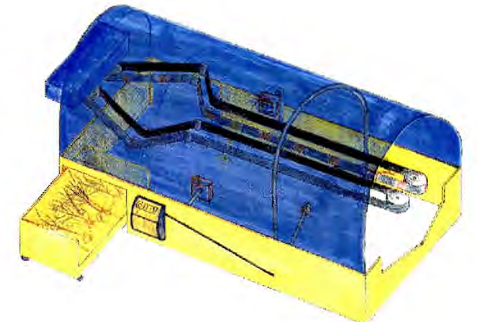
Control of the Large Pine Weevil, *Hylobius abietis*, L.



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A thesis submitted for the degree of
Doctor of Philosophy of the University of London
And the Diploma of Imperial College

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Abstract

The Large Pine Weevil, *Hylobius abietis*, is a serious pest of temperate forestry throughout Scandinavia, the United Kingdom, the Republic of Ireland and Eastern Europe. In the absence of control measures, 100% mortality of planted saplings frequently occurs. The main methods of control involve the prophylactic use of pesticides, although the use of silvicultural methods and nematodes are becoming common.

This thesis presents a new treatment system for the application of pesticides to saplings of the species, Sitka spruce and Scots pine. This system was shown to provide a level of protection for these saplings against *H. abietis* for at least one year, at a level equal to current methods. Aspects of the design, efficacy and safety of this new system are presented.

A second part of the thesis examines the anti-feedant and repellent effects of pyrethroid insecticides against *H. abietis*. This work clearly demonstrates the ability of *H. abietis* to detect and avoid pesticide treated food sources and also to potentially recover after pesticide exposure.

The thesis concludes by discussing the future of pesticides and anti-feedants in European forestry and the potential for this work to be exploited.

Declaration

The work with in this thesis is entirely my own except for the parts listed below

Chapter 3

The design of the field trials was carried out by Stuart Heritage, Forest Research, Forestry Commission, in order to ensure that the data would meet criteria allowing it to be submitted to the Pesticide Safety Directorate. In addition the field trial work was carried out by Forest Research, Forestry Commission, UK.

Chapter 6

The experimental work investigating the recovery of weevils after exposure to pesticides was carried out by a student of Imperial College working on an undergraduate project. However, all experimental design was my own.

Signed Daniel Rose

Confirmed Graham Matthews

Simon Leather

Date: September 2002

Acknowledgements

To my two supervisors, Professor Graham Matthews and Dr Simon Leather for your vast wealth of knowledge and advice that you have given to me during my PhD

To my friends and office mates for putting up with me even when I did my best to send you all insane

And especially to Clare and my family for your support that drove me to achieve something so worthwhile,

Thankyou.

Copyright Statement

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Chapter 1
Introduction and Literature Review

1.1 Introduction

Although reports of *Hylobius abietis* as a temperate forest pest have been made since the early 1800s (Blomqvist, 1883; Holmgren, 1867; Ratzeburg, 1839), the available literature is often conflicting despite the many of years of research on the pest. The Large Pine Weevil (*H. abietis*) is relatively long lived and its development varies in different climates, but Bejer-Petersen *et al.* (1962) list no less than nine different opinions by various authors as to its development and biology. A range of 3-4 months up to 21-22 months has been recorded for larval development, which may even be as long as five years in very cold climates. A key problem with the study of *H. abietis* is partly the difference in behaviour in laboratory conditions compared to the field and secondly, the degree of plasticity in its development and behaviour in different climatic regions. Even micro-climatic variations can have an effect, for example, the condition of the stump in which the larva feeds and the temperature within the stump, (Bejer-Petersen *et al.*, 1962). Therefore there are many confounding views of *H. abietis* and its development to the extent where agencies such as the British Forestry Commission focus their research on practices which results in a reduction in damage to stocks and less towards an understanding of the ecological and biological mechanisms behind the damage it causes.

