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DISTRIBUTION OF ELATERIDAE AND TIPULIDAE PEST SPECIES IN AN AGRICULTURAL LANDSCAPE

By

HELEN MARY HICKS

A thesis submitted to the University of Plymouth in partial fulfilment for the degree of

DOCTOR OF PHILOSOPHY

School of Biological Sciences Faculty of Science

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DISTRIBUTION OF ELATERIDAE AND TIPULIDAE PEST SPECIES IN AN AGRICULTURAL LANDSCAPE

HELEN MARY HICKS

Wireworms and leatherjackets, the larvae of click beetles (*Agriotes* spp) and crane flies (*Tipula* spp) respectively, are subterranean pests of grass, cereals and vegetables and are present throughout the UK and Europe.

Soil cores, pheromone traps and water traps were used to measure populations. The populations of larvae were compared to those of adults. Leatherjackets and crane flies were found to be correlated, as were *A. obscurus* click beetle and wireworm populations. Annual variation in trap catches was observed. Populations were related at a number of spatial scales to cultural, physical and chemical variables.

Spatial structure of populations was identified at all scales (sub field, field and landscape) using SADIE analysis, Moran's I and Taylor's Power Law, but the presence or absence of discernable spatial structure was dependent on species, population and annual variation. Environmental variables were linked to populations using multiple regression; the most common variable for all species were the number of years in grass, but the populations of each species and life stage were defined by

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its own set of variables. Important population defining variables were assessed as potential organically compliant cultural pest control methods. These included rolling, cultivation and crop rotation, but none could be definitively recommended as control methods without further work.

In addition, pheromone traps for click beetles were characterised, it was found that the three species of click beetles had differential speeds of travel and the traps had differing effective ranges. *A. lineatus* were found to travel the fastest, *A. obscurus* slower and *A. sputator* travelled slowest. The maximum sampling range (over 30 days) was greatest for *A. lineatus*, less for *A. obscurus* and least for *A. sputator*.

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At no time during the registration for the degree of Doctor of Philosophy has the author been registered for any other university award without prior agreement of the Graduate Committee.

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South West Soils discussion group 'Soil function, quality and indicators - useful concepts?'

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Chapter 1 Introduction

1.1 Pests in Agriculture and Horticulture

All agricultural systems lose productivity due to a number of factors. These include: adverse weather conditions, weeds, disease and animal pests. The Horticultural Development Council estimates that 10 to 20 per cent of horticultural production is lost to pests and diseases each year (National Audit Office 2003). Little empirical data on pest damage to organic crops is available, with no resources such as annual reports of damage available. Pests do not cause significant damage on conventional crops, because insecticides are used to prevent this. So, figures for conventional systems are usually defined by the quantity of pesticide used. As most pesticides are used on a range of target species it is therefore not possible to back-calculate to the prevalence of theses specific pests in conventional systems.

Different pests cause different degrees of crop loss. This may be as a result of consumption of the crop by the pest causing a reduction in yield of the pest may cause damage to crops, which affects quality. A reduction in crop quality may be much more damaging economically although no absolute values are available. Therefore, identifying those pests responsible for the greatest amount of harm is not a simple task. Horticultural crops are becoming increasingly vulnerable to significant economic impact by pest feeding as consumer demand for unblemished produce becomes stronger. The pests identified by growers as responsible for most damage in UK horticulture are aphids, slugs, vertebrates, nematodes, caterpillars, thrips; leatherjackets, slugs, cutworms and cabbage root fly (Table 1-1). Table 1-1 Key animal pests observed in UK organic field vegetables, 1999-2001, from Davies et al. (2002)

Problem observed:	* problem in	one season ** problem in two seasons		
	*** problem	in three or more seasons		
Grower Priority:	Low	• • Medium	a 'o e	High

Crop	Disease/Pest	Farm System		Grower	Causes of problem as	
		Small	large	Priority	identified by growers	
Carrots	Carrot fly	-	*	• • •	Market specifications, products	
	Slugs	-	-	• •	Rotations, knowledge, cost of control	
	Vertebrates	-	-	••	Cost of control	
	Nematodes	-	-	• •	Rotations	
Brassicas	Aphids	*	* * *	•		
	Vertebrates	*	**	• • •	Cost of control	
	Slugs	-	*	•••	Products, knowledge transfer	
	Cabbage root fly	*	* * *	••	Cost of control	
	Caterpillar	-	* * *	• •.•	Market specifications	
Leeks	Thrips	-	* *	-	Market specifications	
Lettuces	Aphids	-	*	• •:•	Large field size, no predators, no aphicides,	
	Vertebrate	-	**	0	Cost of control	
	Leatherjackets	-	*	•	Rotation (leys)	
	Slugs	-	*	•	Rotation, knowledge, cost of control	
	Cutworms	-	-	•	Rotation (leys)	

Table 1-1 highlights crop rotation issues as a primary cause of pest problems in organic field vegetables. Short term grass based phases (leys) are an important factor in organic vegetable rotations as they build fertility and help to reduce the build-up of some pest populations. However, many soil pests, especially nematodes, cutworms, leatherjackets and wireworms (Table 1-2), can become a problem if a grass ley is present. Pests that spend part or all of their life in the soil are intrinsically more difficult to study and this is reflected in the reduced quantity of

literature relating to them.

Pest	Lifecycle in soil	Feeding Method	Crops affected
Cutworms	Larval stage	Root or	Potato ^{1,2}
		foliage	Carrots/parsnips/ celeriac²
			Leeks ¹
			Lettuce ²
			Onions/leeks/garlic ²
			Brassica ²
			Herbs ²
			Beet Crops ²
Leatherjackets	Larval Stage	Root Feeder	G rass ^{1, 2}
			Brassica ²
			Cereals ^{1, 2}
			Beet Crops ²
			Potatoes ²
Nematodes	All	Root Feeding	
Slugs	All	Foliage, roots	Potatoes ²
		or tuber feeder	Brassica ²
			Cereals ²
			Lettuce ²
			G rass²
			Beet Crops ²
Weevils	Larval stage		Brassica²
			Beans ²
			Peas ²
White Grubs		Root feeder	
Wireworms	Larval stage	Root Feeder	Potatoes ²
			Beet Crops ²
			Cereals ^{2,3}

Table 1-2 Common UK soil pests and the crops most commonly damaged by them

¹ Green (2003)

² Whitehead (2004)

³ Eizaguirre (2005)

Five percent of organic growers named leatherjackets (*Tipula* spp.) and wireworms (*Agriotes* spp.) as their worst pest, whereas no conventional growers considered them their most serious pest (Peacock and Norton 1990). A recent Defra project (Cuttle *et al.* 2006) identified leatherjackets and wireworms as the worst invertebrate pests in fertility building crops, especially grass/clover leys and consequently prioritised the search for organically-compliant control methods for these pests .

Neither the damage caused by leatherjackets or wireworms independently has been sufficiently quantified in vegetable production. Davies *et al* (2002) estimated that 10% of organic crops were suffering serious losses due to either pests or diseases, and almost half the organic growers questioned by Peacock and Norton (1990) estimated their loss of revenue to pests and diseases at more than 10%. Insecticides accounted for 36% of the total pesticide-treated area of vegetable crops grown in Great Britain in 1999 (Garthwaite *et al.* 1999), although it is difficult to quantify what proportion is used against these particular pests.

1.1.1 Leatherjackets as pests

In heavily infested cereal fields damage by leatherjackets shows as bare patches where the crop has been destroyed (Anon. 1984). Farmers are advised that chemical control in cereals should be used if, in 17.5cm spaced drills, a total of 15 or more leatherjackets are found on examination of ten 30cm lengths of drill taken at random across the field (Anon. 1985).

Potatoes, leeks, lettuces, brassicas, courgettes and swedes (Anon. 1984; 1985; Davies *et al.* 2002) are the vegetables most at risk from leatherjacket damage by

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direct consumption of roots, seeds and seedlings. With extremely high populations it may also be possible to see damage in recently re-seeded grass. It is unlikely that damage is visible in established grassland, however, it has been shown that even moderate populations (i.e. <1 million ha⁻¹) can cause a significant reduction of dry matter yield in established grassland (Blackshaw 1984).

1.1.2 Wireworms as pests

Wireworms have also been identified as a pest in potatoes, cereals and vegetables (Anon. 1948; Parker and Howard 2001), most commonly causing damage to crops when a cereal or root crop is planted after permanent pasture has been ploughed. In cereals, the wireworm eats through the stem of the young plant just above the seed. In potatoes, the wireworms make small tunnels into the tuber when feeding; these small holes do not cause a large reduction in yield but cause the potato to fall below quality standards required by the consumer. The small tunnel may also allow ingress by other pests such as slugs or millipedes. It would also be logical that these small tunnels would allow ingress by storage pathogens; however, I have seen no reports of this.

Average damage levels may be related to average infestation levels but the level of damage within individual sites can be highly variable. It has proved very difficult to relate wireworm population levels to damage to crops no matter what form of population sampling has been employed (Parker 1994; Ross *et al.* 1948; Samson and Calder 2003). Only the larger/older wireworms cause significant damage to crops (Parker and Howard 2001), because wireworms live for a number of years a population may contain individuals of varying ages, therefore a high population of

20

young/small wireworms may do little damage, whereas a medium population consisting primarily of large wireworms may do considerable damage.

1.2 Leatherjacket Biology and Ecology

1.2.1 Leatherjacket life cycle

Leatherjacket biology and ecology has been reviewed in detail by Blackshaw and Coll (1999). Leatherjackets are the larvae of crane flies, *Tipula paludosa* Meigen and *Tipula oleracea* L. (daddy-long-legs). *T. paludosa* females typically produce about 350 black, oval eggs, which are laid in the soil in late August and September (Coulson 1962). Where light traps have been used in South Devon, peak numbers of females have been caught between 15th September and 9th October between 1967 and 1974 (Mayor and Davies 1976). The eggs hatch in about 14 days to be small legless grubs. The overwintering leatherjacket larval stage lasts 25 weeks (Laughlin 1967) (Figure 1-1).



Figure 1-1 Leatherjacket: larva of Tipula spp.

Although leatherjackets feed all winter, their main growth period is in spring, when they grow to about 4cm long (Coll and Evans 1999; Laughlin 1967). They are commonly found in grassland and feed on roots and below ground shoots of winter cereals and grass (especially clover) (Anon. 1984; Blackshaw 1984; 1990), and also brassicas and strawberries (Anon. 1984; Vernon *et al.* 2000). During summer the leatherjackets pupate within the soil and adults emerge in August and September that year, but then live for only a few days (Figure 1-2).

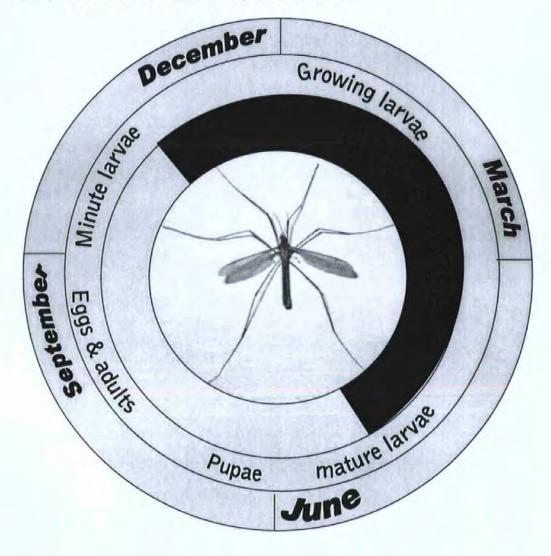


Figure 1-2 From Anon. (1985) diagram of *T. paludosa* cycle. Photo of adult male *T. paludosa* in centre

Although less common than *T. paludosa*, *Tipula oleracea* has been identified as a significant pest in parts of the UK. It is a bivoltine species (having two generations per year) with one emergence period in spring and one coinciding with that of *T. paludosa* (Coll 1996).

young/small wireworms may do little damage, whereas a medium population consisting primarily of large wireworms may do considerable damage.

1.2 Leatherjacket Biology and Ecology

1.2.1 Leatherjacket life cycle

Leatherjacket biology and ecology has been reviewed in detail by Blackshaw and Coll (1999). Leatherjackets are the larvae of crane flies, *Tipula paludosa* Meigen and *Tipula oleracea* L. (daddy-long-legs). *T. paludosa* females typically produce about 350 black, oval eggs, which are laid in the soil in late August and September (Coulson 1962). Where light traps have been used in South Devon, peak numbers of females have been caught between 15th September and 9th October between 1967 and 1974 (Mayor and Davies 1976). The eggs hatch in about 14 days to be small legless grubs. The overwintering leatherjacket larval stage lasts 25 weeks (Laughlin 1967) (Figure 1-1).



Figure 1-1 Leatherjacket: larva of Tipula spp.

Although leatherjackets feed all winter, their main growth period is in spring, when they grow to about 4cm long (Coll and Evans 1999; Laughlin 1967). They are commonly found in grassland and feed on roots and below ground shoots of winter cereals and grass (especially clover) (Anon. 1984; Blackshaw 1984; 1990), and also brassicas and strawberries (Anon. 1984; Vernon *et al.* 2000). During summer the leatherjackets pupate within the soil and adults emerge in August and September that year, but then live for only a few days (Figure 1-2).

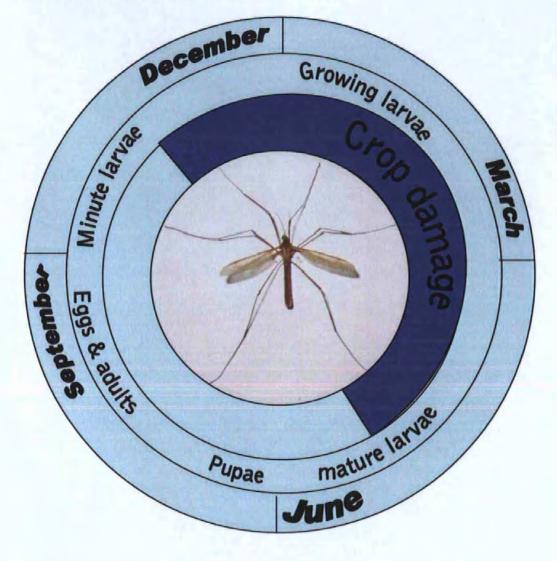


Figure 1-2 From Anon. (1985) diagram of *T. paludosa* cycle. Photo of adult male *T. paludosa* in centre

Although less common than *T. paludosa, Tipula oleracea* has been identified as a significant pest in parts of the UK. It is a bivoltine species (having two generations per year) with one emergence period in spring and one coinciding with that of *T. paludosa* (Coll 1996).

1.2.2 Distribution and factors affecting distribution of leatherjackets

Leatherjackets are found throughout the UK in agricultural and amenity land. There is consistency of population from year to year, McCracken (1995) found the leatherjacket population of the field in the previous year was correlated with leatherjacket numbers. This is probably due to the females being hardly able to fly when fully gravid at emergence and thus laying eggs close to the point of emergence (Frouz and Paoletti 2000; Pritchard 1983).

Many studies have looked at the relationship between environmental variables and leatherjacket numbers (see Table 1-3). The most comprehensive, recent study was McCracken *et al.* (1995), who recorded a large number of environmental variables when carrying out a survey of leatherjacket populations. Some of these variables individually had a statistically significant effect on leatherjacket numbers. Others were not individually statistically significant (organic & inorganic fertiliser, grass height, altitude, grazing, age of pasture, size of field, drainage, weediness, % loss on ignition, pH, P, K, salinity, bulk density, organic fertiliser and grass height) on leatherjacket populations. When the seven most influential variables were combined, they accounted for 50% of the total variability in leatherjacket numbers. This model was then used for predicting leatherjacket numbers, and although it did not work as 'an effective predictive model' on this occasion, it suggested that the approach had potential.

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Table 1-3 Factors affecting leatherjacket populations

Factor Affecting Population	Study	Effect on populations	
Nitrogen	M ^c Cracken <i>et al</i> . (1995)	Slurry and dung application increase LJ populations.	
Silage	Purvis and Curry (1981)	Fewer leatherjackets found in fields cut for silage than in grazed fields.	
Distance from the Atlantic ocean	M ^c Cracken (1995)	The distance of a field from the Atlantic ocean had a statistically significantly effect on leatherjacket numbers.	
Rainfall	Mayor and Davies (1976)	Low rainfall at time of egg and first instar (autumn) is a major cause of leatherjacket mortality.	
	Mayor and Davies (1976)	There was a relationship between the mean population (1963-1974) in Gloucestershire, Somerset, Wiltshire and Dorset and the percentage of average rainfall for September and October. The populations for Devon and Cornwall did not relate to rainfall.	
	Blackshaw (1983a)	Negative correlations between leatherjacket populations and rainfall in July, August and November.	
	Milne <i>et al</i> . (1965)	Low rainfall reduces egg hatching (16.2% at lowest rainfall equivalent compared to 84.8% hatch) and leatherjacket survival (24.6% at high rainfall and 7.4% at low rainfall).	
Aspect	M ^c Cracken (1995)	N, E, S and W aspects are statistically significantly different in their effect on leatherjacket numbers.	

Natural enemies	Anon (1984)	Birds (Rooks, Starlings, Lapwings, pheasants, Blackheaded and Common gulls), and mammals (moles, shrews, yellow-necked wood mouse were all found to predate leatherjackets (Kelly 1989).
Soil Moisture	Coulson (1962)	Eggs and leatherjackets are susceptible to desiccation and are therefore more likely to be found in damp soils in wet conditions.
	M°Cracken (1995)	Fields that had a tendency to waterlogging had more leatherjackets. Waterlogging may be a subjective measurement.

1.3 Wireworm Biology and Ecology

1.3.1 Wireworm lifecycle

Wireworms are most commonly found in undisturbed permanent grassland, although they have been reported in fields with no history of long-term grass (Fox 1961; Parker and Howard 2001; Parker and Seeney 1997). Wireworms are the larvae of click beetles (*Agriotes* spp.) (family: Elateridae). There are three species of click beetle commonly found in Britain whose larvae cause the majority of pest damage: *Agriotes sputator* L., *Agriotes lineatus* L. and *Agriotes obscurus* L. (*Furlan et al. 2001b*). The three species are not always all found together and not all species are found all over the UK (Parker and Howard 2001). Generally, click beetle females lay their eggs in the soil in May and June and the wireworms hatch about 1 month later. At hatching the wireworms are less than 1.5mm long and transparent white. They live in the soil for about 4-5 years and as they grow their colour darkens to a brown/orange. Young (<one year old) wireworms and adult click beetles are not believed to have a significantly damaging effect on crops (Parker and Howard 2001). They feed most intensely in two periods each year, March-May and September and October. In July and August the wireworms pupate. It takes between three and four weeks for the wireworms to become adult click beetles, they then continue to hibernate underground until their emergence in the following spring.

Many aspects of the biology of *Agriotes ustulatus* Schäller (Furlan 1998) and *Agriotes sordidus* (Furlan 2004) have been studied in detail. However, many details of the lifecycles of the main UK species (A. *lineatus, A. obscurus* and *A. sputator*) remain elusive. For example, little is known about adult survival times and the prevalence of overwintering; lifecycle stage duration, larval migration, adult habitat, mating, natural mortality rates, female fecundity or oviposition period or factors that influence larval development. It is also not known how far and by what mode of transport adults migrate (Parker and Seeney 1997), it is believed that click beetles will fly only very rarely. Knowledge of these factors is important when considering potential control options.

1.3.2 Distribution and factors affecting distribution of wireworm

The wireworm survey of England and Wales 1939-1942 was the most detailed survey of wireworm populations in agricultural land, sampling 16,000 fields (roughly 160,000 acres) (Anon. 1948). There was found to be a marked variation in abundance across the UK (increasing from west to east) and from field to field. The survey identified a strong association between population densities in fields that were geographically close but a stronger association between fields that were under the same management. Lefko (1998) found that wireworm populations were significantly affected by historical management practices. This indicates that it may be possible to manipulate wireworm densities by changes in management practices.

Ester *et al* (2004b) trapped adult click beetles using pheromone traps in the Netherlands (1999-2003). They found large numbers of *A. lineatus* and *A. obscurus* in agricultural fields of seed grass, summer barley, alfalfa and grass clover. They also found these two species in grass roadside verges along with *A. sputator*. They concluded that *A. sputator* is not a major pest in the Netherlands. The same researchers noted that trap catches were depressed when the weather was wet, and that beetle catches were more plentiful in the evenings than during the day.

There is evidence that wireworm densities are affected by chemical and environmental variables, such as soil moisture and altitude (Table 1-4). However many of these studies are contradictory, with effects sometimes shown on wireworm densities and sometimes not. For example studies appear to show that wireworms avoid both high and low moisture. It is possible that there is an optimum soil moisture for wireworms and they will avoid soil either dryer or moister than the optimum.

Factor affecting populations	Study	Effect on wireworm population
Altitude	Parker and Seeney (1997)	No significant difference between altitude of infested and uninfested fields, but there was a trend for southern slopes to be infested more often than north facing slopes ² .
	Erichsen-Jones (1944)	More <i>A. obscurus</i> adults found as altitude increases, fewer <i>A. sputator</i> and no change in adult <i>A. lineatus</i> adults.
Soil Type	Nadvornyj (1968)	Wireworms prefer heavy soils.
	Anon. (1983)	Fewer wireworms on light soils.
	Anon. (1948)	Light soils have lower populations ¹ .
Sand, silt & clay content	Ross <i>et al</i> (1948)	High sand is associated with successful crops in the presence of wireworms ¹ .
		No association with silt & clay ¹ .
	Parker and Seeney (1997)	No significant difference between %sand, %silt or %clay of infested and uninfested fields².
CaCO,	Ross <i>et al</i> (1948)	Crop failures associated with higher CaCO ₃ ¹ where wireworm densities were high.
Bulk Density	Parker and Seeney (1997)	Infested sites had a significantly lower bulk density than uninfested fields.
Organic Matter (OM %)	Ross <i>et al</i> (1948)	No relation between OM% and the incidence and severity of wireworm attacks ¹ .
	Parker and Seeney (1997)	No significant difference between OM% of infested and uninfested fields ² .
	Ibbotson (1958)	The species of each wireworm was ascertained wherever possible and A . <i>lineatus</i> was the dominant species where the % loss on ignition was high (58.9- 10.1), A . obscurus was the dominant where the % loss on ignition was lower $(7.6-8.7)^2$.

Table 1-4 Factors known to affect wireworm populations

Soil Moisture	Campbell (1937) (<i>Limonius</i> californicus)	Larvae occupy soil with moisture 9-12%.
	Lees (1943) (unspecified <i>Agriotes</i> species)	Wireworms avoid dry soil in laboratory experiments- effect on population unspecified.
	Lefko (1998)	The greater the number of days of high soil moisture the less suitable the habitat is for wireworms.
рН	Parker and Seeney (1997)	No significant difference between pH of infested and uninfested fields ² .
	Yates and Finney (1942)	See sand, silt & clay content.
	Ibbotson (1958)	The species of each wireworm was ascertained wherever possible and <i>A</i> . <i>lineatus</i> was the dominant species where the pH was high, (5.9-7.4). These were plots which had received basic slag annually for 40 years. <i>A. obscurus</i> was the dominant where the pH was lower (5.5) ² .
	Ladell (1938)	No data given, but observed that there was no relationship between pH and the number of wireworms.

 1 No evidence of statistical analysis or data provided; results show only suggestions of significance 2 P-values not given

1.4 Control Methods

1.4.1 Chemical control

Before the advent of broad spectrum insecticides, such as DDT and aldrin in the 1940's, soil pests were a significant problem. Since then and until recently, insecticides have been available and have minimised crop losses due to soil pests. These insecticides have often been responsible serious negative impacts on populations of non-target organisms, human health and on the general environment due to their persistency. The recent demand for organic produce and tighter environmental legislative controls on pesticides has reignited a requirement for alternative, non-chemical methods to control subterranean pests (Kuhar *et al.* 2003).

Future pest control is being directed by a different collection of external forces and is unlikely to be defined by the introduction of new chemicals. Two of the major drivers for future pest control are:

- 1. changes in environmental regulation legislation
- 2. rise in organic production systems

Both of these require 'non-chemical' solutions to pest control.

1.4.1.1 Insecticide withdrawal

The World Health Organisation has calculated that pesticides cause 20,000 deaths per year (Ahmad 2002) although others place this much higher (Jeyaratnam 1990). There is global pressure for a reduction in pesticide usage, both environmental pressure against harmful chemicals and consumer pressure on health grounds (particularly within Europe). Both new European and UK legislation is requiring the withdrawal of a number of well used pesticides, often with no efficacious alternatives. Chemicals such as dieldrin (withdrawn 1981), aldrin (withdrawn 1991), Lindane (withdrawn 2001) and methyl bromide, previously used to control soil pests (Dewar *et al.* 2001; Hancock *et al.* 1986; Thomas 1996) are no longer permitted in the European Union under Council Directive 79/117/EEC. Whilst some alternatives have been found (see Table 1-5 and Table 1-6) they are often less effective (Dewar *et al.* 2001; Hancock *et al.* 1986) and there are fewer. Ethoprophos is the only pesticide effective against wireworms that is permitted in Holland (Ester *et al.* 2004b). The Stockholm Convention on Persistent Organic Pollutants which was completed in 2001 and came into force in 2004, outlaws or strongly limits the use of a number of the most polluting chemicals globally, although, the chemicals still permitted include organophosphorus insecticides (Ethoprophos, Fosthiazate, & Chlorpyrifos). There are now many vulnerable crops in the UK which have no suitable pest control agents for leatherjackets and/or wireworms such as strawberries and brassicas.

Crop	Recommended Pesticide 1996	Recommended Pesticide 2000	Recommended Pesticide 2004	Recommended Pesticide 2005
Potatoes	Ethoprophos	Ethoprophos	Ethoprophos	Ethoprophos
	Phorate	Phorate		
			Fosthiazate	Fosthiazate
Maize/ Sweetcorn	Gamma-HCH	Gamma-HCH		
	Bendiocarb			
Strawberries	Gamma-HCH	Gamma-HCH		
Grassland	Gamma-HCH	Gamma-HCH		
Brassicas	Chlopyrifos + dimethoate			
	Gamma-HCH			

Table 1-5 Decline in the availability of products for the chemical control of wireworm over ten years

Information from The UK Pesticide Guides (Whitehead 1996; 2000; 2004; 2005). HCH = Hexachlorocyclohexane

Crop	Recommended Pesticide 1996	Recommended Pesticide 2000	Recommended Pesticide 2004	Recommended Pesticide 2005
Brassicas	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos
	Gamma-HCH			
Potatoes		Methiocarb	Methiocarb	Methiocarb
Leys	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos
		Methiocarb	Methiocarb	Methiocarb
	Triazophos			
Permanent	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos
Pasture	Gamma-HCH	Gamma-H <u>.</u> CH		
	Triazophos			
Amenity grass	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos	Chlorpyrifos
	Gamma-HCH	Gamma-HCH		
Maize/ sweetcorn	Gamma-HCH	Gamma-H <u>.</u> CH		
		Fenitrothion		
Strawberries	Gamma-HCH	Gamma-HCH		
Peas	Chlorpyrifos	Chlorpyrifos		

Table 1-6 Decline in the availability of products for the chemical control of leatherjackets over ten years

Information from (Whitehead 1996; 2000; 2004; 2005)

1.4.1.2 Organic Farming Requirements

Recent years have seen a change in British consumer attitudes towards food safety; pesticides are viewed by consumers as potentially harmful substances that are detrimental to health (Baker *et al.* 2004; Chinnici *et al.* 2002; Makatouni 2002). This has led to a commercial surge in the demand for organic produce. The UK agriculture industry adds £6.6 billion to the UK economy annually (Defra 2004). The organic sector is a specialised sector of this market which is expanding rapidly. UK organic sales are now close to £2 billion annually and are increasing by an average of £7 millon per week (Soil Association 2007). This is a small but high profile and rapidly increasing proportion of the market.

The Compendium of UK Organic Standards (Defra 2004b) states that:

'Organic production systems are designed to produce optimum quantities of food of high nutritional quality by using management practices which aim to avoid the use of agro-chemical inputs and which minimise damage to the environment and wildlife.'

Thus the pesticides available to conventional farmers are unavailable to organic farmers and alternative pest control methods are required. Increased consumer pressure for pesticide reduction has led to a change in the attitudes of the whole food industry (Brouwer and Bijman 2001; Levidow and Bijman 2002), leading to a general pressure to reduce pesticide inputs in all food production systems.

1.4.2 Current cultural control methods

Although the control of pests using pesticides is the principal choice of farmers and growers it has a number of inherent drawbacks. The most familiar drawback of pesticide usage is toxic residues. But the development of pest resistance and secondary pests filling the niche of the 'controlled' pest are also problems (Lewis *et al.* 1997). Alternative pest control measures may be employed, these include cultural

and biological methods. Cultural control methods may be any modification to the environment of a pest, which is introduced to have a detrimental effect on the population of that pest, for these soil pests this may include crop rotation and avoidance amongst others. Biological control strategies suppress pest populations with biological agents; these may be pathogens, predators or parasites. Many biological control methods such as infection of the pest with a mortality-inducing disease would satisfy stringent organic guidelines by having no toxic residues, but would still allow pest resistance, secondary pests and pest resurgence. Cultural methods are not generally associated with any of these problems.

