenleaf	0.06 ± 0.01	0.73 ± 0.47	11.40	99.1
IS 7333C	0.27 ± 0.01	1.63 ± 0.27	6.00	83.4
EH-Sart	0.30 ± 0.04	1.60 ± 0.21	5.33	87.1
IS 5893C	0.19 ± 0.02	0.33 ± 0.07	1.74	82.4
Btx3042	0.19 ± 0.02	0.30 ± 0.06	1.58	77.7
B Redlan	0.15 ± 0.03	2.67 ± 1.31	17.80	80.2
R N97	0.25 ± 0.00	1.70 ± 1.15	6.80	81.6
Piper	0.17 ± 0.01	0.17 ± 0.06	1.00	98.6
IS 1269C	0.20 ± 0.03	0.77 ± 0.15	1.10	83.2
IS 7041C	0.37 ± 0.02	0.60 ± 0.31	1.62	87.9
IS 1098C	0.08 ± 0.01	0.20 ± 0.10	2.50	88.2
IS 12611C	0.27 ± 0.02	0.67 ± 0.03	2.48	84.1
B Wheatland	0.33 ± 0.06	0.83 ± 0.23	2.50	78.3

^{*}The data are means (and SD) of three replicates of 25 seedlings each. Taken from Nimbal et al. 1996.

Since direct binding studies for sorgoleone onto thylakoid membranes are not possible in the absence of ¹⁴C sorgoleone, we determined the potential binding of sorgoleone to the D1 protein of the PS II complex by competitive binding of sorgoleone versus atrazine. The

binding behavior was then evaluated in triazine susceptible (S) and triazine resistant (R) redroot pigweed thylakoids. Sorgoleone, diuron, and metribuzin did not show competitive binding to thylakoids of R-pigweed, even at higher concentrations (Figure 1). A quantitative analysis of competitive binding data in which data are transformed to linear relationships is possible in double reciprocal plots (Nimbal et al, 1996b). When atrazine concentration was varied in the presence or absence of 0.5 mM concentrations of the 3 inhibitors (Figures 1A, 1C, and 1E), regression lines generated for plots with and without inhibitor had similar ordinate but different abscissa intercepts. These findings confirm that all 3 inhibitors competed effectively with atrazine for binding to S pigweed thylakoids. No competition between atrazine and these inhibitors was evident in R thylakoids. Binding constants were calculated and were similar to values reported in past literature with diuron and metribuzin (Buman et al, 1992). The affinity for binding, from the estimated binding constants, indicates that sorgoleone has an intermediate affinity between that of diuron and metribuzin (data not presented, Nimbal et al, 1996b).

SITE OF ACTION OF SORGOLEONE

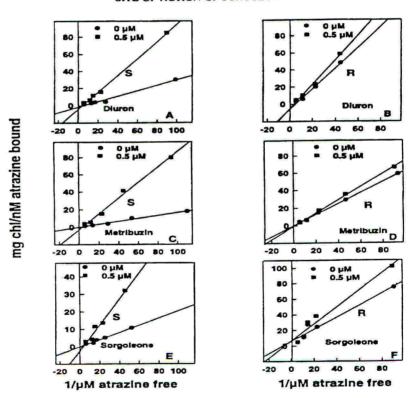


Figure 1. Double reciprocal plots for binding of ¹⁴C atrazine in competition with sorgoleone, diuron, or metribuzin to thylakoids isolated from triazine-resistant (R) and triazine-susceptible (S) redroot pigweed. Taken from Nimbal et al, 1996b.

The use of 3D computer aided design to evaluate structural activity relationships among herbicides and potential herbicide binding sites is widely utilized to predict potential biological activity. An evaluation of sorgoleone in the 3D binding pocket of the QB site at the D1 protein has shown that the electrostatic charge distribution of sorgoleone is highly similar to that of metribuzin and other PSII inhibitors and placement of the molecule within the binding pocket is similar to that of other PSII inhibitors. The charge distribution may account for the strong binding of sorgoleone at the PS II reaction center, in a similar manner to that of diuron-type inhibitors and triazines. Further structural activity work is currently in progress and necessary to evaluate the binding potential of sorgoleone and other related quinones.

To determine if sorgoleone had soil activity against a number of weed species, bioassays were conducted by impregnating soil with sorgoleone at concentrations ranging from 10 to 80 ppmw. After a 3 week growth period, sorgoleone presence resulted in inhibited shoot growth with little or no effect on root development (data not presented). A concentration dependent inhibition of growth was observed in selected species. Shoot fresh weight as well as shoot length of common purslane, velvetleaf, sicklepod and pigweed were reduced at concentrations 40 ppmw or higher, while crabgrass and green foxtail appeared to be less susceptible for growth inhibition at these concentrations. Einhellig and Rasmussen (1989) also noted in field experiments in which grain sorghum residues were located the previous year that the inhibitory effects of grain sorghum were primarily on broadleaf weeds, with little activity observed on grass weeds. They observed these effects the following year, indicating that the allelopathic potential of the crop and or chemical(s) may persist. Our experimentation with soil extraction of sorgoleone-innoculated soil over time attempted to evaluate soil persistence (Nimbal and Westen, 1997). The hydrophobic nature of sorgoleone makes it difficult to extract from soil using aqueous extraction. However, extraction with acetonitrile and water (80:20 v/v) improved recovery and extraction for short periods of time (1 hr) gave recovery rates of up to 85% when compared to longer extraction periods (24 hr) with recoveries averaging 45%. Methanol:water extraction (80:20 v/v) gave low rates of recovery at all extraction times. Recovery of parent sorgoleone was highest initially, and declined substantially over a 6 week period (data not presented). Thin layer chromatography of soil extracts indicated the presence of sorgoleone-derived metabolites, but structure of these metabolites is currently unknown.

Allelopathic crops offer potential for development of model herbicides as well as providing a source of germplasm that could be manipulated to enhance weed suppression in an environmentally compatible manner (Weston, 1996). As we learn more about the mechanisms of allelochemical selectivity, mode of action and regulation of biosynthesis, we should be able to successfully manipulate our germplasm resources to select for enhanced weed suppression. The strong weed suppressive ability of *Sorghum* cover crops and phytotoxicity of sorgoleone offer interesting possibilities for effective biorational approaches to weed management.

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