Hylobius abietis is one of the most damaging pests of temperate forestry (Langstrom, 1982). It causes extensive damage to restocking sites, devastating stocks of coniferous saplings and even broadleaf species (Plate 1. 1). *H. abietis* is found across Northern Europe and as far afield as Asia. However, it is not found in North America, instead *H. pales* Herbst. (Coleoptera: Curculionidae) , fulfils a similar niche. *H. abietis* is often found with a related species, *H. pinastri* Gyll. (Coleoptera: Curculionidae), with the latter typically occupying 2-23% of trap catches, but usually around 10% in Fenno-Scandia (Luik & Voolma, 1989).

Until recently *H. abietis* was not a very serious threat to UK forestry, but when the Forestry Commission changed its planting regulations in the mid 1980s and shifted to the use of clear fell sites rather than planting on new land, the problem became acute. At present, the most effective method of control in the UK is the prophylactic use of pesticides (Moore, 2001), however, in Sweden the treatment of seedlings with pesticides will be prohibited in the near future. Therefore in Sweden, there is added pressure to fund research into the use of host plant volatiles as attractants to be used in push-pull, trapping strategies and silvicultural techniques (Orlander & Nilsson, 1999; Orlander *et al.*, 1997). Other nations are investigating methods of control, such as accurate forecasting of potential damage, so that control can be applied at the correct and most efficient time (Moore, 2001; Wilson *et al.*, 1996).

Plate 1. 1 Spruce Damage

Typical damage by *H. abietis* on a Sitka spruce sapling. Note the exposure of the lower layers of wood after the bark has been completely removed. Girdling of the stem in this nature can result in wilting of the sapling and its eventual death.

Despite the shorter seasons and the colder weather in Sweden, *H. abietis* actually poses a greater problem and threat there than in England. The occurrence of damage is more intense, although over a shorter period (Evans H.F., *Pers. Comm.*). This may be due to the single emergence of the whole population, which attack the saplings at once, rather than the staggered emergence as in the UK.

1.2 A Description of *Hylobius abietis*

The Large Pine Weevil, *H. abietis* belongs to the family Curculionidaeulonidae, super-family Rhynocophora, and order Coleoptera. They have a Holarctic distribution, covering the Scandinavian region, across to the UK (Langstrom, 1982). *H. abietis* is one of the most destructive pests of reforestation areas (Langstrom, 1982) frequently causing 30-100% mortality on untreated saplings and eating a range of trees including Pine and many broadleaf species (Heritage, 1996).

Measuring 10-20mm in length, averaging 15mm, *H. abietis* closely resembles another forest insect, *Pissodes* weevils, at both adult and larval stages. However these are generally about two-thirds the size of *H. abietis*, and have antennae attached near the middle of the snout. *H. abietis* has jointed antennae which attach to the tip of the snout, but often fold backwards, so they appear to originate from the middle (Scott & King, 1974). Another way to distinguish the two is that *H. abietis* can grip a finger with considerable force, often tearing the skin and is not easily dislodged.

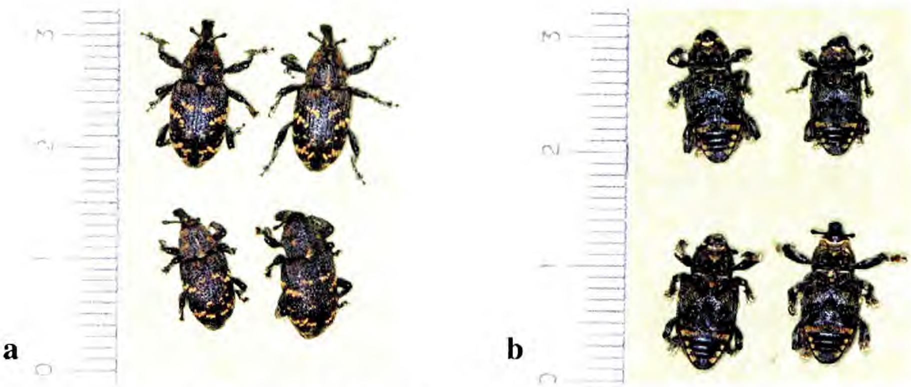


Plate 1.2 **Typical size range of *Hylobius abietis* varies from 10-20mm. There is little difference between males and females, except that males have a small dimple on the last segment, which is covered in fine hairs. A – Dorsal view. B – Ventral view (Figure 1.2b, top right – male; bottom left – female).**