The Soil Association has issued a briefing paper on the control of leatherjackets in organic systems (Soil Association 2003). This mainly draws on cultural methods of control. The recommendations have, in the main, been drawn from general pest control strategies. Avoidance of wireworm damage is primarily achieved by the avoidance of planting in wireworm-infested fields (Parker and Howard 2001), especially avoiding planting susceptible crops as the first crop after a permanent pasture field.

Flooding is being investigated as a potential control option in California and Canada (van Herk and Vernon 2006), but is unlikely to be applicable in this part of the UK as the area is primarily hilly ground (see figure 2-6 for example) and water flows downhill.

1.4.2.1 Crop rotation

Crop rotation is of great importance for organic growers. It involves changing the crop planted in an area every year or few years in order to maintain fertility or break pest population build-up (most notable in brassicas). As discussed in sections 1.2.2 and 1.3.2, grass duration has a considerable effect on the populations of both wireworms and leatherjackets. Wireworm populations build up over a period of years in grass and populations are often highest in permanent pasture (Jedlicka and Frouz 2007; Strickland et al. 1962). Thus, it is not advisable to plant wirewormsusceptible crops immediately after cultivation of a permanent pasture field. It has been suggested that seeds in permanent pasture should be planted as soon as possible so that wireworms are still eating inverted turf whilst germination occurs (Salt and Hollick 1944). In contrast, the effect on leatherjackets is less clear cut. McCracken (1995) found that populations were unaffected by duration of grassland and yet the number of cuts of silage taken from Scottish fields in the previous three years had a statistically significant effect on leatherjacket numbers. Blackshaw (1988) found cereal crops are only at risk of leatherjacket infestation (above economic thresholds) if they follow grassland in a rotation.

Evidence suggests that the abundance of soil pests is greatly influenced by previous crops (Seal *et al.* 1992a). Schepl (2004a) found that including red clover or broad beans in a crop rotation caused more damage to potatoes by wireworms than if peas or lupins were included (either with or without clover/grass ley). Frost *et al.* (2002) suggested that mustard reduces wireworm populations, although their reported reduction was not statistically significant it does suggest opportunities for further investigation.

1.4.2.2 Avoidance

Avoidance covers a variety of tactics employed by growers, from not ever growing a vulnerable crop, to adjusting planting and harvesting dates. Seventy five per cent of growers surveyed by Peacock and Norton (1990) had stopped or were about to stop growing one or more crops that had suffered serious pest problems in the past. Peacock and Norton (1990) found this form of avoidance was the growers' most common adaptation method to pest attack. One widely used method of cultural control of wireworm damage in potatoes is to lift the crop as early as possible (Schepl and Paffrath 2004b).

Monitoring of pests is an important factor when considering using avoidance tactics. However, this is particularly difficult with soil pests due to the level of effort involved sampling the subterranean environment. Sampling before planting and then not planting if populations are present is the most effective way of avoiding wireworm damage (Parker and Howard 2001). For wireworm bait traps (see 3.2.1 for description) the relationship between larvae found when monitoring is very prone to error when predicting populations, so finding no wireworms in traps is not always sufficient to reassure the grower that there is no risk of damage. Pheromone traps can be used in the UK to capture the click beetle males of the three major wireworm pest species (*A. lineatus, A. sputator* and *A. obscurus*) using synthetic analogues of female sex pheromones (Furlan *et al.* 2001b; Toth *et al.* 2002). However, there is as yet no reliable way of relating trap catches to crop damage.

A more efficient approach to growing susceptible crops would be to have an accurate prediction system that was reliable enough to provide all the information required for

effective avoidance. This prediction system may be on a number of scales, regional or sub-field, or may be spatial or temporal.

1.4.2.3 Soil health

Peacock and Norton (1990) state that for almost all growers, their major defence against losses to pests is growing healthy plants that are able to withstand attack. Organic growers rely on crop rotation and not artificial fertilisers to build fertility in the soil. There is a belief that organic soil is inherently more 'healthy'. Healthy soils encourage healthy plants, and healthy plants are believed to be more resistant to pests and diseases (Peacock and Norton 1990; Soil Association 2003). Good soil biology and high organic matter content lead to a low abundance of pests and disease (Chau *et al.* 2005), whereas nutrient imbalances caused by excessive use of inorganic fertilizers can lead to lower pest resistance (Altieri and Nicholls 2003).

Although not compatible with organic production, added Nitrogen can compensate for crop losses due to pests in grassland (Blackshaw and Newbold 1987; French *et al.* 1990) and well-fertilised plants can grow away from pest damage. However, McCracken (1995) found that the application of slurry and dung increased leatherjacket populations, which they suggested may be as a result of increased nutrient levels.

1.4.2.4 Cultivation

The act of cultivating a field is known to reduce the populations of leatherjackets and wireworms (Blackshaw 1988; Frouz and Paoletti 2000; Jedlicka and Frouz 2007; Parker and Howard 2001; Seal *et al.* 1992a). Leatherjackets can be controlled by

timing of ploughing; the land should be ploughed in July or early August before the main egg laying period (Anon. 1984). Rolling is recommended to reduce leatherjacket damage (see section 5.1). High populations of *T. paludosa* are found in spring barley crops that are grown in fields that are ploughed grassland (Anon. 1984; Blackshaw 1988). It was also shown in these studies that the greater the number of successive years of cultivation, the lower was the population of leatherjackets in the emerging spring barley crop.

1.4.2.5 Weeds

There is evidence that wireworms prefer to eat weeds rather than potatoes or maize (C. Pazmandi pers. comm.). Higher populations of wireworms were found in weedy fields than in weed-free fields (Peacock and Norton 1990; Salt and Hollick 1944; Seal *et al.* 1992a), although populations of wireworm-consumers (Staphylinids and Carabids) are also increased in weedy fields (Anderson 2003). It has been found that many growers use mulches as a preventative measure against pests (Peacock and Norton 1990), however, fleece has been observed to exacerbate problems if pests are present (John Richardson pers. comm.)

1.4.2.6 Biological Control

There are currently few suggested methods of biological control for wireworms. *Metarhizium anisopliae*, an entomopathogenic fungi, has been investigated and produces a high mortality in greenhouse and laboratory studies (Kabaluk *et al.* 2006); however it still requires development before field usage. Some species of nematodes, such as *Steinernema feltiae*, have been found to effectively control T. *oleracea* and *T. paludosa* populations (Peters and Ehlers 1994) and are commercially available but not economically viable for commercial use.

1.4.2.7 Arthropod Biodiversity

Organic farms often report fewer pest problems than conventional farms. There is an implication that reduced insecticide usage or increased maintenance of field margins, increases the predatory beetle population. These predators then act to keep the pest population in check. One study that has shown a connection between the arthropod community and levels of pest damage in a crop is by Letourneau and Goldstein (2001). When conventional and organic tomato production in California was compared, there was no significant difference between the level of damage to foliage or fruit. The authors then examined the diversity of arthropod communities and found there was a greater abundance of natural enemies and a greater diversity of all functional groups in the organic systems. In contrast, Armstrong (1995) found that the abundance and diversity of carabid beetles was greater on the fields under conventional management than under organic management. However this study was only carried out over a one year period and covered only four fields. There is some evidence that Carabidae and Staphylinidae consume wireworms (Fox and MacLellan 1956) so an increase in these may cause a reduction in wireworms.

1.5 Future work

Lewis *et al* (1997) have suggested that the three future avenues of investigation for pest control should be (i) ecosystems management; (ii) crop attributes and multitrophic level; and (iii) therapeutics with minimal disruptions (treatments for pest control that cause little damage to non-target organisms). This thesis focuses primarily on aspect (i) – ecosystems management, and the crop attributes aspect of (ii). As recommended by Lewis *et al* (1997), the focus of this investigation will be on a small number of locations all being managed in different ways to achieve similar commercial objectives namely, seven farms located in South Devon, between what is now known as Haldon Forest Park and the River Erme, (Figure 1-3). This is in contrast to previous reductionist work which tended to focus on one pest in a range of geographic environments (national or international) and then try to find a 'magic bullet' to destroy the pest in all situations (Lewis *et al.* 1997). Future pest control methods are likely to involve the entire ecosystem surrounding the pest; predators, landscape, management and features to maintain populations at a non-damaging level. This approach to pest control is impossible to implement without a detailed knowledge of the ecology of the pests.

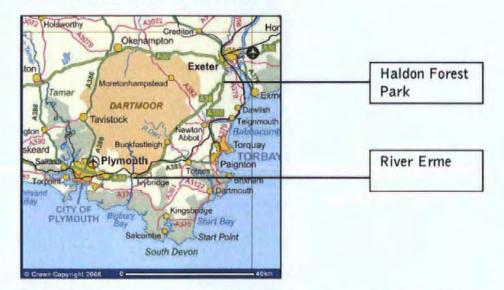


Figure 1-3 Map of South Devon, UK: the area containing all farms used for fieldwork in this study. Reproduced from Ordnance Survey map data by permission of Ordnance Survey.

This work aims to identify the factors which encourage high wireworm and leatherjacket populations in an area of South Devon and to investigate the effect of scale on the distribution of these pests. Cultural methods for inhibiting high populations of soil pests, such as, crop rotations and cultivation methods, will be studied; this is a gap in current knowledge highlighted by Parker and Howard (2001). Parallel to exploring methods of cultural control of soil pests, other aspects of their ecology will be studied, namely, the distribution of wireworms and leatherjackets at a number of scales and the movement and density of click beetles around pheromone attractants.

Prediction of damage, by estimating populations of leatherjackets and wireworms remains a slow and expensive process, despite many years of investigation. Sampling methods are not currently reliable enough to provide a definitive assessment of damage risk for any particular crop. In order to solve this problem, either a more thorough knowledge of larval distribution or a more thorough understanding of population dynamics is required. This may include factors like dispersal and field characteristics to more accurately predict and monitor populations.

This report will look in depth at the distribution of leatherjackets and wireworms (and their adult stages) to try to identify which environmental variables control distribution and whether any can be manipulated as a form of cultural control. Chapter two will observe the effect of environmental variables on click beetle and crane fly population sizes at a landscape scale. Chapter 3 will then focus on how the distributions of larvae at the field scale. Chapter four will look at a large number of environmental variables and how they influence both larval and adult distributions. Chapter five will begin to assess the effects of manipulation of environmental variables of larvae. Chapter 6 will characterise pheromone traps for UK click beetles.

Chapter 2 Investigation of landscape factors influencing the distribution of four pest species in the UK

2.1 Introduction

At the landscape scale, the spatial distribution and factors affecting the distribution of three UK pest species of click beetles (wireworms) and crane flies (leatherjackets) are poorly understood. Consideration of spatial scale is crucial when determining the distribution of pest organisms (Dungan et al. 2002; Schweiger et al. 2005; Vialatte et al. 2007). Previous work on the distribution of pest populations has often either been on a national/international scale (Anitha et al. 2006; Copeland et al. 2006; Furlan et al. 2001b; Kudryavtsev et al. 1993) or more often, on a field scale (Ferguson et al. 2006; Hofs et al. 2006; Strickland et al. 1962; Vialatte et al. 2007). Large scale (national/international) studies are often appropriate for highly mobile pests, such as moths or aphids, and field scale studies for those and other relatively immobile pest species such as nematodes and other soil pests. However, in a heterogenous landscape typical of organic vegetable production rotations, factors affecting distribution are present primarily at landscape scale. The factors affecting the distribution of leatheriackets at national scale are known to be different from those at field scale (Blackshaw 1983a). Although the wireworms and leatherjackets. are believed to be relatively immobile, the adults are capable of moderate-scale dispersal (although that of click beetles is currently uncertain). Different factors affecting insect distribution work at different scales. Knowledge of crane fly and click beetle distribution at landscape scale should highlight the factors that affect distribution at this scale.

In 'Wireworms and Food Production' (Anon. 1948) almost half of the beetles captured nationally were *A. obscurus*, and there were more *A. lineatus* than *A. sputator*. The exception to this was in the area between the Haldon Forest Park and the Erme valley (Figure 1-3) where about half of the click beetles were *A. sputator* and the other two species were found in approximately equal numbers. This is the area where this fieldwork took place. Furlan *et al* (2001b) found that *A. obscurus* click beetles have a high population in the South West of England, whereas *A. lineatus* and *A. sputator* have medium level populations. In the North and the West of England, *A. obscurus* populations were medium and *A. obscurus* and *A. lineatus* were low. For this study, only five fields were sampled over the whole of the UK and so may not reflect populations accurately and the scale over which this changes is completely unknown. Gouch and Evans (1942) found that ratios of *A. sputator* to *A. obscurus* were approximately 20:1 and 1:25 in two fields only 400yrds (366m) apart.

Grass is known to encourage wireworm populations to develop (see section 1.3.2) (Fox 1961; Parker and Howard 2001; Parker and Seeney 1997; Strickland *et al.* 1962). Due to their long lifecycle, high populations are typically found in permanent pasture. Although, little is known about click beetle population variation from year to year. Gouch & Evans (1942) observed *A. obscurus* click beetles selectively lay on grass in preference to wheat, clover, kale or bare earth.

Wireworms migrate to suitable moisture conditions (Campbell 1937; Lafrance 1968; Lees 1943). Rainfall has been observed to have a major influence on leatherjacket densities, both positive and negative (Blackshaw 1983a; Mayor and Davies 1976; Milne *et al.* 1965). By studying the densities over such a restricted area the effect of rainfall should be minimised. Moisture has a marked effect on the survival of

leatherjackets (Coulson 1962; McCracken *et al.* 1995). If the larvae of both pests are affected by moisture one would expect that the distribution of adults would show some variation dependent on water. However, as water content of the soil is highly variable due to time of year and weather conditions, distance from watercourse can be used as a more consistent measure of waterlogging and soil moisture retention. It also has the advantage of not requiring specialist equipment for measurement by farmers and growers.

The proportion of *T. paludosa* to *T. oleracea* crane flies is not known in South West England but annual leatherjacket surveys show large-variation in larval numbersfrom year to year (Mayor and Davies 1976). Crops are also known to influence crane fly species ratios (*T. oleracea* populations can build up under oil seed rape (Coll 1996)). The nature of vegetable cultivation requires many patches of bare earth between rows and between crops. Reduction in leatherjacket population due to mechanical cultivation should show reduction in adult populations as there will be fewer juveniles to mature into adults (Blackshaw 1988; Seal *et al.* 1992a).

The aims of this study were:

- a. To assess the distribution of click beetles and crane flies on a landscape scale.
- To investigate the local geographic factors which may be influencing the distribution of the pest species.

2.2 Methods

The study area in South Devon, UK was selected as it provided an opportunity to study variation in population levels across an organically farmed landscape. This landscape provides substantial changes in elevation, land use and cropping patterns, all within a compact area (Figure 2-2).

Pheromone traps for click beetles were used in three transects were across this landscape. Traps along two transects were sampled each year (Figure 2-2), foe two years. Transect C, traps 19-26 (0.7km long), running North-South was sampled in 2005; transect A, traps 1a-18a (1.7km long), running East-West, were sampled in 2005 and 2006 and transect B, traps 1b-18b (1.7km long), also running East-West, were sampled only in 2006. Initially, transects A and B, arranged perpendicularly to one another were used to as this was thought to provide maximum data; subsequently, the variation within 100m was found to be the most useful, therefore, in the second year transect B was added, parallel to transect A to increase the number of pairs of trap 100m.apart. Transect C was not available for use in 2006.

Three pheromone traps (Plant Prot. Inst. Hung: Acad. Sci., Budapest, Pf 102 H-1525) (Figure 2-1) each containing pheromone for one of the three species of click beetles (Furlan *et al.* 2001a) were placed at 100m intervals along each transect for their emergence period (May-July). Trap manufacturers provide the pheromones geranyl octanoate for *A. lineatus*, geranyl-butanoate for *A. sputator* and geranyl hexanoate plus geranyl octanoate for *A. obscurus* pre-infused into a removable plastic vial. The traps were emptied weekly and the click beetles identified to species. All

catches were stored at -20°C within 12 hours of sampling. The pheromones were replaced after six weeks.

Pheromone traps can interact with each other and the position of the trap in a line in relation to wind direction can affect the number of insects it captures (Wall and Perry 1981). Catch data were checked for any gross effect of interaction (Wall and Perry 1981) between traps and none was present (this conforms to expectations arising from conclusions in Chapter 6).

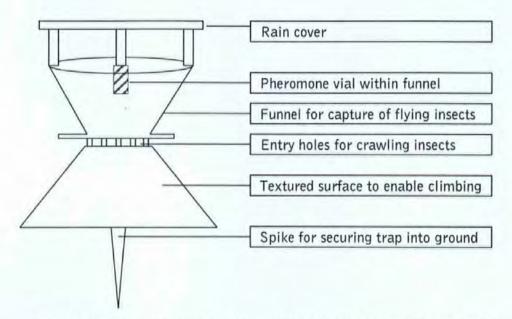


Figure 2-1 Pheromone trap (Yatlor) used to attract click beetles. A speciesspecific pheromone was placed in a vial at top of funnel.

Crane flies were sampled using water traps, consisting of a green plastic bowl, 29cms diameter placed on the ground and filled with water to which about 3ml of liquid detergent was added to reduce surface tension (Blackshaw 1983b). The water traps were placed in the same positions as the pheromone traps for the period of crane fly emergence (August – October). The traps were emptied weekly and the pest species were identified (La Gasa and Antonelli 1999). All catches were stored at -20°C within 12 hours of sampling.

Cumulative catches and cropping history were then stored on a database in ESRI ArcMap 9.0 which provides an interactive base map containing data from the Ordnance Survey obtained via Edina Digimap.

2.2.1 Spatial distribution

The presence or absence of spatial structure was examined within ArcGIS using Moran's I, a measure of spatial autocorrelation (Cocu *et.al.* 2005; Diniz-Filho and Telles 2002; Moran 1950). This index can identify population structure as dispersed, random or clustered. Then the accepted convention of studying soil pests on a field scale was assessed by examining within and between field differences. The withinfield and between field variation in the populations were compared by taking every pair of traps 100m apart and calculating the ratio of the difference in catch over 100m (i.e. smallest catch of pair divided by the largest catch of the pair), this enable the differences between pairs to be compared irrespective of insect density. The ratios for the pairs of traps within the same field were compared with those for the pairs in different fields using a two sample t-test. There were 30 'within field' pairs and 31 'between field' pairs. The data were tested for normality and were log transformed or square root transformed as necessary, variances were homogenous.

2.2.2 Annual variation

A paired t-test was used to compare the cumulative populations for 2005 and 2006 per trap for both click beetles and crane flies for locations 1a – 18a (transect A). The data were tested for normality and were log transformed or square root transformed as necessary, variances were homogenous.

2.2.3 Land use

To test the effect of land use on click beetle and crane fly numbers, the locations of the traps were defined as either permanent pasture (grass unploughed for more than ten years), ley (grass in rotation, ploughed more recently), vegetables (cultivated and planted with vegetables in rotation), permanent vegetables (herbs, vegetables and soft fruit not in a rotation) and not cultivated (farmyard etc). All trap catches were then analysed with a one-way ANOVA which tests whether the means of all populations are equal. The data were tested for normality and were log transformed or square root transformed as necessary, variances were homogenous.

2.2.4 Distance from watercourse

A watercourse was defined as a stream or reservoir (ditches were excluded as there was low rainfall in the period of sampling and therefore water presence could not be guaranteed). The numbers of click beetles and crane flies collected were correlated using a Pearson Product-moment correlation, against the distance from the nearest watercourse to the trap location. All data were log transformed to achieve normality.

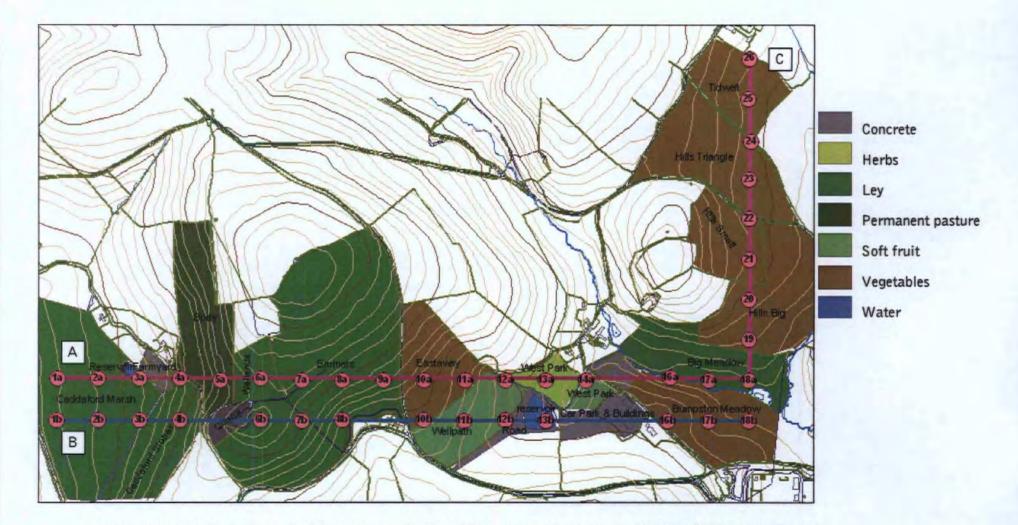


Figure 2-2 Locations of traps in three transects (A, B and C) at 100m intervals, also showing field boundaries and names, crop types, watercourses and contours.

2.3 Results

2.3.1 Spatial distribution

Total cumulative click beetle catches for 2005 and 2006 are displayed in Figure 2-3 and Figure 2-4 respectively. The relative proportions of the three beetles species showed considerable variation in numbers within 100m (Figure 2-5).

Moran's I scores of spatial aggregation showed that the distribution of the insects studied was rarely random and showed highly significant clustering for all but *A*. *lineatus* in 2005 (Table 2-1). For all other species there is a high degree of clustering, however, this index does not describe any aspects of that clustering. A trap is more likely to have a high catch if the traps adjacent to it also record high catches, and equally, low catches can also be predicted in this way.

Species	Year	Moran's I Index	Distribution	P Value
A. sputator	2005	0,06	Clustered	< 0.01
A. sputator	2006	0.23	Clustered	< 0.01
A. lineatus	2005	-0.03	Random	>0.10
A. lineatus	2006	0.14	Clustered	< 0.01
A. obscurus	2005	0.14	Clustered	< 0.01
A. obscurus	2006	0.02	Clustered	<0.10
Male <i>T. paludosa</i>	2005	0.3	Clustered	< 0.01
Male <i>T. paludosa</i>	2006	0.18	Clustered	< 0.01
Female T. paludosa	2005	0.29	Clustered	<0.01
Female T. paludosa	2006	0.07	Clustered	< 0.01

Table 2-1 Moran's I scores for insects captured along a transect in 2005 and 2006.

There was a general trend for the difference within fields to be less than the difference between fields (Table 2-2). However, when this observation was tested (T-

test) by comparing the means of the two groups the results are only significant for *A*. *lineatus* (Table 2-2).

There were no *T. oleracea* caught throughout this study. The cumulative weekly catches of crane flies show considerably less variation (Figure 2-6 and Figure 2-7) over the length of the transect than can be seen in the click beetle data. However, there is still a marked difference between the highest and lowest catches. Male and female catches were similar in every trapping location. In addition, crane fly catches are similar within a single field but there is not a statistically significant difference when comparing the catch ratios between and the within-fields (Table 2-2).

Table 2-2 Comparison of the catch ratios for pairs of traps within and between fields

	Between	Between Fields		ields		
	Mean	SD	Mean	SD	Т	Р
A. lineatus ¹	0.5072	0.2781	0.6201	0.2400	6.33	0.000
A. obscurus	0.5548	0.3010	0.5861	0.2783	0.42	0.675
A. sputator ¹	0.4208	0.2835	0.4117	0.2750	-0.13	0.899
Male <i>T. paludosa</i> ²	0.489	0.290	0.589	0.323	1.20	0.234
Female <i>T. paludosa</i> ²	0.489	0.337	0.580	0.322	1.06	0.295

¹ Log transformed

² Sgirt transformed

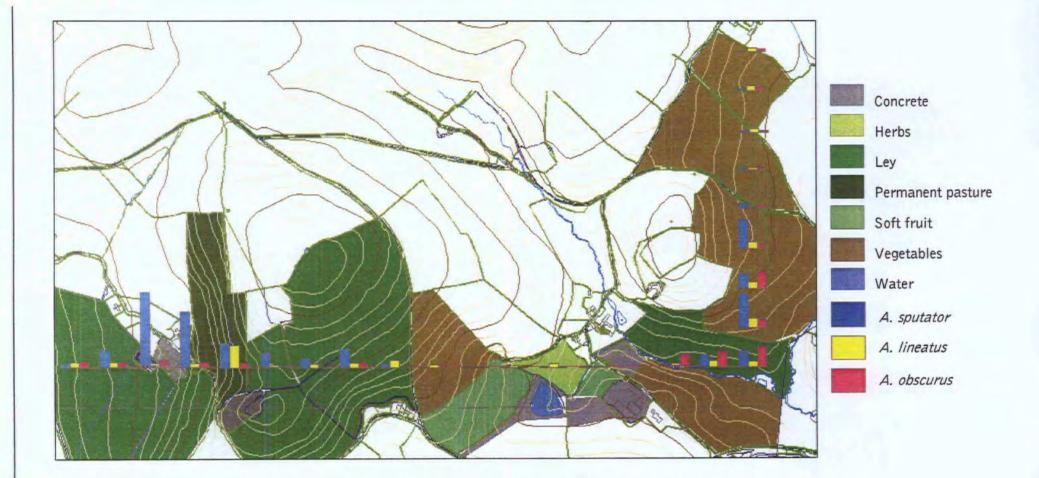


Figure 2-3 Cumulative catches of three species of click beetles: *A. sputator*, *A. lineatus* and *A. obscurus* in 2005. The maximum catch of *A. sputator* was 182 beetles per trap in one season, *A. obscurus* was 51 and *A. lineatus* was 55.

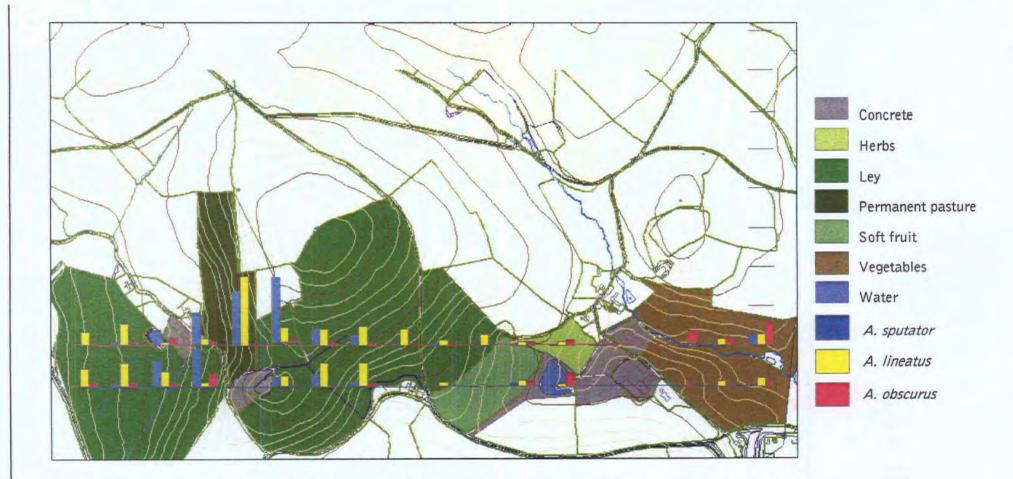


Figure 2-4 Cumulative catches of three species of click beetles: *A. sputator, A. lineatus* and *A. obscurus* in 2006. The maximum catch of *A. sputator* was 296 beetles per trap in one season, maximum catch for *A. obscurus* was 101 and for *A. lineatus*, 314.

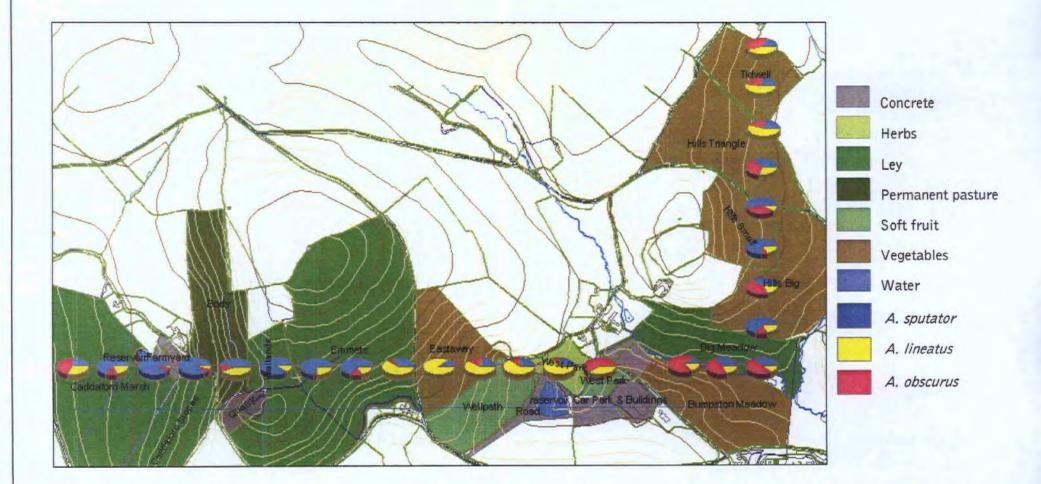


Figure 2-5 Pie charts showing relative proportions of catches of three species of click beetles (cumulative for 2005)

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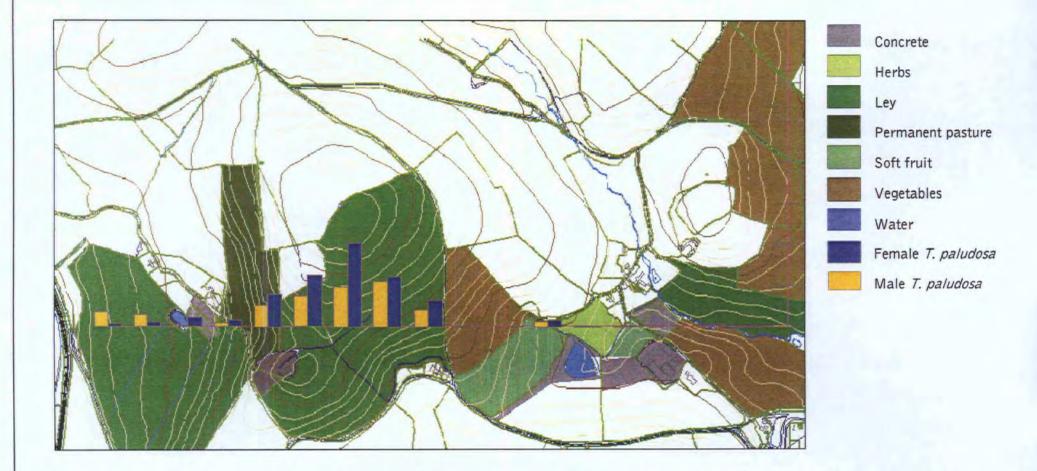


Figure 2-6 Cumulative catches of male and female crane flies along a transect in 2005. The maximum catch per trap in one season of male crane flies was 125 individuals and the maximum catch for females was 230.

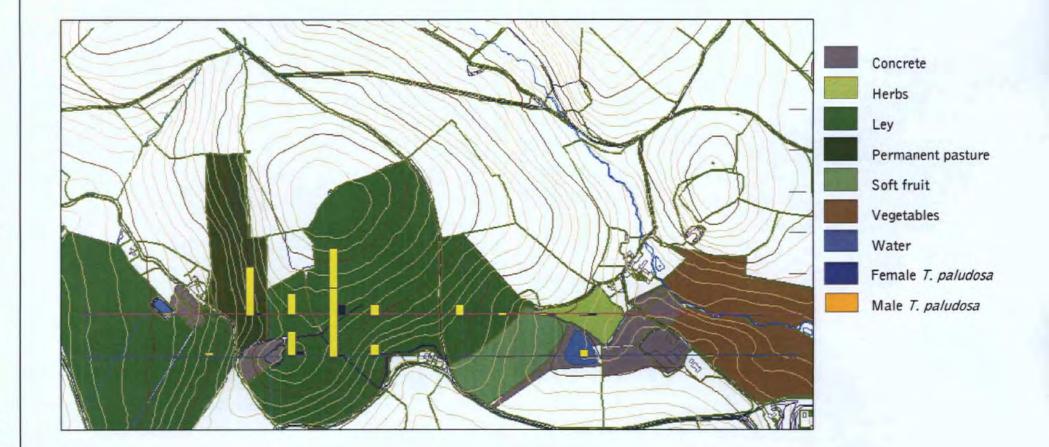


Figure 2-7 Cumulative catches of male and female crane flies along a transect in 2006. Maximum catch per trap in one season of male crane flies was 162 individuals and the maximum catch for females was 26.

2.3.2 Annual variation

There was no significant difference between the populations of the three click beetle species in 2005 and 2006 (Table 2-3) or either sex of crane fly for the eighteen sampling points along transect A.

	2005		2006			
	Mean	SD	Mean	SD	т	Ρ
A. lineatus	14.25	13.00	15.75	32.07	-0.17	0.871
A. obscurus	13.94	9.58	13.56	20.12	0.07	0.945
A. sputator	34.00	32.36	30.81	55.98	0.36	0.726
Male T. paludosa	21.68	43.61	20.16	46.59	0.14	0.892
Female T. paludosa	2.39	5.21	15.71	56.35	-1.32	0.197

Table 2-3 Comparison of mean catches of click beetles and crane flies in 2005 and 2006, across one 1.8km transect (transect A)

2.3.3 Land use

The distributions of catches for the four different crop groups were similar for *A*. *lineatus* and *A. sputator* (Figure 2-8) and were statistically significant for these two species (*A. lineatus* F=4.05, P=0.012 and *A. sputator*, F=5.00, P=0.004), but not for *A. obscurus* (F=0.60, P=0.617) where crop shows very little effect on distribution (Figure 2-8). The null hypothesis as formulated: crop has no effect on click beetle numbers must be accepted.

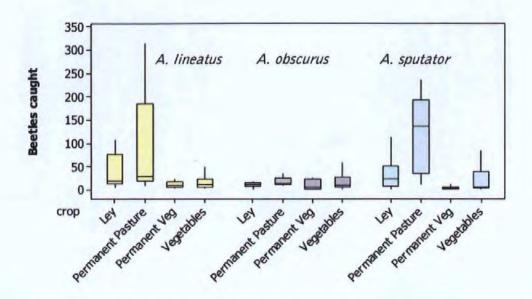


Figure 2-8 Boxplots of cumulative catches of three species of *Agriotes* beetles catches in different crop types in 2005 and 2006 in a heterogeneous organic vegetable production landscape in the UK. The box indicates the interquartile range, the middle horizontal line being the median and the upper and lower whiskers (vertical lines) showing the maximum and minimum data points within 1.5 heights of the box.

Catches of male crane flies were significantly related to crop type (F=3.03,

P=0.03). There is no significant crop effect on female crane flies (F=1.13,

P=0.348). The null hypothesis as formulated: crop has no effect on female crane fly

numbers must be accepted. But high populations of both male and female populations

were found on permanent pasture and ley (Figure 2-9), suggesting that populations

may be more dependent on crop type, than cultivations.

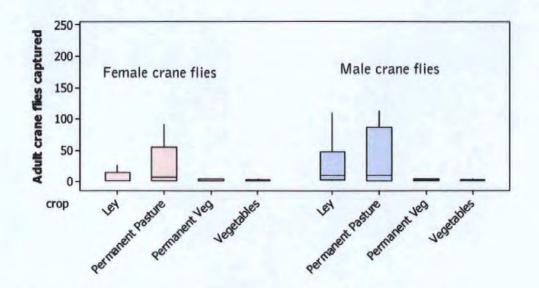


Figure 2-9 Boxplots of cumulative catches of male and female crane flies catches in different crop types in 2005 and 2006 in a heterogeneous organic vegetable production landscape in the UK. The box indicates the interquartile range, the middle horizontal line being the median and the upper and lower whiskers (vertical lines) showing the maximum and minimum data points within 1.5 heights of the box.

2.3.4 Distance from watercourse

Catches of both male and female crane flies were positively correlated with distance from the nearest watercourse i.e. as the distance from the watercourse increased (Table 2-4). *A. lineatus* and *A. sputator* click beetles also showed positive correlations with distance from watercourses, though it was significant for only *A. lineatus. A. obscurus* had a highly significant negative correlation with distance from a watercourse i.e. the number of *A. obscurus* increased as distance to a watercourse decreased. Scatter plots are shown in Figure 2-10 and Figure 2-11.

Table 2-4 Correlations of populations	of crane flies and click beetles with the
distance of traps from watercourses.	All data are log transformed. Statistically
significant results are shown in bold.	

	Correlation	r	Р
Female T. paludosa	Positive	0.51	0.003
Male T. paludosa	Positive	0.63	< 0.001
A. obscurus	Negative	-0.63	<0.001
A. lineatus	Positive	0.47	0.015
A. sputator	Positive	0.30	0.184

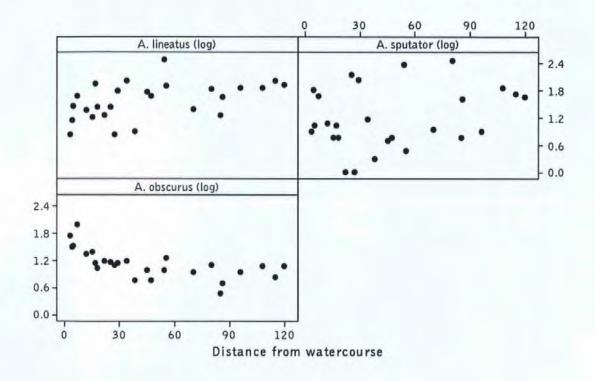


Figure 2-10 Total number of *A. obscurus*, *A. lineatus* and *A. sputator* click beetles (all log) captured in pheromone traps with distance from watercourse (metres)

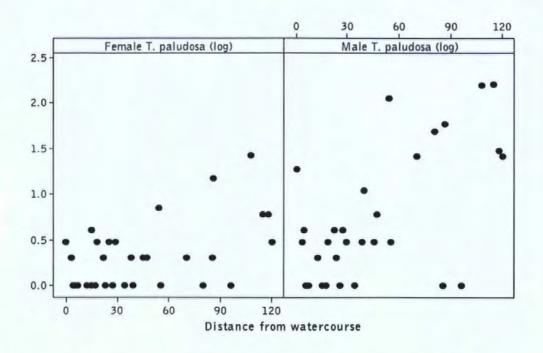


Figure 2-11 Total number of male and female crane flies (log) captured in water traps with distance from watercourse (metres)

2.4 Discussion

2.4.1 Click beetles

2.4.1.1 Spatial Distribution

The results show that there can be a considerable difference in the number of click beetles caught only 100m apart (Figure 2-3, and Figure 2-4). The strong differences observed between the three click beetle species are also illuminating. There were many *A. sputator* captured in this study, though *A. sputator* is less populous elsewhere in the UK (Anon. 1948; Furlan *et.al.* 2001a) except in arable areas in the East of the UK (B. Parker, pers. comm.)

Although all three click beetle species showed strong evidence of spatial clustering, the analysis of ratios shows *A. lineatus* had a strong similarity of population size within fields and relatively greater variation in population size over field boundaries, whereas *A. sputator* and *A. obscurus* populations appear to be less affected by field boundaries. *Agriotes* wireworms have often been treated as one species due to the difficulty of differentiating the species; these results (with adults) show that wherever possible the species should be treated separately. As the field scale seems appropriate for *A. lineatus* from these results, and this is the scale used for previous work (McCracken *et al.* 1995; Parker and Seeney 1997), this will be the scale at which future investigations in this study will be effected. There is also evidence that mosaics of landscape habitat allow different microhabitats within fields (Delettre 2005; Strickland *et al.* 1962). However, it would be valuable to investigate the optimum study scale for *A. sputator* and *A. obscurus*. In spite of *A. sputator* and *A.*

lineatus being less spatially defined by a field this is a valid scale at which to study these organisms as it is a familiar scale for practical implementation of control methods by farmers. Chapter 3 will investigate spatial structure of all species at a sub-field scale; this may enable identification of factors affecting distribution at a smaller scale.

The dispersal behaviour of each species may affect the results observed in this study. If *A. lineatus* moves faster, it may be more likely to move to the best environment. If the environment within a field is homogenous then the results we have seen would support *A. lineatus* being more mobile and either clustering towards pheromone traps within fields or crossing field boundaries to the best environment. Conversely, faster movers may have a smoother distribution as they move towards a pheromone trap from a wide area around the trap, covering a range of habitats. To assess these theories it is necessary to study the speed and distances moved by different species of click beetle (See Chapter 6).

Some evidence of uniformity of within-field populations may be obscured in this study due to the effect of hedges. Ester (2004a) reported that *A. sputator* were primarily found in verges whereas *A. obscurus* and *A. lineatus* were found in both verges and the centre of fields. Most wireworm damage to potato occurs in small fields (Parker and Howard 2001). Small fields have more headland (the uncultivated area of a field adjacent to a fence or hedge where machinery turns), and this headland may act as a reservoir for these pest species. If cultivations reduce populations as postulated, hedges and verges may act as reservoirs for soil pest larvae (Blackshaw 1988). This

study is not at a sufficiently small scale to observe any within-field effects; however, studies with a higher resolution, within one field, will be explored later (Chapter 3).

2.4.1.2 Annual variation

The mean catches of all species in 2006 were very similar to the mean catches in 2005, this is similar to that found in Blackshaw and Vernon (2006b). The click beetle species spend a number of years in the larval phase underground and so a relative consistency of densities from year to year might be expected. In order for a wireworm to develop to adulthood, a suitable habitat needs to be stable for a number of years. The population emerging each year will have shared its subterranean habitat with next year's emerging click beetles for a number of years. If any event had occurred to cause mortality to the emerging population, it is likely the following year's emerging population would also have undergone the same mortality-inducing event. Given this long lifecycle, any patterns in temporal distribution of click beetles may not be visible without many years of population recording (although *A. sputator* wireworms have been observed to experience epizootics (Keller 1994)). The slight differences in population densities that can be seen between species may be a function of their differing dispersal (Blackshaw and Vernon 2006b), where the faster a species travels the less apparent population change from year to year would be expected.

2.4.1.3 Land use

It would be expected that the occurrence and frequency of cultivation would have a greater effect on the quality of larval habitat of that location than the crop type. However, Figure 2-8 suggests that the biggest differences in populations of A_i *lineatus* and *A. sputator* are between grass and vegetable crop types, with a less marked difference between the permanent and rotational grass, or between permanent and non-permanent vegetable crops. Looking at the differences between permanent and rotational crops in Figure 2-8 and Figure 2-9 indicates that the crop type grown is shown to have more influence on populations than the occurrence of cultivations. *A. obscurus* appears to be affected by neither cropping nor cultivations to any large degree.

Byers (2000) found that communities of subterranean macroinvertebrate were more affected by biological factors than climactic factors, e.g. crop type and distance from water as opposed to temperature or altitude. One of the factors affecting this may be selective egg-laying. Gough and Evans (1942) found that *A. obscurus* selectively lay on grass in preference to wheat, clover, kale or bare earth. This suggests higher catches of *A. obscurus* in grass fields but this has not been shown in this study. It may not be possible to directly compare those data with those presented here, since the methods used were different. Pheromone traps attract males and the research of Gough and Evans (1942) relates to females. In addition, pheromone traps probably attract males for mating and egg-laying will occur after mating, so there may be a time delay.

2.4.1.4 Distance from watercourse

This transect study has also shown a difference between the three *Agriotes* species relating to another potential factor affecting distribution: distance from watercourse. Catches of *A. sputator* and *A. lineatus* populations were independent of distance from water courses. *A. obscurus* showed significantly higher populations closer to watercourses. There is strong evidence of preference for particular soil moistures by wireworm (Campbell 1937; Lees 1943), and it may be that this analysis is highlighting a distribution based on soil moisture rather than distance from a watercourse *per se*. It may be that distance from a watercourse determines the prevalence of damp habitats (e.g. marshes, ditches and reservoirs), and thus, that *A*. *obscurus* is more successful in damper habitats than either *A. sputator* or *A. lineatus*. Whatever the explanation, this analysis shows that the three species are differentially associated with distance from a watercourse, and this may be found to be true for their larvae. However, this differentiation of species responses is not present in most existing literature.

2.4.2 Crane flies

2.4.2.1 Spatial Distribution

The distribution of crane flies shows less variation than that of any of the click beetle species studied (Figure 2-3, Figure 2-4, Figure 2-6 and Figure 2-7). This is potentially a result of *T. paludosa* crane fly's ability to fly, which increases their dispersal ability compared to the beetles walking. As the observed crane fly populations (male or female) did not show a strong within-field grouping but showed a relatively smooth distribution over the 2km transect (Figure 2-6), their effective population distribution may be determined at a scale greater than that of individual fields. Alternatively, tall Devon hedges (a substantial earth bank, well covered in vegetation, which may be up to 8 feet high) may impede the migration of gravid female crane flies (Frouz and Paoletti 2000; Pritchard 1983), so captured populations of recently emerged females are likely to have originated within that field. If this were true, a greater variation in numbers would be expected between

fields than within the same field. Ifoulis and Savopoulou-Soultani (2006) noted that *Lobesia botrana* 'Denis & Schiffermuller' (Lepidoptera: Tortricidae), which also has a female with limited mobility prior to oviposition, shows a degree of within field aggregation (particularly, an edge effect). It is necessary to sample at a within-field scale to see evidence of such spatial patterning (see Chapter 3).

2.4.2.2 Annual variation

Many of the fields in this study were in a vegetable-grass rotation and underwent cultivation between the two years of sampling; it is therefore surprising that there was not more variation in populations between the two years. Although the numbers appear to vary considerably in the two years sampled, the differences were not significant. In contrast to the situation with click beetles, any mortality-inducing event would affect one cohort of subterranean larvae only, so we may expect to see a large variation in year-to-year populations of leatherjackets, as seen by Mayor and Davies (1976). This may be because adult females emerge fully gravid and this impedes their dispersal until the majority of their eggs have been oviposited. (Pritchard 1983) in a similar location to the point of emergence, thus maintaining year on year population stability. If females remain close to the point of emergence it is to be expected that the males will remain in the vicinity of the females. When the majority of eggs have been oviposited, females are able to travel greater distances to lay their remaining eggs, this may enable colonisation of new areas by the few remaining eggs.

2.4.2.3 Land use

Crane flies show a clear preference for grass, whether it is permanent or a rotational ley (Figure 2-9). This is probably the habitat in which they are most likely to

complete their larval stage without considerable mortality and the roots act as a food source for the larvae of the next generation. The significant result for male crane flies indicates potential for further investigation. Particularly, it may be possible that the populations are defined by the cropping history over a number of years, especially with regard to range of data for the male crane flies.

2.4.2.4 Distance from watercourse

The catches of both male and female crane flies were positively correlated with distance from the nearest watercourse. As with the wireworms in section 2.4.1.4, leatherjackets are heavily influenced by soil moisture, with extremes causing high mortality (Blackshaw 1983a; Meats 1970; Pritchard 1983). The trap catches indicate that moister areas closest to watercourses may provide the most suitable habitats for the crane flies, suggesting that drought, and not flooding, is the limiting factor on leatherjacket populations. A more thorough testing of this concept would have been achieved by measuring the soil moisture at each point along the transect, however, this was not achieved in this project.

This chapter has shown that all click beetle species (and male and female crane flies) show evidence of spatial clustering at a landscape scale. However the different species appear to be reacting to different factors at this scale, *A. lineatus* and *A. sputator* and male *T. paludosa* appear to be strongly affected by crop type, whilst male and female *T. paludosa*, *A. lineatus* and *A. obscurus* are affected by the distance from the nearest watercourse, which may reflect soil moisture status. None of the studied species showed a significant difference in population levels from one year to the next.

This chapter has raised many other questions. Whilst variations in populations may be viewed at this, landscape scale, they may or may not be present at other scales. Therefore subsequent chapters will examine spatial patterns of click beetle and crane fly species at larger (regional) and smaller (field) scales to examine their spatial patterns and associated environmental causes. This is necessary as studying ecological factors is very scale-dependent (Legendre *et al.* 2002). This chapter and Chapter 3 indicate that there are other factors affecting distribution for these organisms; these will be examined in Chapter 4. This chapter has also a new question regarding the possible effect of differences in dispersal ability and speed of travel in explaining differences in spatial distribution by dispersal; this will be examined in Chapter 6.

The distribution of *T. oleracea* in this landscape was found to be zero, whereas *T. paludosa* and click beetles were found to be present throughout and to show spatial clustering. The population levels were found to be similar in locations sampled in two consecutive years, however population levels were found to vary depending on distance from the nearest watercourse and the crop type cultivates in the field. These geographic factors studies influenced the population levels differently depending on the species.

Chapter 3 Spatial structure of larval pest populations

3.1 Introduction

Knowledge of the spatial structure of subterranean populations has consequences for both population prediction and the development of sampling schemes. This chapter examines the spatial distribution of soil-borne pests at regional and field scales to complement work focusing on the landscape scale (Chapter 2). Moran's I and Taylor's Power Law will be used to analyse field samples taken at a regional level, whilst SADIE will be used to measure spatial pattern within a single field. The effect of edaphic factors on population distribution will be examined at single field scale using SADIE and correlations.

Salt and Hollick (1946) found aggregation of wireworm at three different scales; field, plot and micro scale (9x9 3-inch (7.6cm) square sampling areas). The transect work in chapter two showed that there was significant spatial structure in the populations of these organisms being studied at a landscape scale, but, systematic sampling within fields will enable identification and characterisation of structure at a more practical (pest management) scale.

There are a number of ways of assessing spatial structure in organisms. Traditionally, spatial analysis methods focused primarily on numerical characteristics of counts, for example, the log-mean log-variance relationship of Taylor (1961) or Moran's I (Moran 1950). Seal (1992b) analysed wireworm populations with Iwao's Patchiness Regression and Taylor's Power law and much better fit was obtained with the latter. Taylor's Power Law (Taylor 1961) is the most frequently applied and more consistently adequate technique for assessing spatial structure (Taylor *et al.* 1978). Both Taylor's Power Law (analysing the mean and variance relationship) and Moran's I (a measure of spatial autocorrelation) can identify whether the distribution of an organism is random or non-random, however, SADIE (Spatial Analysis by Distance IndicEs) not only provides a measure of aggregation (I_a) but the red-blue plots that can be produced enable the visualisation of spatial structure, in the form of patches and gaps at a scale below the gross sampling area (Perry 1996). SADIE also allows the spatial correlation of spatially variable edaphic factors with populations, which Moran's I and Taylor's Power Law cannot.

Knowledge of the existence and pattern of spatial structure can help in the development of sampling schemes (Arnaldo and Torres 2005; Binns and Nyrop 1992; Lessio and Alma 2006), which can reduce the labour requirement of sampling. The current standard sampling scheme for leatherjackets and wireworms is 20 cores sampled randomly throughout the field (Parker and Howard 2001). It is very labour intensive and even when only 20 cores are sampled there are still very wide margins of error (Blackshaw 1987a; Jones 1937; Salt and Hollick 1944; Yates and Finney 1942), there is a need for a more accurate sampling scheme which requires less effort. A thorough knowledge of spatial structure would allow the development of such a scheme and would enable assessments of the accuracy of population estimates.

Knowledge of spatial pattern has implications for control. If patches of high populations densities can be identified, control can be limited to the areas of the field most at risk. In organic crops this may be avoidance (see section 1.4.2.2), in

conventional crops this knowledge may allow differential application of pesticides, leading to a reduction in pesticide usage.

The previous chapter showed that both cultivation and moisture influence the distribution of soil pests, confirming previous work (Blackshaw 1988; Campbell 1937; Coulson 1962; Hofman and Mason 2006; Lees 1943; McCracken *et al.* 1995; Parker and Howard 2001; Seal *et al.* 1992a). The work on these relationships has often focused on the field scale effect on populations, the finer structure of distribution has not been observed.

The hypothesis that click beetles find suitable habitats in headlands (Ester *et al.* 2004b; Parker and Howard 2001) may be evaluated by observing the distribution of wireworms at this sub-field scale. It may also be possible to observe any aggregations caused by the limited mobility of gravid female crane flies at this scale.

The aims of this study were:

- a. To determine if spatial structure is discernable and quantifiable.
- b. To relate any distribution patterns found to environmental factors.

3.2 Methods

3.2.1 Systematic sampling

Systematic sampling (samples taken in a uniform grid pattern from defined locations within the sampling area) was carried out in ten organic fields from two farms in Devon, UK, that had previously been identified as having populations of leatherjackets, wireworms or both. The fields were sampled in January and February 2005 and then resampled in January and February 2006 with the 2006 samples located as close to the 2005 sample points as possible. A non-differential hand-held GPS (Garmin Gecko) was used to relocate sampling points; its accuracy was confirmed with tape measurements to landmarks.

The extent of the sampling area and the lag (distance between samples) varied, with a target of *ca.* 70 cores to be collected across the entire field or a substantial part of it (Table 3-1). The sample grain (size) (Dungan *et al.* 2002) was constant; one soil core 10cm diameter, 10cm deep (Jedlicka and Frouz 2007) was processed using wet-sieving (Simmons *et al.* 1998) within 48 hours of sampling.

Soil sampling is the traditional method of wireworm (Salt and Hollick 1944) and leatherjacket sampling (Blackshaw 1983a; McCracken *et al.* 1995). This method has many drawbacks, including; being very labour intensive, requiring laboratory processing and having a detection threshold of approximately 62,000 per hectare when 20 x 10cm cores are taken, which is above the probable damage threshold for potatoes (Parker and Howard 2001). The accuracy of the larval counts may be improved by taking larger numbers of soil cores; however this requires a concomitant increase in effort. Yates and Finney (1942) determined that a greater number of smaller soil cores was more efficient than the same amount of soil collected in larger samples (4" diameter cores as opposed to 6" squares).

Soil conductivity, as a surrogate for soil moisture, was measured using a Thetaprobe (Delta-T devices) adjacent to each core. Soil shear strength was measured in 2006 with a hand held shear vane with a 19mm blade (Edeco Pilcon).

Parker (1994) developed a method for the detection of wireworms using traps bailed with a seed mix and moisture gel. Bait traps used for the systematic sampling of one field, consisting of a 1:1 wheat-barley seed mixture inside a 300ml plastic pot, predrilled with 25 x 2mm holes. These were placed in voids formed by soil core removal and the space around the trap was then backfilled with loose soil from another part of the field (Parker 1996; Seal *et al.* 1992b). These traps were left undisturbed for two weeks. The traps were then collected and hand sorted for specimens. The backfilled soil surrounding the traps was also collected and hand sorted. Bait traps in pots have been found to work more effectively than plate bait traps or mesh bags containing baits (Brunner *et al.* 2006).

Farm	OS Grid Ref	Field	Crop (2005)	Crop (2006)	Lag (m)	No. of Cores
Home Park	SX740606	C	Permanent Pasture	Permanent Pasture	12×12	74
Home Park	SX739609	G	Ley	Brassica	20x20	71
Home Park	SX738606	А (Тор)	Permanent Pasture	Permanent Pasture	12x15	58
Home Park	SX737608	F	Ley	Ley	14x18	72
Home Park	SX739605	A (Bottom)	Permänent Pasture	Permanent Pasture	6x6	72
Riverford	SX781645	Broad Park	Ley	Brassica	30x30	68
Riverford	SX782648	Bakers Park	Ley	Broad Beans	25x25	61
Riverford	SX771657	Hills Small	Lettuce	Lettuce	25x15	76
Riverford	SX772655	Hills Big	Leeks	Not sampled	20x20	74
Home Park	SX740606	C (Traps)	Permanent Pasture	Permanent Pasture	3x3	90

Table 3-1 Location and crops of fields and numbers and distributions of systematic sampling points for leatherjacket and wireworm counts in 2005 and 2006.

3.2.2 Mean variance relationships

The data from the systematic sampling was used in addition to the data from the survey in Chapter 4. Fields with zero counts were excluded, after discussion with the project steering committee. Taylor's Power Law (Log $s^2 = a + b \log \bar{x}$) was applied to all of the data to ascertain whether populations were random, uniform or clustered (see page 86).

3.2.3 SADIE Analyses

SADIE uses spatially referenced data to provide an 'index of aggregation' (I_a) (Perry 1996). The I_a is a single value of the spatial aggregation of two dimensional count data in an area; it is calculated with an associated P value. The I_a is calculated by dividing D (the minimum distance moved by counts to achieve a completely regular arrangement of counts within the sampling area) by E_a (the distance moved by counts to achieve a completely regular to achieve a completely random arrangement). Thus, where $I_a=1$ the counts are in a random distribution and where $I_a > 1$ there is aggregation of the count data (Perry 1996). The counts in this instance are the number of larvae found, and the numeric measures of soil shear and moisture.

As the counts are 'shared' between sampling points within the sampling area to achieve a random distribution, it follows that some locations will be net donors of counts, and some will be recipients of counts. Where neighbourhoods of donors (i) predominate the area is termed a 'patch', and a neighbourhood where recipient (j) locations dominate is termed a 'gap'. These areas are identified as having an Index of Clustering, v, (v, or v_j) of greater than 1.5 or less than -1.5 (these values are arbitrary but conventionally used). These v_i or v_j values can then, after being interpolated with a kriging function (projection of the values between known points), be displayed as a contour map using Surfer 6.04 (Golden Software Inc., 1997, Golden, CO USA) to easily identify the geographic locations of patches and gaps (Perry 1996). These are customarily displayed as red patches and blue gaps with white areas depicting random zones (Perry *et al.* 1999).