Males and females can be distinguished by the different shapes of the lower abdomen. Females have a more pronounced and rounded abdomen, whilst males have a slight depression in the last segment, which is covered in fine hairs (Plate 1.2). The elytra of *H. abietis* completely cover the abdomen; they are a purplely brown in young adults becoming black later in life (Plate 1.3). These wing cases house a pair of long veinated wings that enable dispersal when the temperature rises above 16°C (Solbreck & Gyldberg, 1979). The wing cases show a mottled brown pattern across the back which greatly increases crypsis of the insect (Scott & King, 1974). This patterning is actually made up of small bristles that tend to get lost in later life. On the underside of *H. abietis* are a series of longitudinal ridges and furrows that have oblong shaped indentations (Scott & King, 1974).

Plate 1.3 ***Hylobius abietis*** **feeding on the bark of a Scots pine sapling.**

The long slender nose is called a rostrum, and at the end of this is a set of powerful jaws. *H. abietis* feeds on the bark of most coniferous species of tree and also many deciduous species. It typically starts feeding from the bottom of the stem (although this may be reversed in deciduous trees) and eats down to the cambial layers (Crystal, 1937). Once the tree has been girdled (bark removed all the way around the stem) the sapling quickly wilts and dies.

1.3 Life cycle of *Hylobius abietis*

Hylobius abietis is capable of breeding once per year and may live for up to four years. In addition larval development can take up to four years in cold climates. Therefore, there is a constant efflux of adult *H. abietis* (Eidmann, 1979; Heritage, 1996). However, female fecundity does decline each season, at least in the laboratory. Lipoprotein content, which is a measure of female fecundity, is less in the second over-wintering period than the first (Guslits, 1969). In extensive mark-recapture studies, adult weevils were caught, marked and caught again up to two years later (Moore, 2001). This is indicative of the longevity in the field being equal to that in the laboratory. Each year there are two different periods of emergence. Hibernating adults reappear in the spring, along with new adults, and a second batch of newly emerged adults appear for a brief period in the late summer (Leather *et al.*, 1995; Scott & King, 1974).

The adult weevil begins its life cycle in the early spring when temperatures typically rise above about 8 °C, but may be active at temperatures as low as 2 °C (Heritage & Johnson, 1997). Adults that are emerging from their hibernation are ready to mate immediately, but usually feed for a short period first. It is uncertain whether old generation adults stay on the breeding ground or migrate away (Nordenhem, 1989). Figure 1. 1 shows the physiological and ecological lifecycles, adapted from Leather *et al.* (1999) and Heritage (1996).

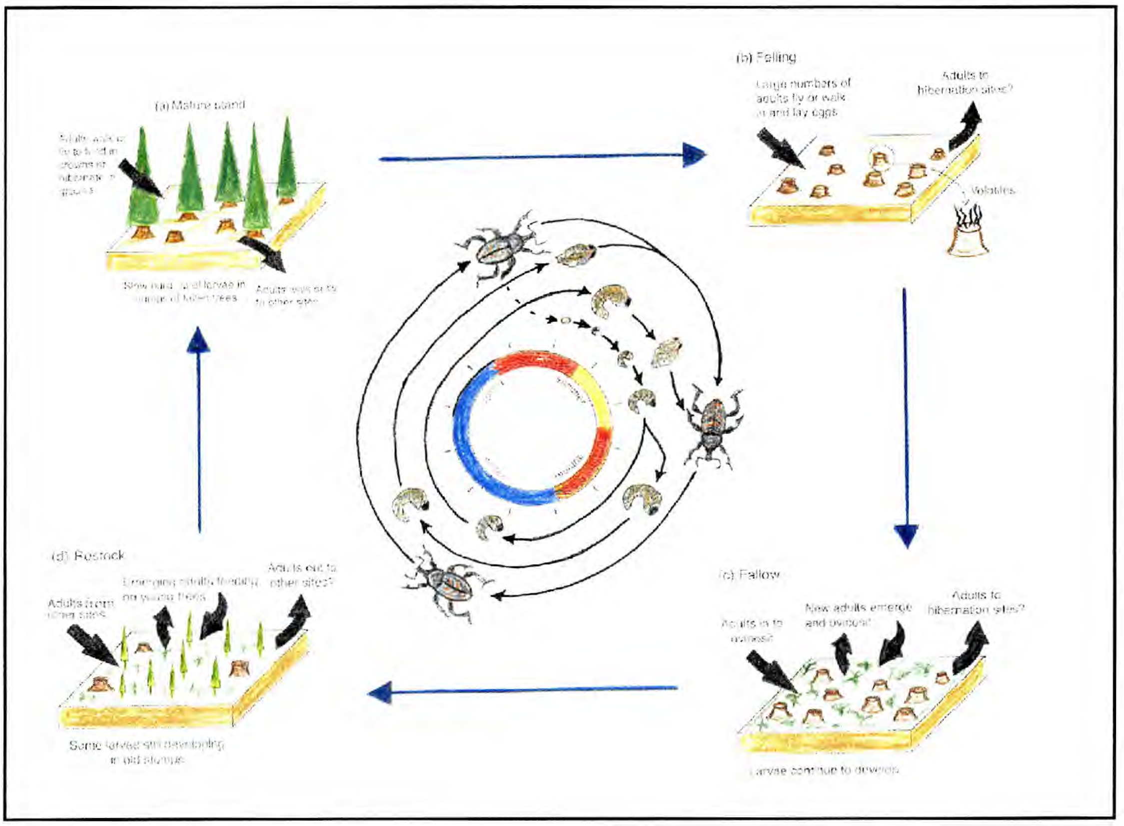


Figure 1. 1 **Physiological and ecological life cycle of *Hylobius abietis*. The life cycle starts in early spring when a new clear fell site is created, adults migrate to the site and oviposit around the old stumps (Outer circle). The larvae then develop over 1-4 years inside the stump (Inner circle). Emerging adults feed on the newly planted**

saplings causing widespread mortality (Modified after Heritage (1996) and Leather *et al.* (1999)).

Few pheromones are used by *H. abietis*, with only one short range sex pheromone that has been identified, but this is believed not to have any long-range effects (Tilles *et al.*, 1988). This appears to be emitted only by females. In addition the presence of a long range pheromone has been suggested by Selander (1978), although no supporting evidence has been found (Nordlander *et al.*, 1986; Tilles *et al.*, 1986).

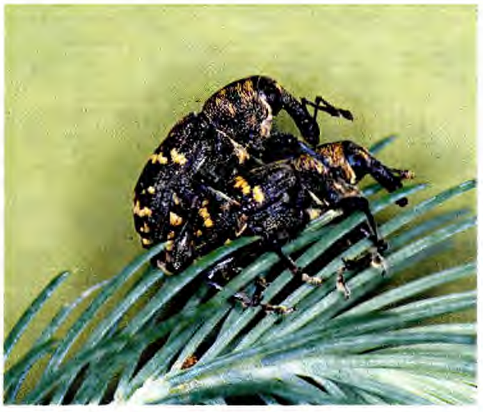


Plate 1.4 Mating Weevils

A male and female *H. abietis* copulating on a Sitka spruce sapling. Mating can often be witnessed on the underside of billet traps in the field.