3.2.4 Correlations between larval distribution and edaphic factors

The relationship between larval distributions and edaphic factors would normally be statistically determined using a regression. However, as the data is non-normal (Kolmogorov-Smirnoff) and cannot be transformed, and there is no practical non parametric regression method, the results were analysed by correlation (which assumes neither variable as cause or effect). A two-tailed Spearman Rank-Order correlation was used in SPSS (SPSS Inc. version 11.5.1 for Windows) to assess relationships between leatherjackets numbers and soil conductance (as a substitute for soil moisture) and leatherjackets and soil shear strength at 5cm and 10cm depth. Relationships between wireworms and the same edaphic factors were tested for correlation in the same way.

In addition, when there are two sets of counts within the same geographic sampling area SADIE techniques can be used to look for associations and/or dissociations between them (Perry 1997). The two sets of counts may be abundances of two different species, or the same species on two different occasions. Alternatively, the two sets of counts may be one count of species and one of an environmental variable, or two environmental variables. There will be an association in a location where a patch appears in both sets of counts and it will also be an association where a gap appears in both counts. Dissociation will occur when the location of a patch coincides with the location of a gap. These associations and dissociations can be plotted on contour maps for visualisation, and are customarily represented as purple (dissociation) and green (association). Associations and dissociations can be used to provide evidence of relationships between species distribution and environmental factors, or intra- or inter species interactions.

3.3 Results

3.3.1 Mean Variance Relationships

The R² for the relationship between mean and variance for both wireworms and leatherjackets are very high (0.985 and 0.964). This shows that the regression lines are representing over 95% of all the variation present in the dataset. For both of these graphs the coefficient of x is extremely close to one (0.989 and 0.995). This means that the fields sampled here have, essentially, randomly distributed populations of both leatherjackets and wireworms and show no signs of aggregation.

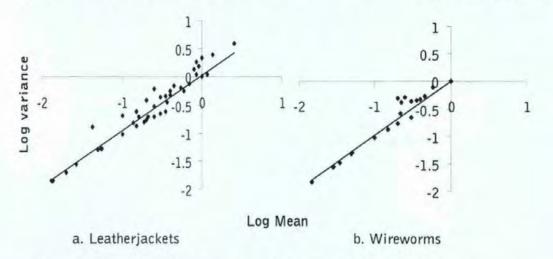


Figure 3-1 log mean - log variance plot for counts of leatherjackets. (a: y=0.995x + 0.032, $R^2=0.964$) and wireworms (b: y=0.989x + 0.008, $R^2=0.985$) in 10cm soil cores

3.3.2 SADIE Red-blue plots

Unlike Taylor's Power Law, SADIE analyses function on a field by field basis and have the capability to look for spatial patterning in one location. In addition, SADIE has the advantage of enabling visualisation of any spatial pattern that is revealed. Each field was analysed each year (Table 3-2, Table 3-3, Table 3-4 and Table 3-5). Significant plots are mapped below (Figure 3-2 and Figure 3-3); those with P values

above the 0.05 significance level are not illustrated here, but in appendix 2.

Table 3-2 SADIE analyses of leatherjacket counts in 10cm soil cores for systematically sampled fields in 2005. I_a is the Index of aggregation and is provided for the overall area, the neighbourhoods of high populations (patches) and the neighbourhoods of low populations (gaps). Significant analyses (P<0.05) are shaded. Listed in order of population size.

Field	No:of Cores	Population (000's ha ⁻¹)	Ia	PI.	I _a Gaps	P I <u>.</u> Gaps	I <u>,</u> Patches	P I <u>.</u> patches
Bakers Park	61	1,004	1.57	0.024	-1.57	0.021	1.51	0.025
Broad Park	68	680	1.62	0.015	-1.59	0.014	1.59	0.015
HPG	71	458	1.37	0.029	-1.37	0.027	1.38	0.026
HPF	72	260	1.58	0.007	-1.59	0.005	1.63	0.004
HPA (Top)	58	237	1.01	0.389	-1.02	0.365	1.02	0.355
HPA (Bottom)	72	174	1.28	0.059	-1.27	0.073	1.31	0.054
HPC	74	68	0.72	0,974	-0.71	0.974	0.80	0.851
Hills Big	74	34	1.34	0.055	-1.34	0.055	1.24	0.074
Hills Small	76	16	1.28	0.957	-1.30	0.085	1.00	0.132

Table 3-3 SADIE analyses of leatherjacket counts in 10cm soil cores for systematically sampled fields in 2006. I_a is the Index of aggregation and is provided for the overall area, the neighbourhoods of high populations (patches) and the neighbourhoods of low populations (gaps). Significant analyses (P<0.05) are shaded. Listed in order of population size.

Field	No of Cores	Population (000's ha ⁻¹)	I _a	P I <u>a</u>	Í _a Gaps	P I <u>a</u> Gaps	I <u>a</u> patches	P I _a patches
HPA								
(Тор)	58	237	1.03	0.348	-1.05	0.323	1.00	0.3933
HPC	74	203	0.89	0.645	-0.87	0.697	0.95	0.5137
HPA								
(Bottom)	72	174	0.98	0.472	-0.98	0.453	1.01	0.3890
HPF	72	69	1.20	0.138	-1.21	0.121	1.23	0.1016
HPG	71	53	1.02	0.426	-1.01	0.426	1.00	0.0652
Bakers								
Park	61	25	0.80	0.800	-0.81	0.787	0.80	0.8173
Hills								
Small	· 76	17	1.70	0.003	-1.73	'0 . 003	1.41	0:0094

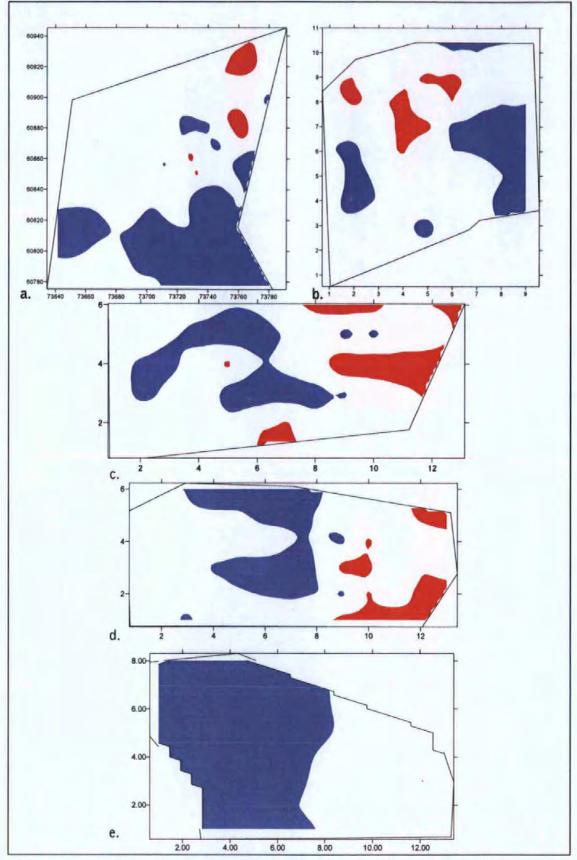


Figure 3-2 Leatherjacket distribution from SADIE analysis from 2005 and 2006. (a. Home ParkF b. Home ParkG, c. Bakers Park, d. Broad Park, e. Hills small). Blue indicates gaps and red indicates patches.

As with Taylor's log-mean log-variance plots there is no evidence of spatial patterning for leatherjackets when populations are low (Hills Small 2006, being an exception). However, the highest populations all show statistically significant spatial patterning. For leatherjackets the threshold for revealing spatial patterning appears to be about 250,000 ha⁻¹ (Table 3-2).

The SADIE plots (Figure 3-2and Figure 3-3) all show both patches (shown as red),

and gaps (blue), (neighbourhoods of significantly high populations and

neighbourhoods of significantly low populations respectively). There is no evidence of

a field edge effect. Generally, the gaps are larger than the patches, but the patches

do appear to be grouped slightly.

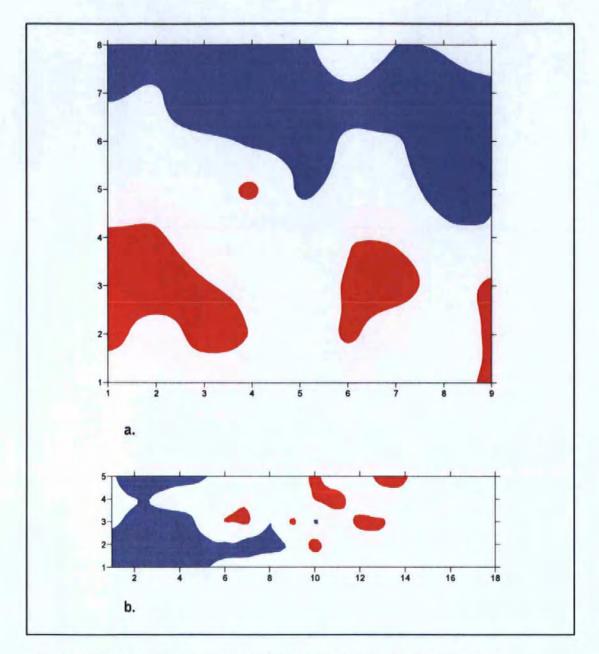
Table 3-4 SADIE analyses of wireworm counts in 10cm soil cores for systematically sampled fields in 2005. I_a is the Index of aggregation and is provided for the overall area, the neighbourhoods of high populations (patches) and the neighbourhoods of low populations (gaps). Significant analyses (P<0.05) are shaded. Listed in order of population size.

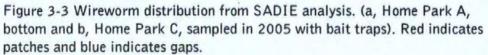
Field	No of Cores	Population (000's ha ⁻¹)	P∘I _a	I _a	I <u>,</u> Gaps	P I _a Gaps	I <u>a</u> patches	P I _a patches
HPA								• • •
(Bottom)	72	729	0.003	1.61	-1.62	0.002	1.67	0.001
HPC								
(traps)	90	556	0.034	1.58	-1.62	0.036	1.51	0.054
HPA								
(Тор)	58	453	0.789	0.84	-0.83	0.793	0.84	0.791
Bakers								
Park	61	615	0.938	0.74	-0.74	0.927	0.77	0.915
HPG	71	352	0.334	1.04	-1.07	0.305	0.87	0.819
HPF	72	347	0.221	1.12	-1.13	0.212	1.04	0.358
HPC (soil								
cores)	74	304	0.381	1.01	-0.99	0.408	0.99	0.421
Broad								
Park	68	184	0.074	1.32	-1.35	0.071	1.00	0.891

Table 3-5 SADIE analyses of wireworm counts in 10cm soil cores for systematically sampled fields in 2006. I_a is the Index of aggregation and is provided for the overall area, the neighbourhoods of high populations (patches) and the neighbourhoods of low populations (gaps). Significant analyses (P<0.05) are shaded. Listed in order of population size.

Field	No of Cores	Population (000's ha ⁻¹)	ΡI,	Ī	I <u>,</u> Gaps	P I. Gaps	I <u>.</u> patches	P I, patches
НРА								
(Bottom)	72	573	0.302	1.05	-1,05	0.304	1.07	0.253
HPA								
(Тор)	58	280	0.128	1.25	-1.22	0.147	1.33	0.876
Bakers								
Park	61	275	0.589	0.91	-0:90	0.615	0.95	0.494
HPC (soil								
cores)	74	270	0.458	0.96	-0.96	0.476	0.983	0.435
HPC				1				
(traps)	90	83	0.279	1.09	-1.11	0.252	1.06	0.210
Broad	(0	40	0 411	0.00	0.00	0 422	0.00	0.4/4
Park	68	42	0.411	0.99	-0.99	0.423	0.98	0,464
Hills	77	24	0 7 7 5	0.9/5	0.04	0 770	0.99	0.4/5
small	76	34	0.725	0.865	-0.84	0.772		0.465
HPF	72	17	0.836	0.82	-0.80	0.836	1.00	0.260

Only two fields, both sampled in 2005, showed significant spatial pattern for wireworms (Figure 3-3). These fields had high populations, although one other, HPA (bottom) sampled in 2006 had an equally high population but did not show significant spatial pattern. The threshold for wireworms could be between 300,000 and 550,000 ha⁻¹ (Table 3-4) if HPA (bottom) is an exception. Alternatively, it can be reasonably assumed that if the bait traps (used in HPC) are attractant, they will positively encourage significant spatial structure. This would place the threshold for detectable structure at a population between 573,000 ha⁻¹ and 729,000 ha⁻¹. Both of these thresholds are considerably higher than the threshold identified for leatherjackets.





The sampling areas showing regions of patches are separated from those showing regions of gaps in both of the significant plots for wireworm distribution, showing as top and bottom for a, and left and right for b (Figure 3-3).

3.3.3 Relating leatherjacket and wireworm distribution to environmental variables

Moisture was significantly correlated with leatherjacket incidence in two fields, one positively and one negatively (Appendix 1). However, fewer than 1 in 20 (or 5%) correlation coefficients are statistically significant. This is equivalent to the false error rate of the P=0.05 significance level used, so it may be unwise to value these significant results. This is also comparable to Salt and Hollick (1944), who estimated that only 5% of fields showed significant patchiness to make differential cropping plans necessary for protection against wireworms.

The correlations assess the relationship over the entire field. A relationship may be present, but, any spatial autocorrelation within the field will invalidate the assumption of independence of samples in these analyses (Legendre 1993). SADIE associations are designed for this type of analysis; it looks at association or dissociation of two variables in smaller patches with uneven distribution throughout the field. Analyses were carried out for each combination of variables for each field and the SADIE output states that the two variables for that field may be associated, dissociated or neither dissociated nor associated. Alternatively, some field were not analysed, often as no larvae were found, or on one occasion, as the field had been covered in black plastic in the second year, which prevented sampling. Table 3-6, Table 3-7, Table 3-8 and Table 3-9 show a count of the output of the analyses for each pair of variables, e.g. in 2005, wireworms were associated with

leatherjackets in one field, dissociated in two fields and neither in four fields.

Table 3-6 Summary of the relationships between wireworm counts in 2005 and other measured parameters as revealed by SADIE analysis. See Appendix 2 for detailed analysis results.

	Leatherjackets	Moisture	Wireworms
	2005	2005	2006
Associated	1	1	3
Dissociated	2	2	0
Neither	4	5	4
Total Analysed	; 7	8	7

Table 3-7 Summary of the relationships between wireworm counts in 2006 and other measured parameters as revealed by SADIE analysis. See Appendix 2 for detailed analysis results.

	Wireworms 2005	Leatherjackets 2006	Moisture 2006	5cm Shear 2006	10cm:Shear 2006
Associated	3	1	0	0	0
Dissociated	0	1	0	1	1
Neither	4	4	8	7	7
Total Analysed	7	6	8	8	8

Table 3-8 Summary of the relationships between leatherjacket counts in 2005 and other measured parameters as revealed by SADIE analysis. See Appendix 2 for detailed analysis results.

	Leatherjackets 2006	Moisture 2005	Wireworms 2005
Associated	3	1	1
Dissociated	1	1	2
Neither	3	7	4
Total Analysed	7	9	7

Table 3-9 Summary of the relationships between leatherjacket counts in 2006 and other measured parameters as revealed by SADIE analysis. See Appendix 2 for detailed analysis results.

	Leatherjackets 2005	Moisture 2006	5cm Shear 2006	Wireworms 2006	10cm Shear 2006
Associated	3	3	2	1	3
Dissociated	1	0	1	1	1
Neither	3	4	4	4	3
Total Analysed	7	7	7	6	7

Leatherjackets in 2006 were frequently associated with the distributions in 2005; this evidence of association was also evident in the wireworm analyses, indicating possible population stability from year to year. There is no consistency between the relationship between the distribution of wireworms and leatherjackets (Table 3-6) and soil moisture, soil shear and each other. Leatherjackets and wireworms are associated and dissociated with each other about an equal number of times (two associations and three dissociations); more often, there was evidence of neither.

3.4 Discussion

Although there is evidence of spatial patterning in this collection of field surveys, the majority of the fields sampled did not show statistically significant results. This contributed to the results of the analysis using Taylor's Power Law (Figure 3-1): random distribution of the larvae. A Taylor's plot requires many samples to be taken, on this occasion each point on the graph is a numerical characteristic of over 60 aggregated samples and there are over 25 points on each graph. The highest populations showed evidence of non-random spatial distribution, Park *et al.* (2006), also found spatial structure at high population densities of a pest (*Homalodisca coagulata*) which was not evident at lower populations. Also like this study, Komonen (2006), studying two congeneric saproxylic beetle species, found significant spatial structure using Moran's I that was not evident when analysed with SADIE.

Although there are fewer samples in each SADIE analysis, each plot is just one group of samples, spread out in their original geographic distribution and thus it is possible

to identify individual populations that show aggregation. Legendre (2002) highlighted the causes of spatial patterning as being biotic, environmental or deterministic (physical), Salt and Hollick (1946) identified oviposition, differential survival and migration as being the causes of spatial distribution in wireworms. Salt and Hollick found aggregations at a micro scale; with the small larvae more aggregated than the large larvae. They found large larvae are essentially randomly distributed; they attributed this to dispersion of larvae after oviposition in a cluster. They also found some clusters of older larvae deeper in the soil that they believed were formed around favourable feeding sites in a generally less favourable feeding environment. The different distribution of small and large wireworms was also observed by Ibbotson (1958) and Seal *et al.* (1992b). The larvae in this study were measured, but with a complex of three species, there is no evidence that size directly relates to age so these measurements have not been analysed and no conclusions can be drawn in this way.

Historical studies using Taylor's Power Law show that both leatherjackets and wireworms reveal different spatial structure at different population densities. There is evidence that spatial distribution increasingly differs from random as populations increase (Taylor *et al.* 1978). Blackshaw (1987a) found a coefficient of 1.14 in fields with moderate to low populations of leatherjackets (not statistically significant from 1), and Finney (1941) found randomly distributed populations when there were less than 250,000 wireworm per acre (617,500 ha⁻¹). In this study aggregation was found for both wireworms and leatherjackets only when populations were high. Unfortunately, the threshold population for identifying spatial structure may be higher than crop protection threshold populations (Parker and Howard 2001).

Damaging populations may be present at population densities that, in this study exhibited no discernable spatial structure.

The importance of spatial structure from a crop protection perspective is the ability to identify areas at risk and reduce the damage to those areas. But Anon (1948) attributed patchiness of crop damage, not to patchiness of the wireworm population but to differential fertility; reasoning that in patches of low fertility plants cannot so easily grow away from the damage caused by larval feeding and observing that "old grassland has particularly patchy fertility". Finney (1941) made a similar argument, that the patchy failure of crops was more likely to be due to the greater efficiency of the wireworm destroying the crop rather than the amount of damage. SADIE analysis offers a method to establish whether crop patchiness is due to differential fertility of fields or clusters of high wireworm population by the comparison of spatial structure of populations with the spatial structure of the relevant variables. Surveys or aerial photographs of crop damage (highly visible just after germination) could be directly related to SADIE red-blue plots of larval patchiness.

There was no evidence for the aggregation of wireworms around headlands as hypothesised by Parker and Howard (2001) and Ester (2004b). There was no discernable edge effect (see sections 2.4.1.1 and 2.4.2.1) for any of the fields that showed significant spatial structure.

The results obtained for the associations and dissociations of the larvae with moisture and shear are inconclusive, with both associations and dissociations present in the analyses of each variable and many analyses showing neither. There are significant

relationships in many of the instances, so soil moisture and soil shear strength are, on occasion, influencing the larval distribution, but the relationship is not simple and it is possible that it is not linear. This is in agreement with Jedlicka and Frouz (2007) who did not find a significant correlation between soil moisture and A. obscurus wireworm populations. Salt and Hollick (1946) also attempted to relate wireworm spatial distribution to soil consolidation and soil moisture but did not find a correlation with either. The most likely explanation is that both moisture and shear strength encompass a range of values with a smaller range in the spectrum of values providing ideal conditions for larvae, with conditions either side being increasingly less suitable, which concurs with field and laboratory observations on leatherjacket soil moisture preferences reported by Kell (1988) and with the findings of Lees (1943) and Lefko (1998). Lees found that wireworms avoided dry soil, yet Lefko found that many days of high soil moisture was unfavourable for wireworms. The impact of moisture on populations applies in the sub-field scale - i.e. influencing the number of larvae in each soil core, but also on a whole field scale, i.e. is the whole field too wet to sustain a population?

Spatial structure of leatherjacket and wireworm populations was found to be present. The edaphic factors, soil shear and moisture are very often significantly related to the populations of soil pests, but cannot currently be used alone to predict neighbourhoods of high and low populations by themselves. However, it is possible that their incorporation with a greater number of variables at the field level may enable the identification of factors affecting distribution and of fields likely to contain damaging populations. In addition, this may enable the identification of variables that can be manipulated to reduce populations. One of the problems identifying environmental determinants was the low population density of these pests in the study area. This area of investigation will benefit from further studies that broaden range of sites sampled and the environmental variables measured (Chapter 4).

Chapter 4 Survey of environmental factors affecting Agriotes and Tipulid distribution

4.1 Introduction

For pest management purposes, it would be extremely useful to be able to predict the presence of soil pests without the need for intensive sampling of subterranean individuals (Chellemi 2000; Ross *et al.* 1948; Salt and Hollick 1944). The adults of these pests are much simpler to study but, unfortunately, there is rarely a consistent relationship between click beetles and wireworms (Krivokhizhin 1991; Samson and Calder 2003). Work to relate the catches of wireworm populations to click beetle populations has been attempted using pheromone traps. Furlan (2001a) linked high catches of *A. ustalatus, A. brevis* and *A. sordidus* with damaging populations of larvae, but did not find a correlation with *A. lineatus* click beetles and wireworms and did not study *A. obscurus* and *A. sputator*. Blackshaw and Vernon (2006a) found a spatial relationship between *A. obscurus*.click beetles and wireworms but failed to find one for *A. lineatus* in Canada. 'However, *A. sputator* was not present at their study site. 'Knowledge of the relationship between wireworms and click beetles was highlighted as a priority by Parker and Howard (2001) in their review on damage to potatoes by wireworms.

The situation with leatherjackets is more straightforward. Blackshaw (1987b) found strong relationships between leatherjackets and female crane flies.

Chapter 2 highlighted some of the factors affecting the distribution of leatherjackets and wireworms in an organic vegetable and grassland rotational environment, namely distance from the nearest watercourse and crop type. However, as discussed in sections 1.2.2 and 1.3.2, there are a large number of other environmental variables also reported to affect either one or both pest species. M^cCracken (1995) and Parker and Seeney (1997) carried out surveys with large numbers of environmental variables recorded over large areas, but had different variables within their experimental design and thus, results from these studies cannot be directly compared to each other. There are three broad categories of environmental variables likely to affect soilinhabiting insect larvae: chemical (e.g. soil pH), physical (e.g. soil texture) and cultural (e.g. frequency of cultivations). Any of these may affect the distribution of a soil borne pest by affecting any of the processes (reproduction, immigration, and mortality emigration) that drive population numbers. Some variables are unalterable but offer potential for predicting larval presence (e.g. altitude) and others may be manipulated by the grower to influence larval populations (e.g. crop type).

Many of the variables observed by McCracken (1995) and Parker and Howard (1997) could be expected to affect wireworms, leatherjackets, click beetles and crane flies, especially soil moisture which causes mortality at extremes (Campbell 1937; Coulson 1962; Lafrance 1968; Lees 1943; McCracken *et al.* 1995; Meats 1974; Milne *et al.* 1965). M^cCracken (1995) studied the influence of few, and Parker and Seeney (1997), no cultural variables. In organic vegetable rotations, cultivation occurs at frequent intervals and may occur more often than in conventional cropping, as cultivation is often used as a form of weed control. Cultivations disrupt the soil environment to a large degree and have a substantial effect on populations of subterranean pests (Blackshaw 1988; Fox 1961; George 1967; Green 2003; Gregory *et al.* 2005; Jedlicka and Frouz 2007; Parker and Howard 2001; Seal *et al.* 1992a). Cultivations have also been shown to decrease the number of carabids and staphylinids (Anderson 1999) which are known to consume wireworms (Fox and MacLellan 1956). Accordingly, variables for a number of cultivations were included in this survey (Table 4-1).

Parker and Seeney's 1997 UK wireworm survey found that multiple linear regression created the best model to explain wireworm distribution. This study highlighted grass duration, aspect (the direction the field faces) and soil bulk density (a measure of the weight of the soil) as the factors most affecting wireworm distribution.

M^cCracken's 1995 survey of leatherjacket populations included 24 variables of which seven were identified as having a significant effect on leatherjacket populations: aspect, tendency to waterlogging, distance from the Atlantic, the population of the previous year, silage (number of cuts in three previous years), organic fertiliser (slurry, dung or neither) and winter grass height. Some of these variables are not relevant to this study of the populations in organic vegetable rotations. In South Devon, for example, there was no considerable difference in the distance of the fields from the sea, and the height of a vegetable crop is much less homogenous than a grass crop and so these variables are not included in this study.

This survey aimed to account for as much variation in populations as possible, thus identifying factors with potential to predict populations, and those factors that might be manipulated to reduce populations. Chapter 3 showed that soil moisture and shear strength can affect the levels of populations of leatherjackets and wireworms. This chapter aims to evaluate the effect of a wider range of environmental, physical, chemical and cultural variables on the occurrence of soil pest populations. One hundred fields were surveyed within a single geographic region currently under organic vegetable production.

4.2 Methods

Ninety-nine organically managed fields from six farms growing vegetables in South Devon, UK were surveyed in 2004, all fields on each farm that were accessible. All six farms grow vegetables in a livestock-based grass rotation involving beef, dairy and/or sheep production. The 99 fields represented the variety and proportions of crops and pasture found in these types of rotation.

4.2.1 Soil core sampling for leatherjackets and wireworms

Twenty 10cm deep soil cores (see section 3.2.1 for description) were taken in February to April from each field using a 10cm diameter soil corer in a 'W' formation (Parker and Howard 2001). The samples were individually bagged in the field and then washed through sieves using high pressure water, the debris was then floated in a saturated salt solution. This method and number of samples provides a good compromise between practicality and accuracy (Mayor and Davies 1976). All leatherjackets and wireworms were collected, labelled and stored at -20°C within 48 hours of sampling. Any 'part' larvae were included if the head was present.

4.2.2 Pheromone traps for click beetles

Three Yatlor traps (see section 2.2 for description and figure illustrating Yatlor pheromone traps) each containing a sex pheromone attractant (Furlan *et al.* 2001a; Toth *et al.* 2002) for either *A. lineatus, A. sputator* or *A. obscurus* were deployed from 7th May 2004 in 92 of the 99 fields from which soil cores were taken. Seven fields from the original 99 were excluded on the request of the landowner or for practical reasons. The traps were emptied weekly and replaced in the same position. All beetles were stored at -20 °C within 12 hours of sampling. The samples from each trap at each sampling point were kept separate and identified to species. The pheromones were replaced after six weeks (as per manufacturers instructions). The traps were negligible for three weeks (12th August 2004).

4.2.3 Crane fly survey

Crane flies were sampled using water traps as described in Chapter 3 (Blackshaw 1983b). In the last week of July 2004 these traps were placed in 90 of the 99 fields from which soil cores were taken. The traps were then serviced weekly; the water was replaced and all catches were removed, recorded, identified to species and sex and stored at -20°C. The traps were removed at the end of the flight period, as identified by a cessation in trap catches (21st October 2004).

4.2.4 Field attributes

Cultural history attributes were obtained using face to face interviews with the farmers. Environmental properties were ascertained using Ordnance Survey maps. X and Y Coordinates were obtained from the National Grid using ArcMap 9.0 (Esri).

To obtain chemical properties soil samples 3cm diameter and approx 25cm deep were taken randomly from 20 sites per field. The samples were air-dried and bulked and then analysed using standard methods (MAFF/ADAS 1986). A full listing of all field attributes studied is provided in Table 4-1.