There has been some debate as to where females actually oviposit after mating. Traditionally it was believed that females make notches in the bark of the stump and deposit their eggs inside (Nordenhem & Nordlander, 1994; Scott & King, 1974). However, there are an increasing number of authors that have reported that *H. abietis* are capable of depositing their eggs in the soil and the larvae then migrate to a suitable pine stump (Nordenhem & Nordlander, 1994; Pye & Claesson, 1981). This may well have the advantage that the larvae can choose between the best food sources, despite the added risk of predation (Nordlander *et al.* 1997; Salisbury & Leather, 1998).

A related species, *H. radicis* Buchanan, deposits most of its eggs in the soil in the wild (Wilson, 1975), yet bizarrely will deposit its eggs in excavated niches in bark in the laboratory (Hunt *et al.*, 1992). Apparently depositing eggs in to an excavated niche is typical of ancestral Curculonidae behaviour (Anderson, 1993). This discrepancy may be due to a lack of choice in the laboratory that causes the weevil to select the only available material and then provide an additional level of care by carefully placing the eggs away from potential predators. Alternatively, *H. congener* Dalla Torre. (Coleoptera: Curculionidae), deposit their eggs in excavated niches and then cover them with frass and bark chips (Lynton-Martin, 1964). Nordlander *et al.*

(1997) suggested that *H. abietis* shows this behaviour to avoid predation by conspecifics inside the stump and that weevil larvae are better at selecting suitable feeding material than ovipositing females. This is quite possibly the only time that larvae will be able to select optimal conditions. Once inside the stump the surrounding soil temperature and humidity can greatly affect the growth and development of the larvae inside the underground stumps and roots (Christiansen, 1971b). When there is a risk of the surrounding soil drying out, females may only lay their eggs in the stumps.

Hylobius abietis usually oviposit in June through to late summer (Guslits, 1969), but this can vary depending on the region (Christiansen, 1971a). The eggs typically take 12-14 days to hatch at 15 °C, laying on average 0.45 eggs per female per day (Salisbury, 1998). Similar findings were reported by Bejer-Peterson *et al.* (1962), although Nordenhem and Nordlander (Unpublished) suggest that this figure may be much greater and Guslits (1969) indicates that laboratory reared females can lay an average of 62 eggs in the first year and 40 in the second year. Leather *et al.* (1999) reported that a minimum of 25 eggs are laid per year per female on average. However, this may be less for 2 year old weevils, which have fewer fat reserves after the second hibernation to build oocytes (Guslits, 1969).

Nordenhem and Nordlander (1997) have shown that under field conditions in Uppsala Sweden, hatching takes about a month. Laboratory experiments showed that there was an average viability of 72.2%. As the eggs develop, the mandibles can be clearly seen as two distinct black markings, approximately 1mm x 0.5mm. This darkening of the mandibles may indicate that the mandibles harden to aid emergence from the egg (Salisbury, 1998).

On hatching, the larvae commence feeding on the pine stump by making long, ever increasing burrows mainly in the cambial region (Scott & King, 1974), although Salisbury (1998) has reported larval feeding in the heartwood in the laboratory. It remains to be seen if this also occurs in the field. The long rambling tunnels of *H. abietis* include several ventilator tunnels to the surface, and densely packed frass behind itself (Scott & King, 1974). Whether this is a defensive mechanism to stop predators coming up from the rear, or just a by-product, is unclear.

The larvae can spend up to five years developing if the conditions are cold and the stump quality is poor. The norm is two years (Bejer-Petersen *et al.*, 1962). However in the UK, in warmer conditions larvae may have completed their development by the autumn and then emerge in early spring (Scott & King, 1974). Most commonly, larvae in upland UK take over a year to develop and emerge in the second autumn to feed (Moore, 2000). Development inside the stump is greatly affected by temperature, but not by surrounding soil moisture content (Christiansen, 1971b). The optimum temperature was shown to be 23 °C in the laboratory, but no upper or lower limits were established. Eidmann (1964) reported that eggs failed to develop at 30 °C, but fully grown larvae were able to tolerate this temperature. It was also shown that there is a critical temperature, which affects the larval entry to diapause. This is a state of temporary dormancy, which the last instar may or may not enter. If external temperatures are warm enough, i.e. if the larva is fully developed and it is still summer, then it can emerge from the stump. If it is winter, however, the larva will stay dormant until the next spring. Eidmann (1964) showed that this critical temperature was between 20 °C and 25 °C.

At the end of feeding, prior to pupation, the larvae hollow out a pupal chamber and form a thatch work of tightly knit wood fibres. This tends to be relatively near the surface in the wood or bark or both, depending on the thickness of the bark (Christiansen, 1971b; Scott & King, 1974). *H. abietis* form naked pupae, with the soft body changing shape and hardening later (Leather S.R., *Pers. Comm.*). The larvae may stay in this cell and over-winter as either a third or fourth instar larvae (Leather *et al.*, 1999). At this stage the larvae enter a stage of cryptobiosis and can be supercooled to a temperature of -12.6 °C before they die (Luik & Voolma, 1989). When supercooling occurs, all metabolism drops to a minimum and the larvae dry out. However, this is not a particularly low temperature compared to many temperate insects such as *Polygraphus polygraphus* L. which supercools to as low as –39°C (Luik & Voolma, 1989). This is an obvious adaptation to their sheltered lifestyle (Leather *et al.*, 1999).

There is generally a two-phase emergence period; some adults emerge in late autumn, whilst others may emerge the following spring (Nordenhem, 1989). This is often dependent on the time it has taken the new generation of larvae to develop. Those adults emerging for the first time in late summer hibernate in the soil over the winter. Consequently there are three different stages of insect emerging in the Spring (Scott & King, 1974). This consists of (I) old weevils which have already produced broods; (II) weevils which have emerged the previous summer or autumn and are ready for their first egg laying; (III) weevils which have over-wintered in pupal chambers (Scott & King, 1974). There is a distinct advantage to emerging in the autumn as it gives the weevils an opportunity to feed and allow their ovaries and flight muscles to develop (Nordenhem, 1989). Therefore, on emergence from hibernation in the spring, they can immediately breed and select the best males. Weevils emerging from pupal chambers in the spring, on the other hand, must feed for a short period before they are ready to reproduce. In addition, weevils emerging in late summer will not develop their flight muscles until the following spring (Nordenhem, 1989). It may well be safer to over-winter in the pupal chamber and therefore have increased survival chances.

Hylobius abietis emerges in early spring, often as early as March. There is a brief period when major damage can occur, which may well decline as the breeding begins in late May or early June. The emergence of weevils can continue from May through to late October, before the weevils return to the soil to hibernate (Scott & King, 1974).

1.4 Current and previous control methods

Hylobius abietis is the only British forest insect pest against which prophylaxis is routinely carried out (Stoakley & Heritage, 1990). The damage caused by *H. abietis* is often disproportionate to the scale of the insect problem. It has been found that *H. abietis* will move continuously across a clear fell until it encounters a young sapling. At this point it will feed until the sapling is killed (Heritage S.G., *Pers. Comm.*). This can occur very quickly and the sapling ring-barked (girdled) with the water transporting xylem vessels quickly severed (Heritage S.G., *Pers. Comm.*). In warmer conditions (approximately 25 °C) the sapling very quickly wilts and dies; yet in colder conditions (approximately 15 °C) they manage to survive the damage (*Pers. observations, unpublished data*). Weevil populations may range from 150,000 adults per hectare (Heritage, 1996) to 220,000 adults per hectare (Leather *et al.*, 1995) with as few as 2000 newly planted saplings in the same area. Consequently there is heavy damage and losses are often seen when there is no pesticide protection (Heritage, 1996). Any form of control, therefore, must either suppress the population to incredibly low levels, or else protect the tree from attack.