 Table 4-1 Environmental, physical and cultural variables (descriptions and abbreviations) used in the survey of *Agriotes* and Tipulid distributions

Environmental	Description
Variable	
Environmental	
altitude	Metres above sea level (range: 20-180m)
Distwc	Distance from watercourse (mm on a 1:25,000 map)
Naspect	Degrees from grid North to an accuracy of 45°
Easpect	Degrees from grid East to an accuracy of 45°
percentSlope	The percentage change in altitude of the field as a fraction of the distance of the change
Chemical	
pH	Measured on air-dried soil in the laboratory
potassium	mg/l
phosphorus	mg/l
magnesium	mg/l
salinity	EC1:5 Air-dried, ground soil was mixed with water in a
	1:5 w/v ratio The mixture was then allowed to settle
	before being measured for electrical conductivity
Physical	
5cmShear	Soil shear strength (a measure of compaction) at 5cm
	depth using a shear vane. Taken <i>in situ</i> on all fields on the same day in August, at 5 positions in each field
10cmShear	Soil shear strength (a measure of compaction) at 10cm
	depth using a shear vane. Measurements taken in situ on
	all fields on the same day in August, at 5 positions in
	each field
Moisture	Conductance in mV as a surrogate for soil moisture
	measured using a theta probe (Delta-T devices), taken in
	situ on all fields on the same day in August, at 5
	positions in each field
Percent_Sand	Sand, silt and clay percentages were measured from soil
	samples using a particle size analyser (Malvern
	Instruments)
Percent_Silt	Sand, silt and clay percentages were measured from soil
	samples using a particle size analyser (Malvern
	Instruments). Note: percentage clay was excluded from
	the analysis as it is redundant when percentages of sand
	and silt are included as variables
BulkDensity	Bulk density was calculated using the method for
	disturbed soil (Tan 1996).
PercentStones	The stone (>2mm diameter & <30mm) content of the
	soil was calculated as a % by weight of air-dried soil
OMpercent	Organic matter measured as percentage loss on
	combustion

Grass Du	Grass duration is the number of years the field has been
	in continuous grass. Permanent pasture was given an 'age' of 75 years (Parker and Seeney 1997)
Silage	The number of silage cuts was taken as a total number of
-	cuts for that field over a three-year period
YrsinGra	The number of years between 1999 and 2004 in which the field had been in grass. This allows for any effect of grass in an organic vegetable rotation
Yrs_sinc	Years since conversion, a count of the years since the conversion period of the field was completed
Org Fert	Organic fertiliser is the number of years of the last five in which either slurry or farmyard manure (FYM) was applied in any quantity
ManureQu	Tonnes Hectare ¹ of FYM or slurry applied to the field in 2003
LimeQuant	Tonnes Hectare ¹ of lime applied to field in 2003 (Mean: 0.45)
Cultivat	Number of cultivations (not including ploughing, rotavating or rolling) in 2003
Additive	Number of additive applications to the field in 2003.
Rotavate	Number of passes by a rotavator in 2003
Roll	Number of passes by a roller in 2003
Bedform	Number of times a bed (for potatoes/carrots) was formed with a mechanical bed former in 2003 (Note: Maximum is one pass)
Destone	Number of times the field underwent mechanical destoning in 2003
Weeding	Number of times the field was weeded (flame, hand or mechanical) in 2003
AvgofMan	Average tonnes Hectare ⁻¹ of FYM or slurry applied to field per year (1999-2004).
AvgofCul	Average number of cultivations (not including ploughing, rotavating or rolling) per year (1999-2004)
AvgofAdd	Average number of additive applications per year (1999- 2004)
AvgofLime	Average tonnes Hectare ¹ of lime applied to field per year (1999-2004)
AvgofRot	Average number of passes by a rotavator per year (1999-2004)
AvgofRol	Average number of passes by a roller per year (1999- 2004)
AvgofBed	Average number of times a bed (for potatoes/carrots) has been formed mechanically per year (1999-2004)
AvgofDes	Average number of times the field has undergone mechanical destoning per year (1999-2004)
AvgofWee	Average number of times a field has been weeded (flame, hand or mechanical) per year (1999-2004)

4.2.5 Analyses

The data were analysed using individual stepwise multiple regressions (Minitab®) for each species (Lawrence *et al.* 2006). Stepwise multiple regressions select the variables to produce a model with the highest R_2 and the lowest S values (alpha to enter and remove was 0.15).

4.2.6 Data processing

In order to analyse these data a large amount of pre-processing was required. The analysis methods used required the number of variables to be reduced so that the variable:instances ratio was increased and overfitting was reduced. Overfitting is where an erroneous predictive model may be created because an abundance of covariates randomly permits a well fitting model. To achieve this, the variables on soil treatments were consolidated from 6 individual variables to one variable index, which relate to the relative quantity of soil amendments used on a particular field. The same was achieved for the cultivation variables (three individual to one index variable). Percentage clay was considered redundant when % silt and % sand are included in the analysis (as 100 minus percentage sand minus percentage silt equals percentage clay) and was, therefore, removed.

An assumption of the statistical methods used here was that there should be no cocorrelation between variables. This is very difficult to achieve given the integrated nature of the data e.g. all fields in permanent pasture will have had very similar management, no ploughing and little fertiliser. Variance inflation factors (VIFs) were used to identify high levels of co-correlation between variables. Variables with high VIFs were removed so as not to violate assumptions of all analysis methods. Avg of ploughing (average number of times a field was ploughed per year over the

previous five years) and Ploughing (number of times plough in previous year) both showed very high VIFs (48.1 and 30.4 respectively) and so were removed from the analysis. Removing the ploughing variables reduced co-correlation to acceptable levels, all other VIFs were less than 20 (the standard VIF threshold).

Practical aspects of data collection resulted in an incomplete data set; some fields which were sampled for larvae (one visit) were not included in the weekly sampling regime for adults. Consequently, the number of fields used in each analysis varied. Where one week's data was not collected (e.g. trap destroyed by grazing cattle) the data was entered as a zero count.

Outliers in the variables were searched for using a Cook's D test and not found to be present. For any analysis where a normal distribution is a pre-requisite, transformed. data were used (Table 4-2).

Table 4-2 Transformations used on species count data to obtain normal
distribution of data for multiple regression.

Population	Transformation		
Female T. paludosa	Log (n+1)		
Male <i>T. paludosa</i>	Log (n+1)		
Leatherjackets	Log (n+1)		
A. lineatus	√n		
A. obscurus	Log (n+1)		
A. sputator	Log (n+1)		
Wireworms	√n		

4.3 Results

4.3.1 Summary of insect catch

All crane flies were identified to species, however, only one *T. oleracea* adult (male) was trapped. This individual was disregarded in analyses and all conclusions relate solely to *T. paludosa*. Larvae were not separated to species level as techniques were not available for click beetles and considered unnecessarily time consuming for crane flies when no *T. oleracea* adults were present. Although there is a method of distinguishing the two species as larvae using isoelectric focusing (Humphreys *et al.* 1993), this was not believed to be an effective use of resources in this investigation.

As expected, the numbers of crane flies and click beetles collected were much higher than the number of leatherjackets and wireworms. Wireworms were detected in eighteen of the 99 sampled fields, whereas leatherjackets were found in 47. Twelve fields had both leatherjackets and wireworms, although they were rarely found in the same core. Table 4-3 Crane flies, click beetles, wireworms (WW) and leatherjackets (LJ) captured 2004. n=99 for larvae, 90 for crane flies, and 92 for the three species of click beetle. N.B. Absolute populations of organisms cannot be calculated when attractant traps are used

	Larvae		Adults			
	ww	LJ	Crane Flies (<i>T. paludosa</i>)	A. lineatus	A. sputator	A. obscurus
Total catch	48	334	1173	7,342 (in <i>A. lineatus</i> trap) (3,064 in others)	4,383 (in <i>A. sputator</i> trap) (491 in others)	6,412 (in A. obscurus trap) (531 in others)
Fields containing organism	18%	47%	87%	98%	84%	96%
Mean population	30,625 ha ⁻¹	210,625 ha' ¹	11.86 per trap over season	79.8 per trap over season	47.6 per trap over season	69.7 per trap over season
Max population	500,000 ha ^{.1}	3,187,500 ha ^{.1}	91 over season	313 over season	529 over season	718 over season

The *T. paludosa* flight period began on the 16th August 2004 (Figure 4-1) and was at its maximum in the week commencing 19th September. There were more male than female crane flies caught throughout the flight period.

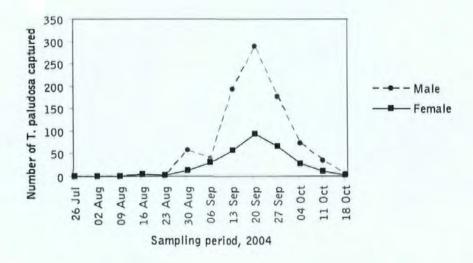
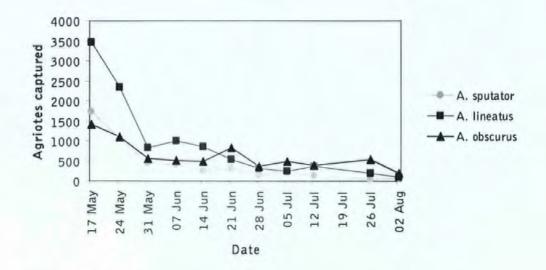
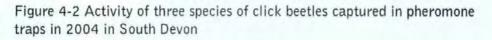


Figure 4-1 Flight period of male and female *T. paludosa* adults captured in water traps in 2004 in South Devon

Click beetles showed a less distinct flight period than the crane flies. High numbers of click beetles were captured immediately on deployment of the traps, the numbers caught dropped very quickly to moderately stable levels, which tailed down to virtually zero by the beginning of August.



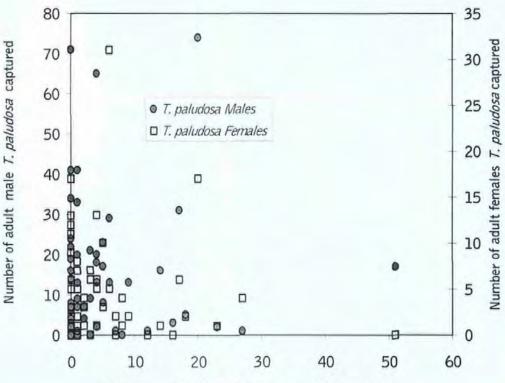


4.3.2 Larval and adult relationships

The distributions of leatherjacket and wireworm catches were both non-normal, when tested using the Kolmogorov-Smirnoff test and so Spearman Rank order correlation was used to examine the relationship between catches of click beetles in pheromone traps and numbers of larvae recovered in soil cores for all fields where paired data existed (n=90). There was not a distinct and obvious relationship between the number of larvae found in one field and the number of adults found in the same field for *A. sputator, A. lineatus* and female crane flies (Table 4-4).

Table 4-4 Results if Spearman Rank order correlation analyses of the relationship between adult catches and larval counts for each of three species of click beetles and wireworms and male and female crane flies and leatherjackets (*p<0.01)

	Wireworms		Leatherjackets	
	Rs	Р	R _s	Ρ
A. sputator click beetles	0.042	0.692	-	
A. lineatus click beetles	-0.073	0.494		
A. obscurus click beetles	0.578	<0.001*		
<i>T. paludosa</i> (F) crane flies		-	0.184	0.085
<i>T. Paludosa</i> (M) crane flies	•		0.336	0.0018*



Number of leatherjackets recovered in 20 soil cores

Figure 4-3 The relationship between leatherjacket counts from twenty 10cm soil cores per field and male and female crane fly catches in water traps during the subsequent emergence period in South Devon, 2004

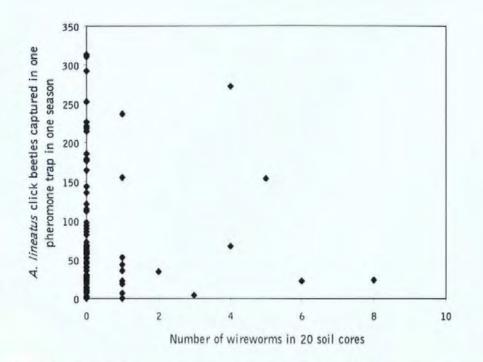


Figure 4-4 The relationship between wireworm counts from twenty 10cm soil cores per field and *A. lineatus* click beetle catches in pheromone traps during the subsequent emergence period in South Devon, 2004

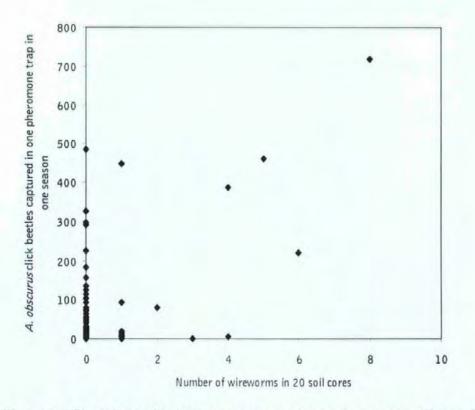


Figure 4-5 The relationship between wireworm counts from twenty 10cm soil cores per field and *A. obscurus* click beetle catches in pheromone traps during the subsequent emergence period in South Devon, 2004

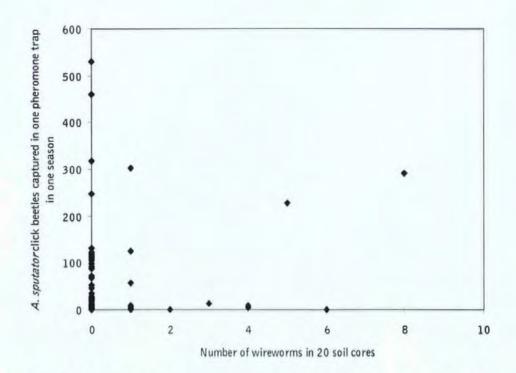


Figure 4-6 The relationship between wireworm counts from twenty 10cm soil cores per field and *A. sputator* click beetle catches in pheromone traps during the subsequent emergence period in South Devon, 2004

There is a relationship between leatherjackets and crane flies, particularly males (Figure 4-3), although this is somewhat masked visually due to the large number of fields containing zero or one leatherjacket. However, the relationship between wireworms and click beetles is less clear cut (Figure 4-4, Figure 4-5 and Figure 4-6). There is a highly significant correlation between the whole wireworm population and *A. obscurus* click beetles but no significant relationship between wireworms and the other two species captured (Figure 4-4).

4.3.3 Results of stepwise multiple regression for factors influencing the distribution of wireworms and click beetles

Variable	Coefficient	P Value
Grass duration	0.0143	< 0.001
Yrsconvert	0.060	0.007
Áspect (E)	-0.00145	0.028
AvgBedForm	-1.36	0.076
AdditiveIndex	1.192	0.029
Salinity	0.069	0.019
Silage	0.045	0.103
Constant	-3.985	1
R²	40.56	
R ² (adj)	35.48	
Model		<0.001

 Table 4-5 Stepwise multiple regression of factors influencing wireworm (sqrt)

 distribution

 Table 4-6 Stepwise multiple regression of factors influencing A. sputator (log)

 distribution

Variable	Coefficient	P Value	
Percent Stones	0.038	0.005	
Potassium	-0.00180	0.013	
0M%	0.163	< 0.001	
AvgManureQuan	0.0247	0.013	
ManureQuan	-0.0103	0.038	
YrsConvert	-0.081	0.004	
Grasdura	0.0134	< 0.001	
RecentGra	-0.224	< 0.001	
1'0cmShear	0.0086	0.002	
Silage	0.111	< 0.001	
Altitude	0.0073	0.001	
Sand [,] Percent	0.055	0.007	
Weeding	0.211	0.006	
AdditiveIndex	-0.44	0.003	
AvgAdditiv	0.46	0.003	
AvgCultiv	0.174	0.046	
AvgWeeding	-0.32	0.120	
Constant	-2.135		
R²	71.04		
R ² (adj)	64.20		
Model		<0.001	

Variable	Coefficient	P Value	
5cm Shear	0.053	0.003	-
Moisture	0.0130	< 0.001	
Bulk Density	15.2	0.005	
LimeQuantity	1.31	< 0.001	
Roll	-2.68	0.001	
Potassium	-0.0137	0.008	
Phosphorus	0.071	0.021	
Slope	0.103	0.041	
Salinity	-0.27	0.112	
Constant	-3.055		
R ²	60.81		
R ² (adj)	56.40		
Model		<0.001	

 Table 4-7 Stepwise multiple regression of factors influencing A. lineatus (sqrt)

 distribution

 Table 4-8 Stepwise multiple regression of factors influencing A. obscurus (log)

 distribution

Variable	Coefficient	P Value	
Moisture	0.00205	0.001	
0M%	0.173	< 0.001	
ManureQuantity	-0.0138	< 0.001	
Potassium	-0.00300	< 0.001	
рH	-0.35	0.003	
AvgCultiv	0.219	< 0.001	
Sand Percent	0.023	0.145	
AvgAdditiv	0.26	0.045	
Rotavate	-0.29	0.008	
Bedform	0.66	0.006	
5cmShear	0.0042	0.063	
Roll	-0.220	0.008	
Phosphorus	0.0090	0.038	
Constant	-0.5695		
R²	73.94	r	
R² (adj)	69.48		
Model		< 0.001	

The models (Table 4-5, Table 4-6, Table 4-7 and Table 4-8), produced by stepwise multiple regression for wireworms and click beetles all show a good fit although all the click beetles species show higher R² values than wireworms, but the wireworms are likely to be a complex of all three species. Multiple regression highlighted a number of significant factors that were different for each species and for wireworm

larvae. It might be expected that some of the factors affecting adult prevalence may be selected for the larvae, however, this was only true for grass duration, years since conversion, additive index and silage. Chemical, physical and cultural factors are all selected for at least one species.

4.3.4 Results of stepwise multiple regression for factors influencing the distribution of leatherjackets and crane flies

Variable	Coefficient	P Value	
Cultivations	-0.207	<0.001	
Rotavate	-0.319	< 0.001	
Slope	-0.0207	<0.001	
AvgRotav	0.63	0.008	
Weeding	-0.190	<0.001	
AvgWeeding	0.303	0.002	
Roll	0.203	0.005	
Moisture	-0.00117	0.002	
AvgBedForm	-1.08	0.027	
Distwc	-0.0137	0.037	
Constant	1.7864	1	
R ²	63.03		
R² (adj)	58.35		
Model		<0.001	

Table 4-9 Stepwise multiple regression of factors influencing leatherjacket (log) distribution

Table 4-10 Stepwise multiple regression of factors influencing male crane fly(log) distribution

Variable	Coefficient	P Value	
5cmShear	0.0091	<0.001	
LimeQuantity	-0.199	0.001	
Sand Percent	0.025	0.082	
YrsinGrass	0.073	0.031	
Aspect (E)	-0.00130	0.043	
Roll	0.20	0.095	
Constant	-0.14607		
R²	32.18		
R² (adj)	27.27		
Mode		<0.001	

Variable	Coefficient	P Value	
5cmShear	0.0063	< 0.001	
Altitude	0.00289	0.007	
AvgRoll	-0.24	0.032	
0M%	-0.042	0.039	
Bedform	-0.34	0.066	
Salinity	-0.029	0.104	
Constant	2.175332		
R²	28.56		
R² (adj)	23.39		
Model		<0.001	

 Table 4-11 Stepwise multiple regression of factors influencing female crane fly

 (log) distribution

The models (Table 4-9, Table 4-10 and Table 4-11) produced by stepwise multiple regression for leatherjackets show and crane flies show a reasonable fit. The model for leatherjackets has an R² of 63.03. However, those for crane flies have an adjusted R² of less than 30, which shows the model is only explaining a small proportion of the variation in the data.

The male and female crane flies and the larvae appear to be affected by different environmental variables. Selected attributes for leatherjacket regression models include a larger number of cultivation variables than appear in the analyses for the male or the female adults. The model for the distribution of female adults contains more variables associated with physical or chemical aspects of the soil (pH and Aspect) than either the males or leatherjackets.

4.4 Discussion

4.4.1 Larval and adult correlations

The potential effects of assuming all leatherjackets are *T. paludosa* are believed to be minimal as only one *T. oleracea* adult was present from over 1000 crane flies captured and identified to species.

Female crane flies are fully gravid at emergence and cannot travel far (Dobson 1973) and they are mated almost immediately, with oviposition beginning about one hour later (Cuthbertson 1929). They can, however, travel considerably further after depositing the majority of their egg load (Dobson 1973). Aggregations around emergence sites occur when males mate with fully gravid females (Binns 1975; Pritchard 1983), then older females emigrate with the remains of their egg load. This enables the majority of the eggs to be laid in sites with a successful history of leatherjacket development the previous year (Frouz and Paoletti 2000), whilst providing pioneers to colonise other habitats. These new habitats have a much more random likelihood of being a suitable leatherjacket and crane fly habitat, but females whose offspring are located in more than one habitat have a higher chance of at least some offspring surviving to reproductive age. In this study leatherjackets were found to correlate significantly with male numbers and with total population, but not statistically significantly with female populations. This would correspond with males aggregating for mating around suitable larval habitats and females initially having a strong association with the same, but dispersal making this association less strong.

The strong correlation between leatherjackets and crane flies further validates the use of water traps for monitoring populations for pest management purposes (Blackshaw 1983b; 1987b). Although the correlations are not significant for all of the click beetles studied, the R_s values of some are reasonable, <40% for tipulids and <60% for *A. obscurus* and thus may be of satisfactory value for pest-management purposes. However, threshold populations of these two species are very low and the potential for loss in the event of an incorrect prediction is very high. The knowledge of which environmental factors support high populations of leatherjackets enables further precision when estimating risk of damage when low numbers of adults are caught.

Sampling was not initiated at the start of the click beetle emergence period due to practical difficulties. The peak capture rates (Figure 4-2) correspond to the time of peak emergence of *A. sputator* in Wales (Parker and Howard 2001). The pattern of collection shown shows many fewer beetles and much less variation week-by-week than that found by Furlan *et al.* (2001b), who collected click beetles in 2001 at a location very close to this study. A peak is shown around July in the Furlan data, however, the click beetles in this experiment showed a gradual reduction in catches and no peak at this time. This may be due to weather conditions initiating a peak prior to the start of sampling, or it may be possible that the farms had a very high existing population. It is not known whether click beetles normally survive overwinter. If this study initially captured a large number of overwintered click beetles, this may explain the high initial populations and the 'hidden' peak in July.

A positive association of *A. obscurus* with wireworms was also observed by Blackshaw *et al.* (2006). This is difficult to explain but there are implications for using pheromone traps in pest monitoring systems. There is a possibility that pheromone traps for *A. obscurus* only will provide a representation of the wireworm. population underground, whereas the current system of adding the catches of the three species together¹ would not be representative of the subterranean population. It is necessary to examine the different responses of the three *Agriotes* species to their pheromones as different behaviours may be evident.

The reasons for *A. lineatus* and *A. sputator* click beetles failing to correlate with larval populations may be a sampling deficiency. Soil samples were randomly taken from the whole of the field, but only one pheromone trap (of each species) was placed near the centre of the field. Ranges of attraction were not known for these pheromones and thus pheromones may have covered part of the field, the whole field or a number of fields and their surrounding features. It is necessary to determine the ranges of attraction for each species' pheromone. The limiting factor of the range of attraction may be the speed and distance at which the insects can travel. If the insects are capable of travelling large distances in a limited time, then finding correlations between soil sampling and pheromone traps is less likely over a prolonged sampling period. It may also be that random sampling is not suitable for this style of investigation and a small systematic sampling scheme would have been a better alternative. Systematic sampling would also enable determination of any patchiness within a field but is extremely labour intensive and beyond the scope of this study. There is no evidence that catches of A. lineatus and A. sputator in pheromone traps can predict wireworm populations within the same year. It is therefore highly unlikely that they could be used to predict damage for future years. The long subterranean section of the lifecycle also makes prediction of wireworm populations from the emergence period of one year very difficult.

¹ See Chapter 6 for details of current advice for using commercial pheromone traps

Wireworms have a larval stage of many years, so the wireworm complex in any field is likely to contain larvae of many different ages. Correlating the adults emerging in one year with the whole complex of wireworms is a not straightforward.

There was, inevitably, a delay between larval sampling in January and February and the emergence period for the target organisms. Nothing is known about larval migration of these species and it is possible that migration by the non-correlated species had occurred between these periods. Larvae may have moved to areas with more favourable soil moistures as the seasons change and the patches of differential soil moisture within a field vary.

4.4.2 Agriotes models

The number of years in continuous grass (Grasdu) was the single most important factor for defining wireworm distribution. This fact is well known (Fox 1961; Jedlicka and Frouz 2007; Parker and Howard 2001; Parker and Seeney 1997; Strickland *et al.* 1962) but provides reassurance of the validity of this survey. It has always been believed that this has been due to a plentiful food source and stable conditions (Miles 1942). As this survey is looking specifically at organic rotations, the number of years in grass (yearsingras) was included as a separate value. This variable counts the number of years of the last five in which the field has been in grass (resulting in a number between zero and five). Five years was chosen as the maximum in the life of any wireworm found in this experiment. The multiple regression showed that this variable is positively correlated with wireworm

populations, suggesting that populations begin to develop over a period as short as would be expected in an organic rotation.

Distance from watercourse was identified as influencing click beetle distribution in Chapter 2. Although the distance from the nearest watercourse was included as a variable in this analysis, it was not found to have a significant effect on the distribution of either wireworms or click beetles.

In this survey, moisture was recorded on a single occasion and thus is a relative snapshot of field conditions, the measurement was taken during a period of high rainfall and may be perceived as an indicator of waterlogging. Previous research has highlighted ways that wireworms react to excess or lack of water in their environment (Lefko *et al.* 1998; Parker and Howard 2001; Salt and Hollick 1946). Variation in moisture can occur either spatially or temporally. Looking at temporal changes in soil moisture for wireworm habitats was outside the scope of this study, and detailed spatial analysis of soil moisture and wireworm distribution in Chapter 3 showed no outstanding patterns. Jedelicka and Frouz (2007) also found a statistically significant association of wireworms and moisture using PCA which was not evident as a direct correlation between moisture and wireworms (*A. obscurus*).

Parker (1997) found that compacted soils were unfavourable for wireworm populations. Soil compaction (measured as soil shear at 5cm depth) was not identified as being related to wireworm populations in this study. Salt and Hollick (1946) also did not find any correlation between wireworm populations and soil compaction, nor did Chapter 3 of this work. However, soil shear strength (at 5cm or 10cm) was identified as a factor influencing the distribution of all three species of click beetles. This was unexpected but may be a result of soil shear influencing click beetle emergence.

Many studies have found that organic production systems create environments that are more favourable for invertebrate populations (Berry *et al.* 1996; Hole *et al.* 2005; Stockdale *et al.* 2001). The increase in wireworm population correlated with years since conversion may be an indication of the creation of these better conditions. This increase in invertebrates is normally interpreted as beneficial for pest management purposes as an increase in beneficial insects is also seen, but in this instance it may be increasing the pest population itself, exacerbating the problem. It is, however, worth noting that many of the fields in this survey have been managed organically for less than five years and so the ecosystem may not yet have matured to create a more sustainable predator/prey balance. Or perhaps, the principle is not in practice here: Armstrong (1995) found that the diversity and abundance of carabids was reduced in organically managed potatoes.

Nadvornyj (1968) looked at moisture, pH, temperature and bulk density and found that only soil texture contributed to explaining elaterid distribution. The elaterids identified were primarily the three species in this survey, but there were an additional 38 species found in Russia. However Nadvornyj (1968) found that *A. obscurus* were negatively correlated with percentage sand, whereas this study has shown a positive correlation between sand and *A. sputator* populations. Strickland (1962) found higher levels of damage in sandy soil; this could be as a result of higher populations, whereas Ross (1948) found more damage with low populations. Unfortunately Nadvornyj's paper does not give quantitative values for any conclusions drawn. Nadvornyj also reported a positive correlation between *A. obscurus* and percentage sand which was not found in this study. Fox (1961) found a positive link between sand and population of *Agriotes* click beetles. Percentage stones also showed an influence on *A. sputator* distribution, the quantity of sand and stones in the soil will influence the drainage of the soil and thus, may influence populations.

The potassium indices for the farms studied tended to be low, although organic production is said to increase those (Stockdale *et al.* 2001). The fields in this study have very low pH's, only two are above 7.0 and half are below 6.0. Soil pH is unaffected by organic, as opposed to conventional production. In this study, potassium and pH are negatively correlated with *A. obscurus* distribution, this concurs with 'Wireworms and Food Production' (Anon. 1948), where acid soils were found to have a tendency for higher populations of wireworms. Ibbotson (1958) found that *A. obscurus* wireworms were dominant in low pH, in agreement with the negative correlation of pH and *A. obscurus* beetles here. However, 'Ladell (1938) found no overall relationship between wireworms and pH but in this experiment the species of wireworms were not determined. This is further evidence for the need to be able to accurately identify wireworm to species level (Ellis *et al.* 2009).

A. sputator and *A. obscurus* had positive correlations with the percentage of soil organic matter, this was also found by Salt and Hollick (1946) for wireworms. This has implications for organic farming specifically as evidence shows that this mode of production increases the percentage organic matter in the soil (Munro *et al.* 2002; Stockdale *et al.* 2001). Salt and Hollick (1946) attributed the correlation they found

to the consumption of organic matter by wireworms. However, Traugott (2006) has shown, using stable isotope analysis, that in a maize field *A. obscurus* eat either weed roots, maize roots or a mixture of both but not soil organic matter. But, an increase in soil organic matter may be related to an increase in plan root density and thus a more plentiful food supply for wireworms.