The period over which saplings must be protected can actually be quite extensive, for example Sitka Spruce (*Picea sitchensis* (Bong.)) is susceptible to damage for up to two growing seasons. It is likely that after this time the quantity of resin produced inhibits feeding in *Pissodes* weevils (Tomlin & Borden, 1997). However, Douglas fir (*Pseudotsuga menziesii* (Mirb.)) will remain prone to damage and mortality for several growing seasons (Heritage, 1996).

There is a huge unpredictability in the timing of maximum damage. There are generally peaks in April/May and August/September, though this is dependent on local weather conditions both during the period of larval development and at the time of emergence or feeding (Heritage, 1997). Therefore, the widespread use of insecticides would be inefficient, particularly as *H. abietis* tend to go to ground when it is damp, thus avoiding direct contact with pesticide (Heritage S.G., *Pers. Comm.)*

Most insect control systems rely on suppression of the pest population. Unfortunately this cannot be the case with *H. abietis* due to the considerable population size and sheltered lifecycle. There is currently no way of treating the stumps to kill off the emerging adults and developing larvae, at least with pesticides (Heritage, 1996). Secondly, each insect is capable of killing the sapling in a very short time so there is a zero tolerance for damage. This situation is similar to that of many pests of fruit, for example, *Ceratitis capitata* that lays eggs in the fruit of mangoes, coffee and oranges. Consumers do not want maggots in their fruit so this is also a zero tolerance pest management scheme (Jones 1989). Therefore, the pest management scheme used to deal with *H. abietis* is based on prophylactic control.

Chemical prophylactic measures used against *H. abietis* can be divided into three broad categories. These are pre-planting, during and post-planting measures. Pre-planting methods include dipping, pre-planting spray and the Electrodyn sprayer conveyor (Heritage, 1996). At the time of planting carbosulfan granules have been used, and after planting, pesticides such as permethrin and gamma HCH may be applied (Heritage, 1996).

1.4.1 The dipping system

The dipping system has been in use for several years. During this time the exact methodology has varied quite substantially to increase daily output. The original dipping systems involved the treatment of individual trees by hand. However, this was time consuming and there was a huge opportunity for operator exposure. Since then nurseries have adapted the system to improve daily output. In one observed system the saplings were bundled into groups of approximately 400 saplings and spread along a wooden bar with an attached tarpaulin sheet (which protects the roots). The tarpaulin is then folded over the roots of the saplings and securely fastened. Only the foliage is immersed in the pesticide bath, the roots must not be treated. The aim when treating pine saplings is to only treat the lower 15cm of stem, from 2cm below the root collar, to 13cm above it. The dipping process treats a much greater area than this, thus using more pesticide (Heritage, 1997).

The dipping system, although effective and based on the use of simple technology, has several disadvantages. The first is that there is a potentially high risk of operator exposure. The second is that the plants must be dried before they can be subsequently handled, especially as not all workers, for example, packers and planters, are required to wear full protective clothing. The current system uses a heated wind tunnel to dry the saplings, but there is always the risk of over drying the saplings and damaging the roots (Heritage, 1997). Mechanised systems are being developed to automate the process, but these currently use huge amounts of electricity to dry the saplings.

1.4.2 Pre-plant spray

The use of a pre-planting spray is not widely used, as the Electrodyn and dipping processes treat most of the saplings from Forestry Commission (FC) nurseries. However, spraying is a useful tool in research trials. Knapsack sprayers are used to treat small potted saplings, and this can be done at dipping or top up concentrations.

Additionally there are some mechanical systems that can treat many more trees at a higher rate (Heritage, 1997). Recent developments in spray application have addressed issues of repetitive strain injury (RSI) when operators are repeatedly required to depress a heavy trigger. This is the case in spot applicators, which are used to deliver a metered dose to saplings. A new system has been designed by Micron Sprayers Ltd, which uses electronic triggers and controls to deliver an accurate metered dose to forest transplants. The new system avoids the associated health risks typically coupled with other backpack sprayers (Anonymous, 2002).