Manure quantity, the variable measuring the amount of manure spread on the field in the previous year, was found to be negatively correlated with *A. obscurus* and *A. sputator* population size. None of the farmers surveyed in this project applied over the legal maximum FY.M or slurry. There is an indication that future work should look at quantities, types, frequency of application on wireworm growth and populations. If wireworm populations were reduced by the application of manure, this has the potential to be a simple and cost effective, short-term pest control measure. It also has the advantage of increasing soil fertility, compensating for crop losses and also assisting crops defence against pest damage (Altieri and Nicholls 2003; Blackshaw and Newbold 1987; French *et al.* 1990). However, the potential' long term increase in organic matter increases pest problems. This is in contrast to many studies (reviewed by Altieri and Nicholls (2003)) that indicate that crops grown with the addition of organic fertilisers suffered fewer pest problems.

When the three species and wireworms are analysed individually it appears that the distribution of each species is being driven by its own set of variables, the variation in the three species is the overriding finding. More interestingly, the factors affecting wireworm distribution on the whole are not a subset of the factors affecting the three adults.

4.4.3 Tipulid models

The correlation of male and female crane flies is likely to be due to the short length of the adult stage for this organism. The adults only have a few days in which to mate and reproduce before dying. Therefore it is to be expected that the male and female distributions are closely linked.

Moisture has a negative correlation with leatherjackets. As these measurements were taken in extremely wet conditions, they may be taken to confirm waterlogging winter conditions and subsequent mortality of the leatherjackets (Blackshaw 1983a; Blackshaw and Perry 1994; Meats 1970; Pritchard 1983) and is more clearly defined than when examined on a single field scale. In Chapter two, the distance from the nearest watercourse was found to be related to crane fly distribution. Distance for watercourse was selected as a predicting variable for leatherjacket distribution in this analysis.

Cultivations, rotavations, average number of bed formations over five years and manure quantity each showed the same pattern. As expected from work by Blackshaw (1988) leatherjacket populations were strongly negatively correlated with the number of cultivations. Leatherjackets were strongly negatively correlated with slope, indicating that leatherjackets are to be found in higher numbers on flat fields, and less on sloping ones. It is possible that this may be due to female ovipositioning behaviour.

Weeding variables, both 'average over 5 years' (near significant) and 'one year count' (significant) were selected in the model of leatherjacket distribution. The average number of weeding occurrences over the previous five years showed a positive relationship with leatherjacket numbers , however an increase in weeding in the one year previously, coincided with a decrease in leatherjacket population. This may be due to the physical process of weeding itself (either machine or hand) or exposing leatherjackets to predators or the removal of alternative food sources. This also applies to cultivations, including bed formation and rotavation (one year and five year) (Pritchard 1983).

Variables selected for crane flies only could be directly related to site selection for ovipositioning. For example the aspect of the field (measured as degrees from North) may relate to soil temperature from ambient solar radiation, salinity may directly affect egg survival & 5cm soil shear strength may enable eggs to be positioned at the correct depth. Unfortunately none of these factors can be manipulated to reduce oviposition, as they may have provided potential for dissuading oviposition in vulnerable crops.

The adjusted R² of the multiple regression models for relating leatherjacket populations with environmental variables (58.35%) was greater than that for either male or female crane flies (27.27% and 23.39%). This is probably because variables were selected for inclusion into this survey by reviewing the literature. As the primary need is to determine leatherjacket numbers (as opposed to adults) there is a bias within the literature to highlight variables associated with larvae and not adult stages. Wireworms and leatherjackets were found not to share any environmental determinants with the exception of the average number of time a bed is formed over five years. As bed formation only occurs for a potato crop, any management strategies for these pests will have to be specific for each and it is unlikely to be possible to develop one 'organic' strategy for the control of all soil insect pests. However a different form of a cultivation variable (e.g. the number of rotavations or rolling occurrences) was selected for each larval type. Overall, the variables selected by the wireworm model could be of use to identify fields at risk of high wireworm populations e.g. aspect, years in grass, but would not be easy to manipulate with the intention of reducing wireworm populations. However, the variables selected to explain leatherjacket distribution include a number that may be suitable to integrate into rotations as control methods e.g. rolling and rotavations. The effect of cultural practices, such as cultivations will be examined in Chapter 5. The range of attraction of the pheromone traps also requires clarification when assessing risk control strategies. This will be addressed in Chapter 6.

Chapter 5 Cultural Control

5.1 Introduction

The subterranean environment of soil pests is highly disturbed by cultural practices such as cultivation and populations may be strongly affected by the availability of food sources. This potential for pest control has been neglected since the advent of pesticides (Katan 2000) and is an area now being identified as requiring further research (Peigne *et al.* 2007). The individual effects of cultural practices could not be fully assessed with the survey methods used in previous studies (Chapter 4). Consequently, this chapter uses manipulative techniques to assess the effect of rolling and crop type at greenhouse and plot scale and the effects of cultivations at plot and field scale. Knowing where a pest may be found (at any scale) is beneficial, but much greater benefit could be derived from the ability to reduce the population of the pest to a level at which its harm is minimized. Cultural control methods avoid many of the problems of chemical or biological control. There are no toxic residues and pest resistance is not a potential problem.

The current advice given to organic growers to prevent damage by pests is primarily prevention, with some methods for cultural control of leatherjackets suggested, although they differ in the methods suggested to reduce populations. The Soil Association briefing paper advises organic farmers and growers about leatherjackets (2003), recommending ploughing before September, cultivations (including hoeing) during crop growth and nematode application. The SAC (Scottish Agricultural College) advice leaflet (1999) recommends that soil should be rolled to prevent movement and feeding of leatherjackets and recommends cultivations to cause mortality. ADAS (1984) advised rolling to control damage to crops and encouraged cultivations to decrease populations. The SAC technical summary (Coll 2001) recommends an additional cultivation when populations are high, rolling of the seed bed and harrowing for weed control as a method of leatherjacket control. Rolling in the summer whilst leatherjackets are pupating is recommended to reduce leatherjacket numbers, the theory being that rolling caps the soil making it impossible for the crane fly to emerge (ADAS, 1984). Thus the lifecycle is then broken and the leatherjacket population is greatly reduced for the following year.

An ADAS advice leaflet (1983) on wireworms recommends disturbance of the soil to reduce wireworm damage, especially early in the year when the insects are pupating and entombed in subterranean cells which can be disturbed by cultivations (Miles 1942). Cultivation also causes direct injury and exposure (to desiccation and predation) (Anon. 1948; Miles 1942). The same 1983 ADAS advisory leaflet highlights the incorporation of crop or weed residues as very necessary. Parker and Howard (2001) stress the ability of repeated cultivations to decrease a wireworm population year on year, however, they observe that this decrease is not by any standard amount. Parker and Howard also recognize that a firm tilth restricts the movement of wireworms within the soil. Much of this advice is presented in the form of observations not experimentation, little empirical evidence is published but it is an area requiring further research (Peigne *et al.* 2007).

The 'Wireworm and Food Production' survey (Anon. 1948) looked at populations of wireworm in grass from 1940-41. These populations were generally high compared

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to those identified in this study so far. The populations of wireworms of fields that were cropped with arable crops were reduced to a greater extent than those which were not, leading to the hypothesis that it may be possible to control wireworm populations by cultivation.

One of the few recent published experiments on cultivation mortality in subterranean pests was a comparison of mortality of leatherjackets under a number of cultivation regimes (Table 5-1). This study was found that full seed bed preparation caused, approximately, a 70% reduction in number of leatherjackets (Blackshaw 1988).

Table 5-1 Effect of cultivations on leatherjacket populations, from Blackshaw (1988)

Treatment	Effect
Ploughing	Non significant reduction
Discharrow	No.effect
Rolling	Significant reduction
Rotavation	Significant reduction
Rolling <u>plus</u> rotavation	No additional reduction

Many studies have observed that wireworm populations are significantly different under different crops. Work on wireworm larvae outside the UK has reported that the inclusion of cowpeas (black-eyed or yard-long beans), peanuts, soy beans, and cotton in a rotation can cause an increase in damage and also that an unnamed species of beans can cause a reduction in damage to subsequent crops (Nettles 1940; Schepl and Paffrath 2004b; Seal *et al.* 1992a). Nash and Rawlins (1941) found that altering the crops in a rotation had a substantial effect on wireworm damage to potatoes, with maximum damage when potatoes follow permanent pasture and reduced damage when manure or beans (soy and field) are included in the rotation. Within the UK Frost *et al.* (2002) found no significant difference in damage in potatoes when mustard was sown as a green manure. It has been known for many years that peas and beans are more resistant to wireworm attack than grass (Miles 1942). The Wireworm and Food Supply Survey found that wireworm populations were reduced by approximately half in fields growing beans (Anon. 1948). But more recently, there has been evidence that having peas or lupins as the nitrogen-fixing element in a rotation can reduce damage levels in subsequent potato crops (compared to clover in the rotation) (Schepl and Paffrath 2004a).

The aims of these studies are to:

- a. Investigate the effect of rolling on crane fly emergence
- b. Investigate the effect of crops on wireworm populations
- c. Investigate the effect of cultivations on leatherjacket and wireworm populations
- d. Determine the difference in weight gain between wireworms feeding solely on wheat and those feeding soley on peas

5.2 Leatherjacket Soil Rolling Trial

5.2.1 Method

One permanent pasture field at Seale Hayne, Newton Abbot, Devon, with a high population of leatherjackets was identified (from previous, unreported work). The field was divided into four plots 2.7m wide and 100m long. Two of the four plots were rolled using a Kidd 4 ton water ballasted flat roller. Three passes of the roller were made as the ground was particularly hard at this time. Four emergence traps (Figure 5-1), modified from (D'Arcy-Burt and Kell 1988) were placed on each plot on 8th September 2005 and 6th September 2006 (approx. 1 week before emergence was due to start). Any gaps between the trap and the soil at ground level were filled with sand.

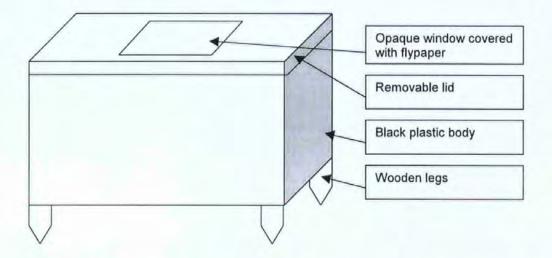


Figure 5-1 Emergence trap for crane flies. The wooden legs are hammered into the ground and all emerging crane flies are trapped within the trap and then captured on the fly paper. The trap was 45cms high.

After one week the lids were carefully opened and the flypapers were removed, replaced and inspected for crane flies. Measurements using a theta probe to provide conductance as a surrogate for moisture were taken at the same time. After traps were removed, the ground was closely visually inspected for discarded pupal cases.

5.2.2 Results

The traps were emptied weekly for the six weeks covering the crane fly emergence period in autumn 2005 and again in 2006. However, no crane flies were recovered in either year.

5.2.3 Discussion

The soil had been thoroughly sampled for leatherjackets in the spring of 2005 and found to have been supporting a high population (>1,000,000ha⁻¹). Traps were positioned in specific areas identified as areas of high populations during previous unreported work. However, throughout the experimental period not one crane fly was captured on the flypaper in either the rolled or the unrolled plots. In addition, there were no crane flies present freely within the trap that had not been captured on the flypaper. It was confirmed that there was no other means of escape for the crane flies. The area where the bases of the traps met the ground was inspected closely and only one showed a small opening suitable for crane fly escape. On deconstruction of the experiment the ground beneath the traps was inspected closely and no pupal cases were found.

In 2005 the non-emergence of the crane flies may have been as a result of a period of hot dry weather for six weeks previously, this may have had the effect of 'capping' the ground, similar to the expected result of the rolling. This hypothesis may be supported by the fact that large numbers of crane flies were only observed around the gateways and a stream running centrally through the field. These areas would have been less prone to drought and so may have allowed the emergence of the flies. There was a higher rainfall throughout summer 2006, although in this year an adjacent field was seen to have large numbers of crane flies emerging throughout the duration of the experiment.

An alternative hypothesis is that the traps (being constructed of black plastic composite) may have heated the air within the traps to a level that damaged insects beneath. Repeating this experiment with higher numbers of traps would permit confirmation of the validity of the trap use.

5.3 Investigation of Crop Effect on Wireworms

5.3.1 Greenhouse trial

5.3.1.1 Methods

On 27th June 2005, 57 plant pots, 20 cm diameter were filled with compost, then planted with either peas (*Pisum sativum* L.) (29 pots) or wheat (*Tritium aestivum* L.) (28 pots). Winter wheat (var. Ashby) was sown at 200 kg ha⁻¹ (15 grains per pot) and the Peas (var. Magnus) at 200 kg ha⁻¹ (three peas per pot).

Wireworms were weighed to 10⁻⁵g accuracy and one was added to each pot on the 7th July 2005 ensuring that equal numbers of similarly sized wireworms were added to peas and to wheat (Table 5-2).

Table 5-2 Average and standard deviation of wireworms when added to pots of peas and wheat

Crop	Average of Initial weight	Standard deviation of initial weight
Peas	0.0255g	0.0101g
Wheat	0.0238g	0.01′03g

The pots were placed in an unheated greenhouse and watered daily. The pots were dismantled on the 29th November 2005 and the recovered wireworms were weighed and the growth stage recorded.



Figure 5-2 Pots containing peas or wheat sown at field rates to establish differences in wireworm growth, each pot contained one wireworm. Drainage holes of pots were covered in mesh.

The experiment was repeated in 2006, for a shorter duration to prevent wireworms pupating (see section 5.3.1.2), the wireworms were added to 16 pots (8 containing wheat, and 8 peas) on 15th June 2006 and removed 8 weeks later.

5.3.1.2 Results

When the 2005 experiment was dismantled, 16 wireworms were not recovered, two were dead and a further 6 had pupated (Table 5-3). In 2006 only 6 wireworms were recovered. These data were considered insufficient and are not included in the analysis (Table 5-3).

Life stage	Year	Co	unt of inse	cts
at end of experiment		Peas	Wheat	Total
INITIAL NUMBER	2005	28	29	57
Click Beetle	2005	4	2	6
Corpse	2005	1	1	2
Nothing	2005	7	9	16
Pupa	2005	6	2	8
Wireworm	2005	11	1:4	25
INITIAL NUMBER	2006	8	1'0	18
Corpse	2006	0	1	1
Nothing	2006	4	4	8 .
Pupa	2006	0	1	1
Wireworm	2006	4	2	6

Table 5-3 Number of wireworms of each life stage recovered when experimental pots containing peas or wheat were dismantled in 2005 and 2006

Of the insects that remained wireworms at the time they were recovered there was a large difference in the growth weights of the larvae between each crop type in both years of the experiment (see Table 5-5). However, wheat promoted the largest weight increase in 2006 (albeit with only six wireworms) whereas peas caused the largest weight increase in 2005. An independent samples t-test reported no significant difference between the growth in those living in peas and those living in wheat (T=0.189, P=0.485) (Table 5-4).

Table 5-4 Average and standard deviation of wireworms when recovered from pots of peas and wheat

Crop	Average of end weight	Standard deviation of end weight
Peas	0.0382g	0.0152g
Wheat	0.0370g	0.0438g

Table 5-5 Mean percentage change in weight of wireworms grown in peas or wheat in 2005

	Peas	Wheat
2005	+53.20%	+38,94%
2006	+18.9%	+47.6%

5.3.2 Plot scale crop effect

The same crops (peas and wheat) were assessed for their effect on wireworm populations at semi-field scale in plots.

5.3.2.1 Methods

Twelve plots, $4m \times 10m$, with grass borders of 0.5m, were randomised to be cropped with either peas (200 kg ha⁻¹) or wheat (192 kg ha⁻¹). The experimental site was at Seale Hayne Farm, Newton Abbot, Devon and had not been ploughed for more than 10 years. The pasture was ploughed on 6th May 2005, and the plots underwent three cultivations, (disc, ring-rolled then rotavated). The plots were then heavy springtined and crumbler-rolled to produce an acceptable tilth, and were drilled the following week. The plots were sampled by taking six standard soil cores from the midline of each plot on 9th November 2005. The soil cores were washed and floated in saturated salt solution. The wireworms were then collected and identified as *Agriotes* or non-*Agriotes* species. The experiment was repeated the following year, repeating the treatments in each plot. Ploughing occurred on 4th May 2006, drilling on the 10th May, and cores sampled on 18th October.

5.3.2.2 Results

The results (Table 5-6) show that too few wireworms were recovered to carry out any meaningful analysis for either *Agriotes* or non-*Agriotes* genera.

Table 5-6 Number of wireworms found in soil cores from field plots containing either peas or wheat

	2005		2006	2006	
	Peas	Wheat	Peas	Wheat	
Agriotes species	1	3	1	2	
Non-Agriotes species	2	10	6	0	

5.3.3 Discussion of greenhouse and field trials

No difference in growth or populations was identified in these two experiments and the results are very limited, so it is not possible to confirm the observations of Schepl and Paffrath (2004a). It therefore cannot be recommended as a method to control wireworms on this occasion.

The primary problem in the greenhouse trial was the availability of wireworms. Those used in the greenhouse experiment were collected from a grass field and consequently cannot be identified to species, nor can their age be estimated. This inability to reliably identify the species of individual wireworms whilst they are living means that any differences in the responses of different species or development stages. may be obscured (Seal *et al.* 1992b). In order to minimise age effects the wireworms were applied to treatments in the greenhouse experiment in alternate size order.

The low numbers of *Agriotes* wireworms recovered complicated this plot experiment. There are normally low proportions of non-*Agriotes* wireworms and therefore the whole wireworm complex is traditionally treated as a unit (Finney 1946; Salt and Hollick 1946). Some non-*Agriotes* species are known to be herbivorous pests of agricultural land and are therefore of interest in this experiment, but some non-*Agriotes* species may be carnivorous (Traugott *et al.* 2006), so when high populations are present there is additional difficulty in interpreting results due to the potential effects of carnivory.

The problems of a mixed wireworm species assemblage may have been reduced if the experiment had included greater numbers of wireworms. In this case, it would have been more likely to have all species and age groups represented in the experiment. After all, from a practical viewpoint, farmers will not know which species are in the damaging population in their fields. If the experiments were run with greater numbers of wireworms any effects may have been visible despite not identifying to species level.

5.4 Cultivation

When the numbers of wireworms recovered from the field plot trial (section 5.3.2) were investigated for potential population reductions under cultivation, no significant differences were found (T = 0.23, P = 0.826) between cultivated and uncultivated plots (Table 5-7).

	Mean wireworms recovered	Mean <i>Agriotes</i> recovered	Standard deviation wireworms	Standard deviation <i>Agriotes</i>
Uncultivated	34,722	69,444	208,333	290,388
Cultivated	231,481	69,444	665,139	375,713

Table 5-7 Mean population of wireworms per hectare for plots which had either undergone standard arable cultivations or had not been cultivated

The pre- and post- cultivation populations from the systematic sampling experiment in Chapter 3 can also be examined. Table 5-8 and Table 5-9 show data from this chapter marked as cultivated if the field was ploughed and planted between the two sampling dates and uncultivated if the field was not ploughed and cropped. This provides data for leatherjackets in addition to that for wireworms, allowing the change in population after cultivation to be compared to the change in population without cultivation using a Mann-Whitney U test.

Field	Number of cores	2005 Population (000's)	2006 Population (000's)	% change in population	Cultivated
HPA (b)	72	174	174	0%	No
HPF	72	260	69	-73.5%	No
HPA (a)	58	237	237	0%	Ňo
HPC (cores)	74	68	203	+198.5%	No
Bakers Park	61	1,004	25	-97.5%	Yes
Broad Park	68	680	0	-100%	Yes
Hills (a)	76	16	17	+16.3%	Yes
HPG	71	458	53	-88.4%	Yes

Table 5-8 Mean population of leatherjackets per hectare for individual fields for each of two years showing whether the field underwent cultivation for cropping between the two sampling events

There was no significant difference in the median value of percentage population change between the cultivated and uncultivated fields (W=20.0; P=0.663) for leatherjackets. Although with such a small number of data points it is unlikely that a significant difference could have been observed.

Table 5-9 Mean population of wireworms per hectare for individual fields for each of two years showing whether the field underwent cultivation for cropping between the two sampling events

Field	Number of cores	2005 Population (000's)	2006 Population (000's)	% change in population	Cultivated?
HPA (b)	72	729	573	-21.4%	No
HPC (traps)	90	556	83	-85.1%	No
HPF	72	35	17	-51.4%	No
HPA (a)	58	43	280	-551.2%	No
HPC (cores)	74	304	270	-11.2%	No
Bakers Park	61	61	275	+350.8%	Yes
HPG	71	35	0	-100%	Yes
Broad Park	68	18	42	+133.3%	Yes
Hills (a)	76	0	34	N/A	Yes

There was no significant difference in the median value of percentage population change between the cultivated and uncultivated fields (W=19.0; P=0.1779) for wireworms.

The evidence from this experiment is very sparse but shows no satisfactory evidence for the use of cultivations for reducing leatherjacket or wireworm populations.

5.5 Discussion

This series of experiments has provided no further evidence to confirm or deny the efficacy of recommended organically-compliant methods for the control of leatherjackets or wireworms. What is clear is that these control methods do not have the same clear-cut efficiency of chemicals (Blackshaw and Newbold 1987; Hancock *et al.* 1986; Strickland *et al.* 1962). In contrast to the situation in this experiment, a grower following advice to roll to prevent leatherjacket emergence would have been delighted to have a zero emergence of crane flies and would be unaware that there would be no emergence in the absence of rolling. A grower relying on the mortality effect of cultivations to reduce a pest population to non-damaging levels would have been at risk of suffering economic loss as a result of the post-cultivation wireworm population in 'Bakers Park' (Table 5-9). The post-cultivation populations of wireworms and leatherjackets in Table 5-8 and Table 5-9 are an example of why sampling for population estimates is always recommended as an important part of a pest management strategy.

The use of rotations is very important in organic vegetable production and is already constrained by a plethora of considerations e.g. fertility building, prevention of

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disease, commercial demands, logistical and geographical demands (e.g. field topography not suitable for necessary machinery). The experiments in this chapter have focused on control methods that are already being recommended and are trusted by growers. However, organic production uses many techniques not commonly used in conventional agriculture that may also have an impact on subterranean larval populations. For example, covering crops with fleece is a commonly used method of reducing aerial pests, as when protecting carrots from carrot fly and this may be expected to have an effect on the soil and potentially the larval population beneath it although this is currently uninvestigated.

This chapter attempted to address the potential control opportunities of cultural practices; in particular rolling, cultivations and crop type were inconclusive in this study. To address these research questions fully would require a much greater research effort, which was beyond the limits of this research project.

Chapter 6 Efficacy of Pheromone Traps for *click* beetle species

6.1 Introduction

Pheromone traps for click beetle have only recently become commercially available (Toth *et al.* 2002) but the responses of individual click beetles and populations to pheromone trapping has not been published for UK click beetle species. The efficacy of pheromone traps for mass capture, their area of effectiveness, implications for adult migration and the relationship between below ground larval populations and adult click beetle catches are all factors for which there is no information currently in the public domain. These factors all require elucidation before the potential of pheromone traps can be maximised. This chapter describes interactions between click beetles and their pheromone traps and focuses on the sex ratios of beetles captured in pheromone traps, the effect of direction on trap catches, the speed of beetles' travel, and the range of attraction of the pheromone traps.

Pheromone traps for click beetles (section 2.2) are currently on the UK market, sold by Syngenta for the purposes of monitoring wireworm populations under the name of Nemathorin Pheromone Traps. The manufacturer's advice is to deploy one speciesspecific pheromone trap for each of the three species, to sum the catches and then evaluate the risk from wireworms based on Table 6-1.

Season-long trap catch per set of three traps	Predicted wireworm population per hectare	Approximate risk of damage to potatoes	
Nil	No or very low infestation	No or very low risk	
Fewer than 50	25,000 to 150,000	Some damage likely	
50 to 100	150,000 to 250,000	Significant damage likely	
More than 150	More than 250,000	Severe damage likely	

Table 6-1 Syngenta classifications of click beetle catches for risk to potatoes (From: <u>http://www.syngenta-crop.co.uk</u>)

High populations of wireworms show spatial structure in the form of patches and gaps (see Chapter 3), and it is likely that this will influence their trap capture rates, mediated by trap recruitment distances. For example, if recruitment distances for the pheromone traps were very small then the number of beetles captured would be highly variable depending on its positioning relative to patches. In contrast, if recruitment distances for the pheromone traps are very large then non-cropped areas of a field (hedges, neighbouring woodland etc) may be sampled inadvertently.

Parker and Seeney (1997) propose that wireworm densities are highly influenced by the initial colonisation of new pasture by click beetles. Knowledge of the distance travelled by the beetles is vital to predict possible reservoirs of populations that could recolonise a site after the original population had been controlled. Calibration of the efficacy of pheromone traps would allow calculations of the density of the resident click beetle population to be made from catches. Without this information catches can only be measured comparatively. If the three beetle species show different reactions to the pheromone then Table 6-1 trap counts could be modified to allow species specific weightings to be included. This could potentially provide a much tighter relationship between pheromone trap catches of click beetles and wireworm populations than are currently possible, as current commercial methods of monitoring is so poor that improvements are probably not difficult to achieve. Pheromone traps have also been suggested as a method for removal trapping, but without characterisation of the pheromone traps, this would be impossible to implement.

Gouch and Evans (1942) conducted preliminary work on click beetle movement, releasing 831 *A. sputator* and recapturing with traps constructed of simple grass bunches. After twelve days, thirteen beetles were recaptured: 7 within 10 yards, 3 within 15 yards, 2 within 20 yards and 1 at 30 yards distant from the release point.

Pheromone traps are not thought to capture females, however, the sex ratios of pheromone trap catches have not been published. Work is required to determine whether beetles captured are all male and being captured as a result of attraction to the pheromone, and not as a result of the YATLOR trap acting as a pitfall trap. Borg (1973) found the sex ratio of *A. lineatus* was 1:1, that of *A. obscurus* and *A. sputator* is unpublished. The current best estimate at present is that the total numbers of click beetles are double that of male click beetles, which may or may not be the total click beetle catch.

The method for the work detailed in this chapter is based on that used by Östrand and Anderbrant (2003) for the capture of European pine sawflies using pheromone traps, which is in turn, based on Turchin and Odenaal (1996). Using the equation:

$$\alpha = \frac{T}{B} = 2\Pi \int_{0}^{n} rP(r)dr$$

from Turchin and Odenaal (1996) where T is the number of beetles captured in a trap, B is the population density (the number of beetles in a specified area), r is recapture, P(r) is the probability of recapture and d is the distance from the trap. The effective sampling area (α) can be calculated, this defines the area around the trap where it is assumed that all trapped click beetles originate from within.

The aims of this study were to:

- a. quantify the spatial parameters of pheromone traps employed to capture adult male click beetles
- b. test the null hypothesis that there are no differences in the responses (speed and distance) of *A. lineatus* , *A. obscurus* and *A. sputator*
- c. investigate the sex ratios of click beetles captured in pheromone traps
- d. elucidate the speed of travel of *A. lineatus*, *A. sputator* and *A. obscurus* click beetles

6.2 Methods

6.2.1 Sex ratios of click beetles in pheromone trap catches

Three Yatlor traps were placed in a permanent pasture field at Home Park, Totnes, Devon (SX 7392 6051). Each trap contained one of each of the pheromone attractants for *A. lineatus, A. sputator* or *A. obscurus* (see section 2.2). The traps were set on 11th March 2004 and emptied at weekly intervals thereafter. The experiment was terminated on 4th August 2004. The click beetle adults were identified to species and then their sex was determined by examination of the genitalia (Figure 6-1 and Figure 6-2).



Figure 6-1 Male click beetle genitalia, magnification x 10

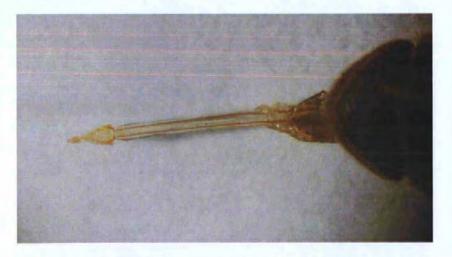


Figure 6-2 Female click beetle genitalia, magnification x 7.5

6.2.2 Click beetles for mark-release-recapture

Click beetles (*A. sputator*, *A. obscurus* and *A. lineatus*) were trapped using pheromone traps (Furlan *et al.* 2001a) from a number of fields within 20 miles of the experimental site in South Devon in May 2006. The beetles were stored at 16°C with vegetation and moist paper to maintain humidity until required (maximum 14 days).

Preliminary studies investigated the optimum method of marking beetles (tags (Piper 2003), lasers (Griffiths *et al.* 2001) and many paint types (Kishita *et al.* 2003)) and indicated that the application of acrylic paints were most suitable. The beetles were marked using a coded scheme of three coloured dots of acrylic paint (Sakura colour products, Japan) representing distance from trap, direction from trap and area of their release (Figure 6-3).

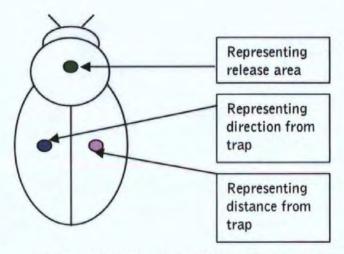


Figure 6-3 Schematic representation of click beetle marking method. Coloured dots on the pronotum represents one of three possible release areas; the dot on the left elytra represents the direction of release from the pheromone trap, north, east, south or west; and the dot on the right elytra represents one of four distances of the release point from the pheromone trap

6.2.3 Experimental procedure for mark, release, recapture

The experimental procedure for this mark, release, recapture work was piloted in

1995. Attractive pheromone traps for each of three click beetle species (A. sputator,

A. obscurus and A. lineatus) were placed in a topographically flat, organically-

managed, permanent pasture field with no wind obstacles in South Devon, UK.

Beetles of each species were released at distances of 4m, 8m, 16m and 32m (found

to be appropriate from pilot work), North, East, South or West of the pheromone

trap (a total of 16 release points). One beetle of each species was released at each of the four points at 4m from the trap; two of each species were released at each of the release points 8m from the trap, four at 16m and eight at 32m, (Figure 6-4, adapted from Turchin and Odenaal (1996)), or 60 click beetles at each of three reps, a total of 180 beetles. This was the maximum number of beetles that it was possible to capture, keep healthy, mark and release, weighted so that greater numbers were released closest to traps where recapture was more likely.

The release design shown in Figure 6-4 was repeated concurrently three times within the same field at separation distances of over 150m (found to be greater than achievable click beetle travelling distances in pilot work in 2005).

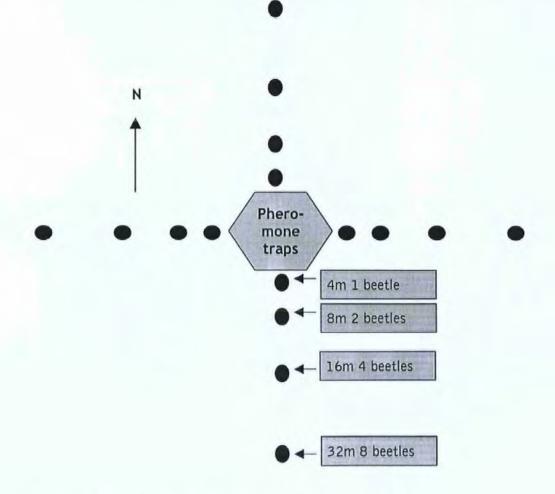


Figure 6-4 Release points of click beetles for mark-release-recapture experiment

The beetles were transported to the release site in small plastic vials on 23rd May 2006. The lids of the vials were removed and the vials placed horizontally on the ground. After twenty-four hours the tubes were collected and any beetles remaining in the tubes were recorded as non-releases. The traps were inspected at 2, 8, 21, 30, 37 and 45 days (arbitrarily chosen) after the release date. Marked beetles were removed for identification. The experiment was terminated when the field became unavailable for further use and trap catches were reduced to very low levels (45 days). Records of wind direction were identified, but were not used as direction had no effect on trap catches.

Sex ratios were determined and the effects of distance and direction were analysed using ANOVA. The sampling range was analysed by regression and effective sampling area was calculated using calculations based on Turchin and Odenaal's equation (1996) for estimating the recruitment area of pheromone traps.

6.3 Results

6.3.1 Sex ratios of click beetles in pheromone trap catches

In total, 943 male click beetles were captured, compared to 10 female click beetles (Table 6-2). The number of female click beetles captured was approximately 1% of the total. There was no difference between the number of females captured in traps containing large numbers of male click beetles and those containing few male beetles (for all three species, Pearson correlation < 0.098, P > 0.24). The number of females captured was independent of the variety of trap attractant used; therefore, it is probable that the females were captured randomly. The number of females is low and therefore should not affect the population estimates made from this data. Table 6-2 Numbers of click beetles of each sex captured in pheromone traps

	Males	Females	% female in captured population
A. sputator	246	7	2.77%
A. lineatus	201	1	0.50%
A. obscurus	496	2	0.40%

6.3.2 Recaptures

The proportion of non-released beetles (defined as individuals placed in a vial, at the release point, that had not exited within 24 hours) was very low: 4 out of 180 for *A. lineatus*, 2 for *A. obscurus* and 8 for *A. sputator* (Table 6-3).

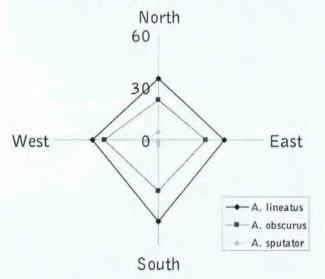
		Direction					
Species		East	North	South	West	Total	
А.							
lineatus	Released	45	43	43	45	176	
	Recaptured	17	15	20	17	69	
A.	Sec. 1. 1.						
obscurus	Released	44	44	45	45	178	
	Recaptured	12	10	13	14	49	
А.							
sputator	Released	41	42	45	44	172	
	Recaptured	0	2	1	3	6	
Total Sum	of Total						
released		130	129	133	134	526	
Total Sum	of Total						
Recaptured		29	27	34	34	124	

Table 6-3 Total releases and recaptures of three species of click beetles over the 6-week emergence period in 2006 from different release directions

6.3.3 Sampling direction

A 4-directional release plan was used to enable the visualisation of the effects of direction on click beetle recaptures (Figure 6-5). The numbers recaptured in every direction are equal, data were checked for homogeneity. Similar numbers of beetles were recaptured from all four directions of release, relative to the position of the trap (1-way ANOVA, direction, $F_{,3}$ =0.14, P=0.932). Consequently, there was no further analysis of directional data.

Figure 6-5 Total numbers of click beetles recaptured by cardinal direction from the trap over 45 days. Lines are an aid to visualisation in the style of Ostrand and Anderbrant (2003)

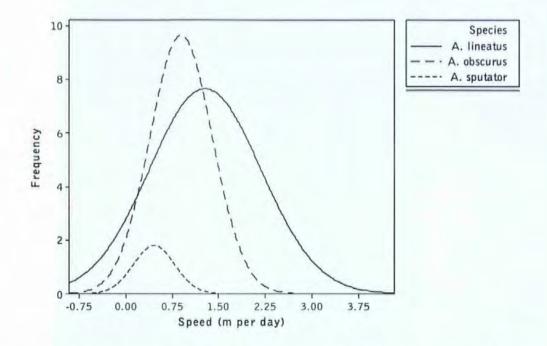


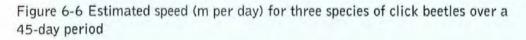
6.3.4 Speed of travel

The distance travelled by, or net displacement of the click beetles was calculated as a straight line between the release point and the trap in which the beetle was caught, and the speed was calculated. This is very unlikely to represent the true pattern of movement (and therefore speed) of click beetles as they are unlikely to move in a straight line, but to be diverted. *A. lineatus* were found to travel the fastest at an

average speed of 1.28m day⁻¹, *A. obscurus* travelled at an average speed of 0.89m day⁻¹, and *A. sputator* travelled slowest at 0.46m day⁻¹ (Figure 6-6).

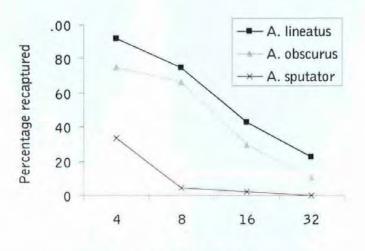
The fastest speed that any one beetle averaged on its return to the trap was also calculated. The maximum speed achieved by one *A. lineatus* was 4.09m day⁻¹, *A. obscurus* was 2.13m day⁻¹ and *A. sputator* was 1.07m day⁻¹.





6.3.5 Sampling range

A two way ANOVA (without replication) showed that the rate of recapture of click beetles (beetles recaptured/beetles released) differed with the distance of the release point from the pheromone trap (Figure 6-7 and Table 6-4, F_4 =11.70 P= 0.006) and differed significantly between the three species (Figure 6-7 and Table 6-4, F_2 =15.61 P=0.004). The Tukey procedure revealed that all pairwise differences among means for distance were significant, p < 0.0001, except for 4m and 8m (p=0.0725) and 16m and 32m (p=0.1718). The Tukey procedure also revealed that the differences between *A. sputator* and the other to species were significant (p>0.0001), but that the pairwise difference between the means of *A. obscurus* and *A. lineatus* were not significant (p=0.9453). Figure 6-7 shows *A. sputator* recaptures decreased over a much shorter distance than either of the other two species, which showed a more mutually consistent decline.



Distance from trap (m)

Figure 6-7 Total percentage of click beetles recaptured from all distances from pheromone trap over 45 days in South Devon in 2006

		Distance				
Species		4m	8m	16m	32m	Total
A. lineatus	Released	12	24	45	95	176
	Recaptured	11	18	19	21	69
		(91.7%)	(75.0%)	(42.2%)	(22.1%)	(39.2%)
A. obscurus	Released	12	24	47	95	178
	Recaptured	9	16	14	10	49
		(75.0%)	(66.7%)	(29.8%)	(10.5%)	(27.5%)
A. sputator	Released	12	23	46	91	172
	Recaptured	4	1	1	0	6
		(33.3%)	(4.3%)	(2.2%)	(0%)	(3.5%)

Table 6-4 Total releases and recaptures of three species of click beetles over the 6-week emergence period in 2006 from different release distances

The distance from which the pheromone trap is unable to attract any click beetles was ascertained by building regression models of overall recapture rates (P(r)) against release distance from the trap (r), from which P(r) = 0 could be calculated (Turchin and Odendaal 1996). P(r) was calculated for cumulated catches over three different time periods: the first 15 days, the first 30 days and 45 days. Using P(r) = 0 and the equation on page 140, sampling range (r_s) and the effective sampling area (α) over a set time period can be calculated.

6.3.6 Sampling range

As the three species have been shown to behave differently to pheromone traps (Section 6.3.2) the species were initially analysed separately to identify the best models using combinations of untransformed and log transformed data (Turchin and Odendaal 1996) in order to find the best fit to explain click beetle movement. However, the models including Log dist – log P(r) behaved either best or a close second in every circumstance, this model was selected for all occasions to enable comparison between species and time (J Perry, pers. comm.).

	Number	Distance	Log distance	Log distance	Distance
	of days	P(r)	P(r)	Log P(r)	Log P(r)
		y=-0.028x	y=-0.906x	y=-0.274x	y=-0.009x
A. lineatus		+0.91	+1.44	+0.45	+0.29
	15	$R^2 = 0.8203$	$R^2 = 0.8279$	$R^2 = 0.8422$	$R^2 = 0.8737$
		y=-0.023x	y=-0.749x	y=-0.214x	y=-0.007x
		+0.90	+1.34	+0.41	+0.28
1	30	$R^2 = 0.7474$	$R^2 = 0.7648$	$R^2 = 0.7744$	$R^2 = 0.7848$
X		y=-0.024x	y=-0.794x	y=-0.223x	y=-0.007x
		+0.94	+1.42	+0.43	+0.30
	45	$R^2 = 0.8303$	$R^2 = 0.8928$	$R^2 = 0.8848$	$R^2 = 0.8512$
		y=-0.021x	y=-0.682x	y=-0.210x	y=-0.007x
S		+0.66	+1.05	+0.33	+0.21
	15	$R^2 = 0.384$	$R^2 = 0.3854$	$R^2 = 0.3792$	$R^2 = 0.3934$
un		y=-0.024x	y=-0.782x	y=-0.240x	y=-0.007x
psq		+0.77	+1.24	+0.39	+0.25
A. obscurus	30	$R^2 = 0.5203$	$R^2 = 0.5669$	$R^2 = 0.5932$	$R^2 = 0.5544$
A		y=-0.024x	y=-0.764x	y=-0.232x	y=-0.007x
		+0.81	+1.26	+0.40	+0.26
	45	$R^2 = 0.6133$	$R^2 = 0.6378$	$R^2 = 0.6764$	$R^2 = 0.6659$
		y=-0.005x	y=-0.173x	y=-0.067x	y=-0.002x
		+0.13	+0.24	+0.09	+0.05
1	15	$R^2 = 0.2746$	$R^2 = 0.387$	$R^2 = 0.3839$	$R^2 = 0.2756$
A. sputator		y=-0.009x	y=-0.339x	y=-0.126x	y=-0.003x
		+0.23	+0.46	+0.17	+0.09
1.5	30	$R^2 = 0.365$	$R^2 = 0.5643$	$R^2 = 0.589$	$R^2 = 0.3853$
A		y=-0.009x	y=-0.339x	y=-0.126x	y=-0.003x
		+0.23	+0.46	+0.17	+0.09
	45	$R^2 = 0.365$	$R^2 = 0.5643$	$R^2 = 0.589$	$R^2 = 0.3853$

Table 6-5 R^2 values for models defining pheromone trap catches against distance. Shading shows the `best-fit' model.

Species	Number of days	Maximum sampling range	Effective sampling area (α) (m²)	
		P(r) = 0 (m)		
A. lineatus	15	43.29	2588	
	30	80.35	6908	
	45	82.00	6768	
A. obscurus	15	37.80	2580	
	30	42.59	2795	
	45	51.31	3636	
A. sputator	15	24.57	1698	
	30	22.40	1335	
	45	22.40	1335	

Table 6-6 Outputs from models built to predict sampling range and effective sampling area of pheromone traps for three click species

Large differences were again found between the species in their effective sampling area and sampling range, although *A. lineatus* and *A. obscurus* are more similar to each other than to *A. sputator*. The effective sampling areas of *A. lineatus* and *A. obscurus* increase with time. However the effective sampling area of *A. sputator* does not increase from 30 to 45 days. The maximum sampling range of *A. obscurus* continued to increase throughout the experiment, however the other two do not increase throughout. *A. sputator* has a much smaller effective sampling area over all timescales than *A. obscurus* which has a smaller effective sampling area than *A. lineatus*. Species are ranked in the same order as their speed of travel but not the same proportions.

6.4 Discussion

6.4.1 Determining the sex ratios of click beetles in pheromone trap catches

Aim C, p141 was to determine the sex ratios of pheromone trap catches, section 6.3.1 provides the numbers of females and males recaptured. The proportion of females captured was negligible, indicating that the pheromone traps are effective at attracting males, and that there is no subsequent attraction of females to the captured males. However, there were indications that female *A. sputator* are more active than either of the other two species as they are more likely to be caught randomly than either *A. obscurus* or *A. lineatus*.

6.4.2 Speed of click beetle travel

Aim D, p141 was to elucidate the speed of travel of click beetles. Section 6.3.4 provides the average and maximum speed of the three click beetle species recorded in this experiment. *A. lineatus* moved quickest and *A. sputator* moved slowest of the three click beetle species, this is the same order as their body lengths, *A lineatus* is the longest and *A. sputator* is the smallest. Their differing dispersal speeds may also have an effect on their distributions and movement throughout an agricultural landscape. Using mark-release-recapture protocols, the speed measurements are based on net displacement and therefore will underestimate both overall movement by individuals and their speed. Consequently, it is not possible to draw any conclusions regarding the movement footprint or area covered by dispersing beetles and the speed values calculated must be taken as minimum velocities. In an interesting contrast *A. sputator* moved the slowest, but was captured most often when the YATLOR traps

were used as pitfall traps. This may be explained by *A. sputator* being less responsive to the pheromone so that they move towards it less directly but covering a lot of ground with random movements.

A confounding factor in this style of mark-release-recapture experiment is that the naturally occurring population of click beetles was untouched before the experiment. This meant that naturally occurring females may have acted as a competing source of pheromones to the males and so affect their progress towards the pheromone trap. The existing populations of females could have been estimated using pitfall traps in conjunction with pheromone traps, but this may have led to the inadvertent capture of marked beetles with a associated knock-on effect on the pheromone trap catch results. The mating habits of click beetles are unknown at present and so we cannot estimate the effect this may have. However, this is likely to reflect 'reality' in field use of pheromone traps.

In this study, the mean speed for *A. sputator* was 0.45m day⁻¹. The mean speed travelled by the *A. sputator* in Gouch and Evans (1942) was 1.08m day⁻¹ which is very similar to the maximum speed of *A. sputator* in this experiment (1.07m day⁻¹). Gouch and Evans used clumps of grass as traps, these traps provided shelter and thus attracted males and females, as opposed to pheromone traps which attract males only. This is very different from the speed of the Japanese click beetle; *Melanotus okinawensis* (Kishita *et al.* 2003) which travelled a mean distance of 77.4m and 221.1m in two days (two experiments). Knowledge regarding the movements of click beetles will also enable the study of click beetle dispersal and metapopulations,

the processes of which are significant in the repopulation of fields after population crashes (Koenig 1999).

Frequency histogram of speeds show smooth curves for all species, however, four individual *A. lineatus* (out of 69) travelled much greater distances than other *A. lineatus*, this suggests that *A. lineatus* click beetles may travel by two methods, walking short distances most of the time and occasionally flying greater distances. It is possible that the travel method may be strongly affected by weather conditions, with high temperatures triggering flight. Larger numbers of beetles would need to be studied in order to repeatedly observe this rare event.

Transect spatial work in Chapter 2 suggested *A. lineatus* had a strong within-field effect. If *A. lineatus* moves faster or further, it has the ability to travel to the best environment, and populations may congregate there.

6.4.3 Direction

There were no directional differences in click beetle recapture, as was also found for *M. okinawensis* (Kishita *et al.* 2003). The typical way of looking at insect behaviour towards pheromones is to visualise a plume through which the insect moves to orientate itself towards the trap (Perry and Wall 1985; Perry *et al.* 1988), mostly because much previous pheromone work has been on flying moths with pheromone release over a metre above the ground. Click beetles are believed to be ground based and rarely, if ever, fly (Furlan *et al.* 2001a). This experiment took place in long grass before a silage cut. The pheromone was placed 25cm from the surface of the ground, so it is possible a plume was present above the grass, but as no directional

response to the pheromone was seen it is possible that the pheromone is more evenly dissipated at ground level between the stems of grass.

6.4.4 Sampling range

Aim A, p141, was to quantify the spatial parameters of pheromone traps. The sampling ranges for three time periods are shown in Table 6-6, for each of the three click beetle species.

The design of this experiment allowed for a maximum range of 40m. This was estimated in a pilot study in 2005, but may be less than the distance a click beetle could travel between emergence and death. Although their life expectancy is unknown, even the furthest distance travelled by a beetle in this experiment will be less than any lifetime dispersal distance as all beetles had already been captured in a pheromone trap before being marked. The maximum trapping distance may be greater than the radius of α (effective sampling area) as α is unlikely to be a circle (Wall and Perry 1985) as conditions within the sampling area will not be homogenous.

Little is known about the differences in biology of the three species and this may confound any conclusions based on direct comparison between them. For example, there may be different mortalities between species, different native female populations with competing pheromone production or the pheromones of different species may be transmitted differently. This makes it difficult to draw conclusions about the causes of the variation in effective sampling area. However, these concerns do not affect the use of the effective sampling area to estimate density of pheromone traps that would be required for removal trapping. It is, however, necessary to allow for the influence of crop type on pheromone dispersal (Wall and Perry 1981).

For removal trapping to be effective there has to be a direct link between the insect captured in the trap and the pest stage of the insect. In an insect with an annual lifecycle, a reduction in pest population should be seen the following year (assuming minimal immigration). However, wireworms and click beetles have a 4-5 year lifecycle; so removal trapping of one year's beetles will not reduce the emergence numbers the following year in any way. It would take at least three or four years before any reduction in pest numbers was possible. With removal trapping it is also essential that the adult males are captured before they mate with females. This is the only way to ensure the population is reduced. If it is assumed that all males captured in this experiment have not mated the number of traps required to remove the male population can be calculated. These calculations are based on the effective sampling area over 15 days to minimise the chances of males mating. An estimate of the costs of removal trapping can be made using current prices of pheromones and traps at €9 per trap and $\in 2.50$ per bait, assuming all traps are reusable each year. A. lineatus and *A. obscurus* would require 4 traps per hectare at a cost over four years of €90 per hectare, and A. sputator would require 6 traps per hectare at a cost over four years of $\in 114$ ha⁻¹. The total cost of $\in 294$ ha⁻¹ is expensive, but conceivably economically viable for high value crops in fields at high risk.

In summarising aim B, p141, the three species examined do show species specific speeds of travel and ranges of attraction. However A. obscurus and A. lineatus show more similarities to each other than either does to A. sputator.

Discussion

6.5 Pest management

The literature promoting organic management of crops focuses on the requirement for a 'natural balance' of the environment which is promoted as a method for minimizing pest populations (Frost 2003; Hole *et al.* 2005; Soil Association 2003). The process of stabilizing pest populations is postulated to take many years. Riverford Farm, the location of much of this fieldwork, was one of the earliest converters to organic production and they are still suffering substantial losses to subterranean larval pests, a viable organic pest control method is required.

The old model of investigating pest control was the hunt for the 'magic bullet', a solution to control a single pest in every environment and geographic region. Lewis (1997) recommended that investigating pest control should be replaced by a new model: the search for pest control should be in a restricted environment and geographic region but effective on a range of pests. This newer model defines the framework for this project. Overall, Chapter Five has shown no definitive recommendations for control have been formulated, however, knowledge of factors determining the distribution of this range of pests has been increased and crop type and cultivations have been identified as potential control methods. It is possible that definitive control methods were not identified in this project as the restricted geographical area used was a heterogeneous environment and a range of habitats were present within a small geographical area. This is not necessarily a characteristic of all organic farms.

6.6 Larval pest sampling

Chapter Four showed that the populations of male crane flies and the sum of male and female crane fly catches were both correlated with leatherjackets, suggesting that adult males are distributed around suitable larval habitats, and has found that A. obscurus click beetles are correlated with wireworm populations. This was also found by Blackshaw and Vernon (2006a). These findings have further validated the use of water traps for monitoring leatherjacket populations (Blackshaw 1987b) and suggest the use of *A. obscurus* pheromone traps for identifying areas of high wireworm populations. If the correlation between wireworms and A. obscurus click beetles proves to be true under a range of conditions and geographic areas, then using pheromone traps for estimating wireworm populations may be a practical, quick, simple and cost effective alternative to the large effort of sampling soil for wireworms. Pheromone traps and water traps give estimates of relative populations, whereas soil sampling gives an estimate of absolute populations. If pheromone or water traps were to be considered for replacing soil sampling they would need calibration to ensure the match of larval and adult populations under a range of conditions. The other pest species studied in this project were not correlated with larval populations. There are a number of potential reasons for this. For *Agriotes*, it is believed that the wireworm complex does not just represent one species so it may be considered surprising that the complex of all wireworm species should correlate with A. obscurus click beetles alone and not with either of the other two species. Another potential cause of the non-correlation of larval and adult populations of pests is the delay between the larval sampling time of year and the emergence time for

adults, it is possible that there was movement of the populations within these times. Alternatively, there may have been mortality of larvae in non-suitable habitats or exposure to mortality inducing factors between the sampling of larvae and sampling of adults or that the sampling techniques used did not sample for *A. lineatus* or *A. sputator* wireworms.

6.7 Larval pest distribution

An alternative strategy to sampling for pests is to predict the populations of pests so that areas of high populations can be identified and treated. This might be chemically, but may also be through a cultural control method, e.g. avoidance. A major finding in this study is that the distributions of the range of subterranean pests studied in this project share few commonalities with regard to distribution. Throughout this project (chapters Two and Four) only one *T. oleracea* was found (in 2004 or 2005), therefore density of *T. oleracea* in this area of the UK appears to be effectively zero with a zero distribution. All species except *T. oleracea* were found throughout the study area, indicating that these are widespread throughout all these agricultural land types and land use situations. The prevalence of all three *Agriotes* species throughout agricultural land is also observed by Parker in the UK (pers. comm.). Leatherjackets and *T. paludosa* crane flies were also found to be present throughout the study area and their distributions were found to be affected by different factors from each other as do the three congeneric species of *Agriotes* and their larval complex.

The spatial distribution of all three click beetle species, crane flies and the larvae of these species has been further elucidated in this project, and significant spatial structure was identified at a number of scales. The only modern European work on the distribution of *Agriotes* was pan-European and showed great differences in the abundances of the species between sampling locations, of which there were only one or two per country (Furlan *et al.* 2001b). Furlan *et al* had only one location in the UK, which had pheromone traps for each of the three species, all traps captured click beetles but no finer detail of distribution was obtained. This project has sampled populations at a much finer scale and has shown that differences in abundances can be present across much smaller distances. This was also shown in Canada by Blackshaw (Blackshaw *et al.* 2007) where population densities changed within a few hundred metres. Two scales were considered in the study, in Chapter Three to identify high population patches within fields and in Chapter Two to identify population variation across a landscape.

The populations at landscape scale showed clustering and when examined at a smaller scale the spatial structure showed a pattern of both patches and gaps. Moran's I showed that all of the organisms studied (with the single exception of *A*. *lineatus* in 2005) showed spatial structure at the landscape scale of the transect. *A. lineatus* populations were found to be defined at a field scale, catches within fields were more similar than catches the same distance apart but in separate fields. Field scale is the traditional scale for observing agricultural pests as this is the scale at which chemical interventions are generally applied, but is not necessarily the optimum scale in population terms.

Within individual fields spatial structure of populations was not generally identifiable except at very high pest populations. Where high populations are present, selective pesticide use on patches is a potential control method (providing non-organic control). The population threshold for treatment of leatherjackets in many row crops is 0.25 million ha⁻¹ (www.dowagro.com), and wireworms can cause damage at populations of less than 100,000 ha⁻¹ (Parker and Howard 2001). These are both below the populations at which field scale spatial patterning is visible in this study. In addition, the very high effort required for sampling subterranean pests would prevent this ever being a practical solution for reducing pesticide usage. However, in organisms that are quicker and easier to sample, the use of SADIE methodology would enable the identification of patches of varying sizes and shapes. An interesting experiment that was outside the limits of this project would be to test for a match between SADIE maps of patches (regions of high populations) and gaps (regions of low populations) and aerial photographs of pest damage to crops.

6.8 Influences on larval pest populations

Assessing pest distribution at the landscape (farm) scale was useful as it enabled visualisation of the distribution of organisms and how that distribution related to the geography and landscape mosaic of the environment. This is of practical value to the grower, as their interest is often at a 'whole farm' scale. Organic vegetable rotations work on a farm scale, with crops rotating through many or all fields on the farm, therefore a grower must have confidence in knowledge of pest populations over the whole farm (although damage may only occur in a vulnerable crop in one year). This is particularly pertinent for wireworms as their many years of subterranean life can

cover a large period of a vegetable - ley rotation. This study also revealed no evidence that larval populations are associated with headlands and hedges at the edges of fields.

Agricultural land, even in the small area of this study, is not homogenous. Chapter Two showed that whilst great differences in populations were present within small distances, populations were associated with certain conditions. For example, A. obscurus was less often found closer to watercourses. Environmental, chemical and physical variables were all shown to influence subterranean populations of all these pests, but different environmental, physical and chemical variables affected different species even within the same genus and variables showed different effects in different life stages of the same species. The transect work has shown that all adult organisms studied were probably associated with grass, as permanent pasture or ley and the multiple regression confirmed this, specifically, populations of wireworms increased with the numbers of years in grass. The Good Agricultural and Environmental Condition of Cross Compliance for the Single Payment Scheme contains a clause to prevent loss of permanent pasture (defined as a field that has been in grass for five years or more, even re-seeded leys), the maximum amount of permanent pasture that can be ploughed in the UK is 5%. From 1st January 2005 any permanent pasture requires an environmental impact assessment before it can be ploughed for cropping (Defra 2004a). It is probable that only fields with a high proportion of ryegrass will be granted permission for cultivation. There is plenty of evidence that higher wireworm populations are found in permanent pasture and we would expect to see a reduction in the damage caused by wireworms in Europe. Though this is currently uninvestigated it may be advantageous to observe which permanent pasture fields are

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obtaining derogations as a result of Environmental Impact Assessments and how this relates to wireworm populations and rates of crop damage.

The multiple regression specifically identified that populations of *Agriotes* (wireworms and some click beetles) were primarily characterised by number of years in grass, but also by moisture, organic matter and soil shear strength, whereas soil shear, aspect, cultivations and manure most strongly influenced Tipulid distributions. The relationship between high populations and these variables may be due to either site selection or immigration by adults or alternatively be as a result of that variable providing a suitable habitat for the survival of larvae. It is very difficult to differentiate between either of these causes. It is possible that as the number of years in grass increases, the complexity of the root structure of the grass field increases and this food source may then become more beneficial to larvae. This would support the positive relationship between wireworms and organic matter, but is contrary to the perceived wisdom that the increased biodiversity of organic systems reduces pest populations. Multiple regression analyses identified different variables as important in defining distribution for each species, and most notably the variables defining the wireworm complex did not mirror the adults of any one species or appear to show similarities with all three together. The distribution of the wireworm complex appeared to be affected by variables independent of those affecting the adult species. This was also true of those variables defining male or female adult Tipulids; the same variables did not define the leatherjacket population. This indicates that the environment required for larvae is different from that required by adults or that the mixture of species in the wireworm complex is masking species specific environmental requirements. There is a need for the ability to identify the species of

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Agriotes wireworms (Ellis *et al.* 2009), this will facilitate the identification of the variables which define the distribution of each individual species of wireworm, and this may facilitate the prediction of areas containing high populations of wireworms.

6.9 Pest larvae life cycles

Wireworms have a lifecycle with the larval phase lasting up to 5-6 years (Miles 1942), T. paludosa leatherjackets have an annual lifecycle (Laughlin 1967). The differences in the length of larval life stage were evident in the transect study, where Agriotes species were found to show a far greater level of year-on-year population stability than was found in Tipulid populations. Both species are more vulnerable to mortality at earlier larval instars, for leatherjackets this is for only a few months over winter but for wireworms this may be a number of years (Gough and Evans 1942; Meats 1974; Nettles 1940). The protracted larval stage of wireworms may mean that populations can be expected to be affected by cultural effects over a number of years and the leatherjacket, with its shorter larval stage may be expected to be more affected by cultural action over the previous 12 months than over the previous five years: but this study found the reverse. Tipulid populations were more influenced by five-year variables and the *Agriotes* complex by single year variables. Wireworms were affected by single year variables whilst leatherjackets were found to be associated with the average number of bed formation occurrences (e.g. for potatoes) over five years, the average quantity of manure over five years and the amount of weeding (mechanical and hand) in the previous year, increased cultivation and greater manure application increased the number of leatherjackets.

Previous work has shown increased cultivations decrease populations of leatherjackets, by exposure to predators or desiccation (Blackshaw 1988), this does not appear to be happening here. This study and McCracken *et al* (1995) found that the long term addition of manure increases leatherjacket populations, but the incidental addition of extra nutrients has been shown to increase the overall crop output, this relationship requires further elucidation. The added benefit of the addition of nutrients would fertilise plants so they are more able to compensate for pest attacks, it is not known if this increased output would compensate for the crop losses due to pests.

Overall, it appears that a number of cultural, environmental and chemical variables are driving all populations and with such a range of habitats it is possible that there is a variety of variables acting as population limiting factors, not one or all having a population limiting influence in every environment. Interestingly, although many of the variables highlighted in this analysis are aligned with those identified by Parker and Seeney (1997) and McCracken *et al* (1995) in their multivariate analyses, many are different. This strongly highlights the difficulties with reliance on single multivariate studies and indicates that pest ecologies are more complex than are definable from this data, especially with the added confusion of the inability to identify wireworms to species level.

The multivariate analyses of the survey data were also used to identify cultural techniques which are potentially mortality inducing, and which may be used to control populations, including manure addition, rolling and cultivations. Greenhouse experiments and field trials were carried out to evaluate these. In the manure and cropping in greenhouse and cultivation experiments there appeared to be large differences between the treatments, however, they were inconsistent.

As at present, with no method to differentiate wireworm species, experiments of this style must potentially use a number of species of *Agriotes* wireworms, which will undoubtedly confound interpretation of the results, there may be no point progressing this until this identification of is possible (Ellis *et al.* 2009). An alternative is to breed wireworms so that their species is confirmed, however, again, the long larval stage of wireworms causes difficulties and their supply is very unpredictable and often they are impossible to obtain. Cropping and cultivation experiments investigating the effect of cultivations on wireworm populations would need to be carried out over a number of years, due to the length of the larval life length of wireworm and that long term variables were identified as having more effect on leatherjacket populations than single year variable were.

6.10 Larval pest movement

Field plot experiments are also based on the implicit assumption that larvae are relatively non-mobile. If the larvae were found to be more mobile, this may provide false results by the movement of larvae out of plots, it is unlikely that this movement could occur on a field scale in a short time frame. Knowledge of the mobility of organisms is necessary, not only for plot experiments, but also concerning repopulation of treated areas from pest reservoirs, time delays between sampling and treating, and gene flow between populations. Larval movement was not studied in this project but knowledge of larval movement may aid interpretation of the 'within field' studies in this and other patch identifying studies. Movement of larvae is

important when considering sampling and control options, reducing the population in one field is redundant if the field is immediately repopulated from a neighbouring pest population. Although larval and *T. paludosa* movement is not studied in this project, the movement of the adults of three species of *Agriotes* was.

The mark, release, recapture work in this study has shown that adult *Agriotes* species have differential mobility, *A. lineatus* were found to travel the fastest at an average speed of 1.28m day⁻¹, *A. obscurus* travelled at an average speed of 0.89m day⁻¹, and *A. sputator* travelled slowest at 0.46m day⁻¹. Studies on the movement of click beetles towards pheromone traps cannot be directly related to movement of click beetles when repopulating depopulated areas. Movement measured in this experiment is the movement of males to locate females for mating; this may be different from movement for the purpose of site selection where adults are moving to a suitable habitat. Studies of gene flow through the environment would enable the understanding of movement of populations through the landscape; this may allow the relevant environmental factors to be identified with more clarity.

Identification of regions of high populations of wireworms by capturing adult male *Agriotes* in pheromone traps has been recommended previously, but has treated all species as equal. Now different travel and speeds and distances may allow for more accurate estimates of larval populations e.g. weighting species catches to allow for lower catches of short distance travelling beetles. The current model makes an assumption that each adult species contributes equally to each larval species and each larval species contributes equally to the damage to crops. However to sort out

these two assumptions makes the accurate differentiation of larval species is necessary.

All studies in this project are limited by the short time frame. There is a requirement especially with *Agriotes* having a long generational time, for a long term study of larval distribution, especially as the pertinent variables are also likely to show temporal variation, such as the weather affecting soil moisture and larval survival, or the timing of cultivations and their effect on larval mortality. Larvae show variable susceptibility to adverse conditions at various growth stages, generally, early instar larvae are more likely to be subject to mortality than late instar larvae and so mortality inducing factors present in previous years might have been expected to influence the populations when sampled years later, the long term stability of patches and gaps of aggregation are also unknown. Consequently, to identify fully the effects of the variables studied in this work it is necessary to compare the relationships between the variables and the populations of different age larvae and at different times of year.

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Appendix 1

	2005		2006	
Field	Rs	P	Rs	Р
HPF	0.16	0.0892	-0.031	0.793
HPG	-0.083	0.491	No WW	No ŴW
НРА (Тор)	0.143	0.284	-0.019	0.885
HPC (soil cores)	-0.013	0.915	0.040	0.737
HPA (bottom)	0.194	0.103	0.002	0,985
Broad Park	-0.065	0.596	-0.033	0.804
Bakers Park	0.192	0.139	-0.074	0.610
Hills Small	No WW	No WW	-0.166	0.158
HPC (traps)	-0,047	0.661	0.042	0.692

Table 7-1 Correlation coefficients (R_s) and significance levels (P) for correlations between the number of wireworms and soil moisture in fields across Devon. No WW means that no wireworms were sampled in that field.

Table 7-2 Correlation coefficients (R_s) and significance levels (P) for correlations between the number of leatherjackets and soil moisture in fields across Devon. No LJ means that no leatherjackets were sampled in that field.

	2005	_	2006		
Field	R _s	Ŗ	Rs	Ρ	
HPF	0.141	0.239	0.150	0.208	
HPG	-0.063	0.603	-0.192	0.108	
НРА (Тор)	0.247	0.062	0.042	0.754	
HPC (soil cores)	-0.063	0.594	-0.019	0.870	
HPA (bottom)	-0.234	0.048	0.127	0.289	
Broad Park	0.053	0.668	No LJ	No LJ	
Bakers Park	0.208	0.108	-0.173	0.229	
Hills Small	iills Small 0.192		-0.055	0.643	
Hills Big	Big 0.279 0.01		Not sampled	Not sampled	
HIPC (traps)	No LJ:	No ĽJ	No LJ	No LJ	

	Wireworms		Leatherjackets		
Field	R _s	Р	Rs	Р	
'HPF	-0.020	0.867	0.012	0.922	
HPG	No WW	No WW	0.079	0.514	
HPA (Top)	0.030	0.822	0.014	0.914	
HPC (soil: cores)	PC (soil cores) -0.067		-0.110	0.349	
HPA (bottom)	A (bottom) 0.065		0.088	0.462	
Broad Park	pad Park -0.069		No LJ	No LJ	
Bakers Park	-0.190	0.186	-0.104	0.472	
Hills Small	-0.072	0.541	0.162	0.168	
Hills Big	Not sampled	Not sampled	Not sampled	Not sampled	
HPC (traps)	0.102	0.341	No LJ	No LJ	

Table 7-3 Correlation coefficients (R_s) and significance levels (P) for correlations between the number of wireworms and leatherjackets and soil shear at 5cm depth in fields across Devon. No LJ means that no leatherjackets were sampled in that field.

Table 7-4 Correlation coefficients (R_s) and significance levels (P) for correlations between the number of wireworms and leatherjackets and soil shear at 10cm depth in fields across Devon. No LJ means that no leatherjackets were sampled in that field.

	Wireworms		Leatherjackets		
Field	Rs	Р	Rs	Р	
HPF	-0.103	0.390	-0.048	0.688	
HPG	No WW	No WW	0:090	0.453	
HPA (Top)	-0.127	0.342 0.204		0.125	
HPC (soil cores) 0.040		0.733	0.069	0.558	
HPA (bottom)	IPA (bottom) -0.046		0.153	0.201	
road Park -0.187		0.156 No LJ		No LJ	
Bakers Park	0.009	0.951	-0.030	0.837	
Hills Small	0.053	0.656	0.184	0.117	
Hills Big	Not sampled	Not sampled	Not sampled	Not sampled	
HPC (traps)	0.103	0.333	No LJ	No LJ	

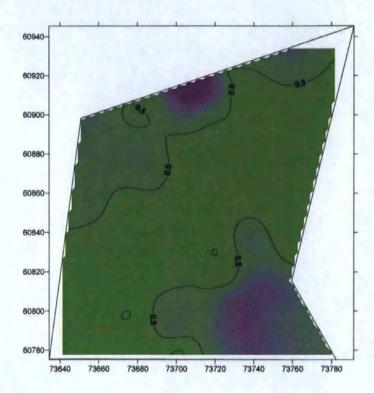
Appendix 2

	Number	Wireworms & Moisture		=	ackets &	Wireworms & Leatherjackets		
Field	of cores	X	P	Х	Р	X	Ρ	
							(a)	
HPF	72	-0.1180	0.5787	0.1411	0:1289	0.6773	<0.0001	
							(s)	
HPG	71	0.0081	0.4739	0.1656	0.0998	-0.4686	0.9998	
HPA (Top)	58	0.1435	0.1386	0.2236	0.0698	0.2705	0.0298	
HPC (soil			(1)					
cores)	-74	-0.2666	0.9889	-0.1111	0.8148	0.0163	0.4394	
HPA			(b)		(u)		(v)	
(Bottom)	72	0.2763	0.0115	-0.2987	0.9893	-0.3795	0.9993	
Broad Park	68	0.1072	0.2200	-0.0376	0.6172	-0.1864	0.9241	
Bakers Park	61	0.1243	0.2358	-0.0208	0.5603	-0.0058	0.5106	
Hills Small	76	-	-	-0.0918	0.7710	-	-	
					(c)			
Hills Big	74	-	-	0.5887	<0.0001	-	-	
			(w)					
HPC (traps)	90	-0.3127	0.9975	-	-		-	

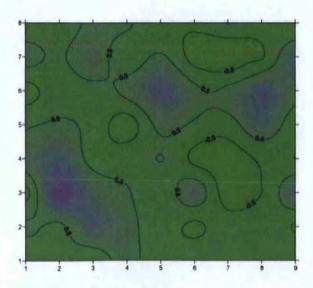
Table 7-5 SADIE associations (X = Chi) for the spatial distribution of wireworms, leatherjackets and soil moisture in 2005.

		2006 LJ.& WW	2006 LJ & moisture	2006 LJ & 5cm Shear	2006 LJ & 10cm Shear	2006 WW & moisture	2006 WW & 5cm Shear	2006 WW & 10cm Shear	05 & 06 니	05:& 06 WW
	х	-0.159	0.008	0.393	0.460	0.152	0.004	-0.084	0.218	0.040
HPA				(d)	(e)					
(Bot)	Ρ	0:906	0:466	<0.001	<0.001	0.103	0.491	0.754	0.049	0.372
	х		0.234	-0.216	0.036				0.442	
		ZERO	(f)			ZERO	ZERO	ZERO	(g)	ZERO
HPG	Ρ	ww	0.015	0.965	0.39	ww	ww	ww	< 0.001	ww
	х	ZERO	ZERO	ZERO	ZERO	0.044	0.109	0.058	ZERO	-0.086
HPC	•	LJ	LJ	LJ	LJ				LJ	0.335
(Traps)	Ρ					0.362	0.201	0.299		0.775
	Х	0.706	0.388	-0.163	-0.339	0.231	-0.239	-0.281	0.596	0.862
		(h)	(i)		(x)		(y)	(ż)	(j)	(k)
HPF	Ρ	0.001	0.006	0:908	0.994	0.072	0.980	0.976	<0.001	<0.001
	х	-0.098	0.036	0.182	0.353	0.020	0.088	-0.196	0,391	0:195
НРА					()				(m)	
(Top)	Ρ	0.762	0.404	0:089	0.003	0.441	0.262	0.929	0.004	0:081
	х	ZERO	ZERO	ZERO	ZERO	0.029	0.165	0.091	ZERO	0:389
Broad		LJ	LJ	LJ	LJ				LJ	(n)
Park	Ρ					0.037	0.105	0.261		0:003
	х	-0.071	-0.078	-0.072	0.067	-0.021	-0.004	-0:059	0.234	0.284
HPC										(o)
(Cores)	Ρ	0.729	0.731	0.730	0.339	0.566	0.507	0.685	0.071	0.011
	х	-0.337	0.26	0:648	0.408	-0.230	-0.120	-0:064	-0.789	
Hills		(aa)	(p)	(q)	(r)				(ab)	ZERO
Small	Ρ	0.997	0.015	< 0.001	< 0.001	0.952	0.957	0.689	0.999	ww
	х	-0.138	-0.176	-0.357	-0.279	-0.140	-0.222	-0:055	0.24	-0:065
Bakers				(ac)						
Park	Ρ	0.837	0.867	0.996	0.967	0.821	0.940	0.640	0.054	0:647

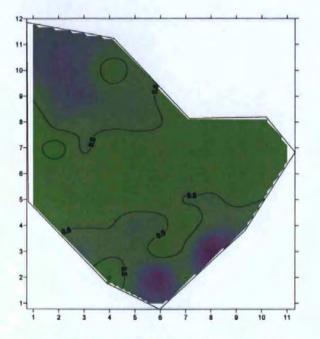
Table 7-6 SAD1E associations (X = Chi) for the spatial distribution of wireworms (WW), leatherjackets(LJ), soil shear strength (shear) and soil moisture in 2006 and 2005.



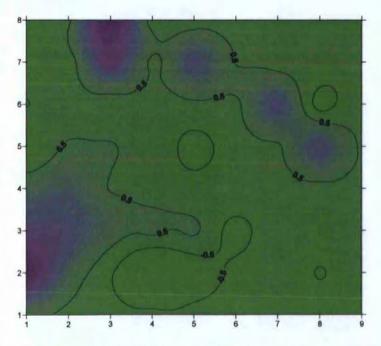
a. (from Table 7-5/Table 7-6) SADIE associations for HPF, 2005 Leatherjackets & wireworms



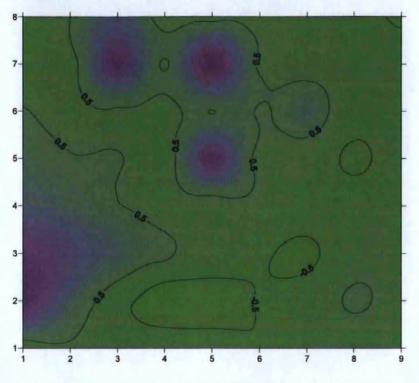
b: (from Table 7-5/Table 7-6) SADIE associations for HPA (bot) 2005 wireworms & moisture



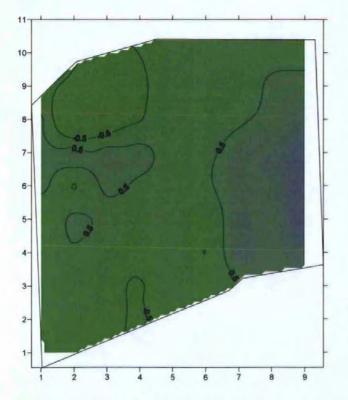
c: (from Table 7-5/Table 7-6) SADIE associations for 2005, Hills Big Leatherjackets & Moisture



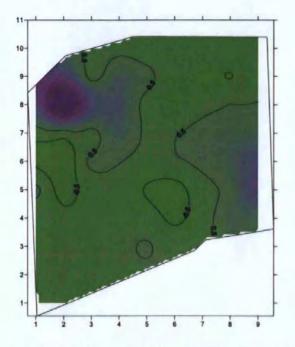
d: (from Table 7-5/Table 7-6) SADIE associations for HPA (bot) 2006 leatherjackets & 5cm shear



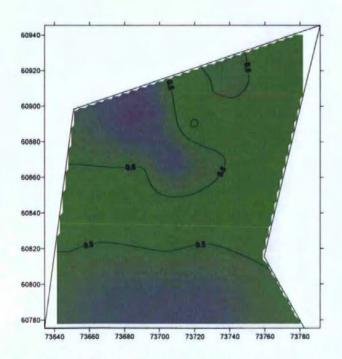
e: (from Table 7-5/Table 7-6) SADIE associations for HPA (bot) 2006 leatherjackets & 10cm shear



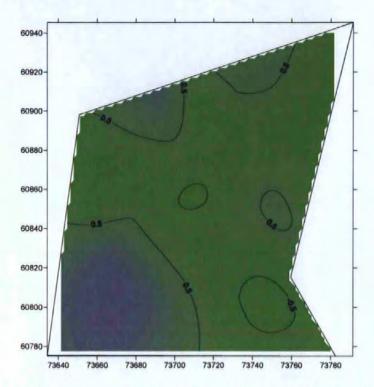
f: (from Table 7-5/Table 7-6) SADIE associations for HPG 2006 Leatherjackets & moisture



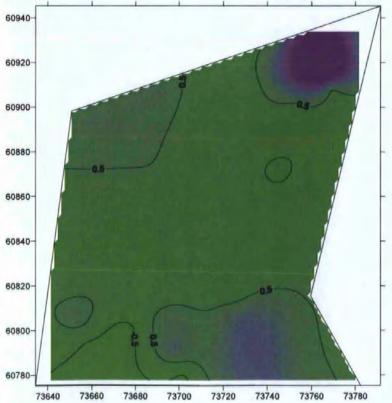
g: (from Table 7-5/Table 7-6) SADIE associations for HPG Leatherjackets 2005 & 2006



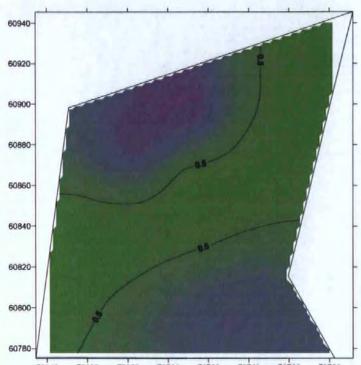
h: (from Table 7-5/Table 7-6) SADIE associations for HPF 2006 leatherjackets and wireworms

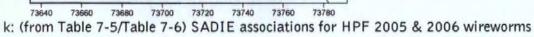


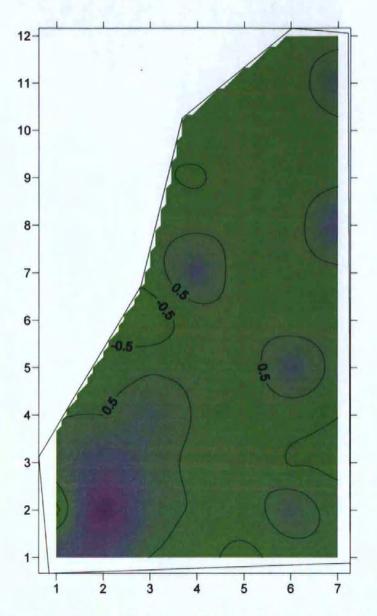
i: (from Table 7-5/Table 7-6) SADIE associations for HPF 2006 Leatherjackets & moisture



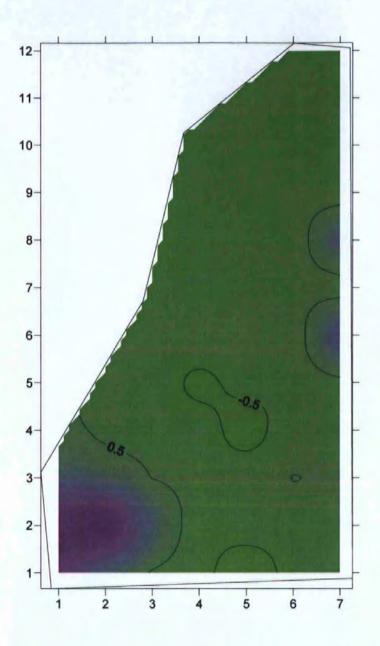
j: (from Table 7-5/Table 7-6) SADIE associations for HPF 2005 & 2006 Leatherjackets



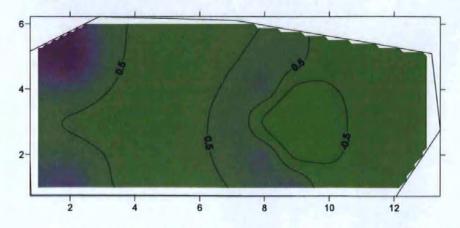




I: (from Table 7-5/Table 7-6) SADIE associations for HPA (top) 2006 Leatherjackets & 10 cm shear



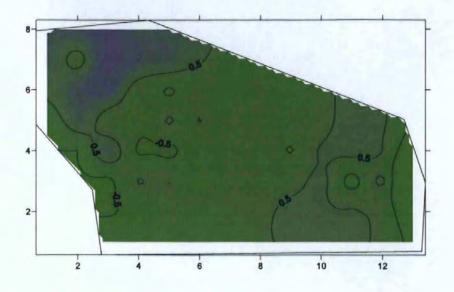
m: (from Table 7-5/Table 7-6) SADIE associations for HPA (top) Leatherjackets 2005 & 2006



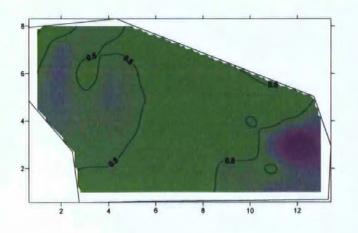
n: (from Table 7-5/Table 7-6) SADIE associations for Broad Park 2006 & 2005 wireworms



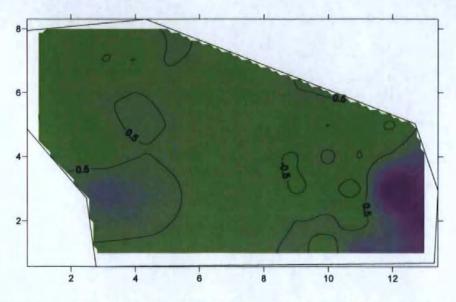
o: (from Table 7-5/Table 7-6) SADIE associations for HPC (cores) wireworms 2005 & 2006



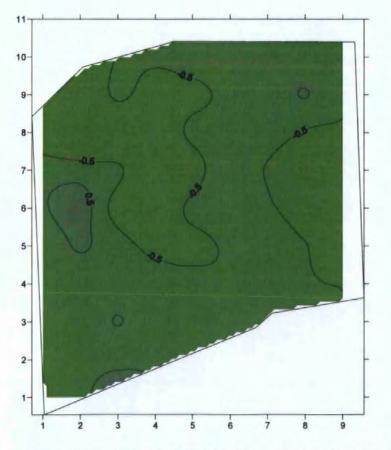
p: (from Table 7-5/Table 7-6) SADIE associations for Hills small 2006 Leatherjackets & moisture



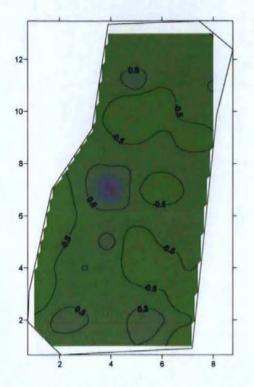
q: (from Table 7-5/Table 7-6) SADIE associations for Hills small 2006 Leatherjackets & 5cm shear



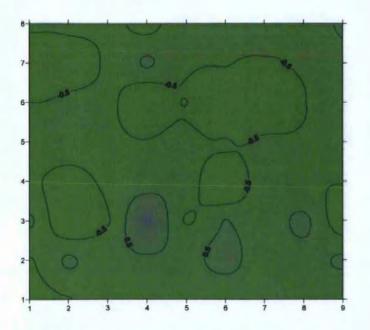
r: (from Table 7-5/Table 7-6) SADIE associations for Hills small 2006 Leatherjackets & 10cm shear



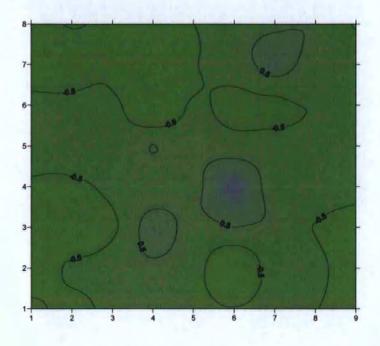
s: (from Table 7-5/Table 7-6) SADIE associations for HPG 2005 leatherjackets & wireworms



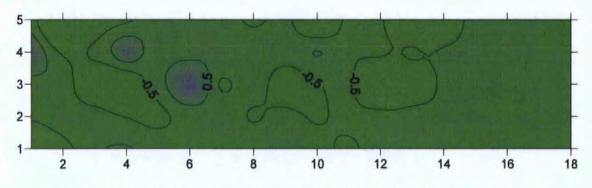
t: (from Table 7-5/Table 7-6) SADIE associations for HPC (cores) 2005 wireworms & moisture



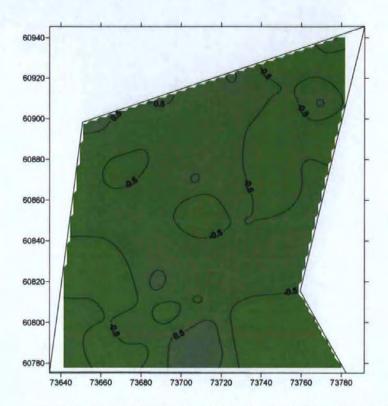
u: (from Table 7-5/Table 7-6) SADIE associations for HPA (bot) 2005 leatherjackets & moisture



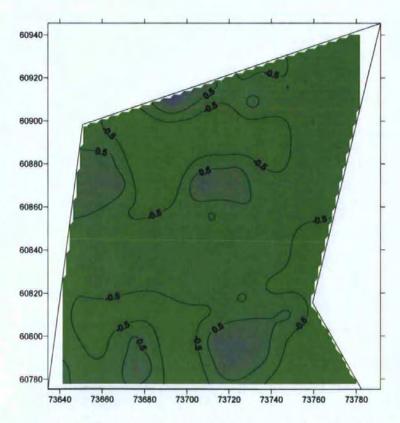
v: (from Table 7-5/Table 7-6) SADIE associations for HPA (bot) 2005 wireworms & leatherjackets



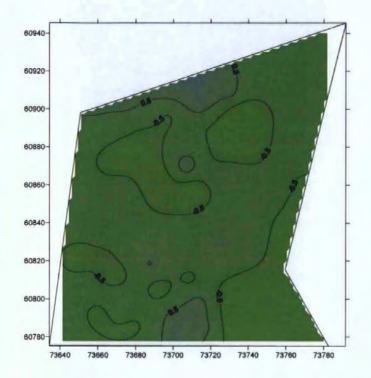
w: (from Table 7-5/Table 7-6) SADIE associations for HPC (traps) 2005 wireworms & moisture



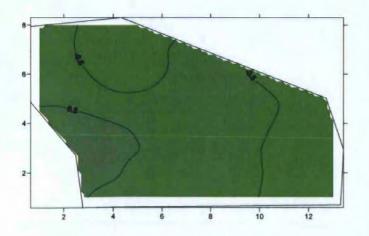
x. (from Table 7-5/Table 7-6) SADIE associations for HPF 2006 Leather jackets & 10cm shear



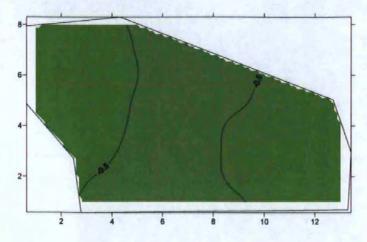
y. (from Table 7-5/Table 7-6) SADIE associations for HPF 2006 wireworms & 5 cm shear



z: (from Table 7-5/Table 7-6) SADIE associations for HPF 2006 wireworms & 10 cm shear



aa: (from Table 7-5/Table 7-6) SADIE associations for Hills small 2006 Leatherjackets & wireworms



ab: (from Table 7-5/Table 7-6) SADIE associations for Hills small 2005 & 2006 leatherjackets

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