Typically these systems use permethrin, (usually Permasect 25EC), although gamma HCH (Lindane) as has chlorpyrifos has traditionally been used for dipping (Heritage, 1997). Lindane has since been revoked by the Pesticide Safety Directorate (PSD), as it is a persistent organo-chlorine (Heritage, S.G., *Pers. Comm.*). This is reinforced by a pan European directive to discontinue the use of lindane.

The systems used in Sweden are slightly different from those used by the Forestry Commission. Concerns in Sweden over the accumulation of permethrin contaminated material have led to the development of the nursery spraying techniques (Torstensson *et al.*, 1999). One such method involves spraying saplings whilst they are planted in the field with a hand sprayer and a nozzle aimed horizontally at the stem of the sapling. The intended deposit was 2ml per seedling, which equates to 4 litres/hectare. This is a high value compared to the Electrodyn system (0.4 l/ha), but lower than conventional dipping (approx 20 l/ha). However, the concentration of the Electrodyn spray solution is substantially higher (Section 1.4.3).

Spraying pesticides in the field compared to an enclosed system in a treatment centre has two key disadvantages. Firstly, there is the inherent risk that over several seasons the permethrin will accumulate in the soil of the nursery. Secondly there is the increased operator risk as the trees are treated one handling stage earlier and therefore there is greater risk of pesticide exposure. However, Torstensson *et al.* (1999) argue that handling will occur when the plants are actually drier than those handled in a treatment centre (which are frequently moist after treatment) and is therefore much safer.

Torstensson *et al.* (1999) tracked the movement of permethrin both in the soil and on different parts of the saplings using chemical analysis. In their tests, it was found that permethrin had a half-life of 3-4 months and 90% had been broken down by 7-9 months. An initial concentration of 4% would maintain the required level of active ingredient (0.30μg mm^{-2}) for two years. This is in line with the quantities set by the Forestry Commission (FC) for pre plant application (Heritage S.G., *Pers. Comm.*). Similar patterns were observed in both the soil and on the trees, however permethrin was shown not to move deeper than 10cm into the soil. This was attributed to the very strong binding action of permethrin to soil, preventing its transport.

1.4.3 The Electrodyn sprayer conveyor

The UK Forestry Commission has developed the Electrodyn sprayer conveyor for use as a treatment system against the Large Pine Weevil to protect newly planted coniferous saplings (Heritage *et al.*, 1997a). This system was adapted from the hand-held sprayers that were originally used to treat cotton with an ultra-low volume insecticide spray in Africa. The original system was patented by Zeneca Agrochemicals, formerly ICI plc, in 1976. An indoor conveyor system was approved to be used by the Forestry Commission only, although an application was filed to develop it commercially (Heritage *et al.*, 1997a). Since this initial date, the PSD (Pesticide Safety Directorate) decreed that the adjuvant cyclohexanone was too toxic, but Health and Safety Executive (HSE) has permitted its use in a well ventilated and air filtered system. Therefore the Electrodyn system will require a new pesticide formulation based on non-toxic oil based carrier, if its use is to continue. Unfortunately development of a new formulation incurs extensive cost and registration (Heritage S.G., *Pers. Comm.*).

The Electrodyn sprayer conveyor uses a specially formulated semi-conductive oil based carrier with an electrical resistivity of 10^4-10^6 ohm m^{-1}. This is gravity fed through a special plastic nozzle (Matthews, 2000). The nozzle is impregnated with carbon and a 20,000-volt electric current is applied across the nozzle. The resulting ligaments are highly charged and break up into even sized droplets of less than 10μm in diameter. The droplet size is easily controlled by altering the flow rate and the charge applied to the nozzle. The highly charged droplets travel with a force fifty times greater than gravity towards the nearest earthed surface. Any sapling (which is earthed) passing through the droplet cloud is immediately coated in pesticide (Heritage *et al.*, 1997a).

There are several key advantages of the Electrodyn system over the other methods of sapling treatment. The biggest advantage is that operator exposure is kept to an absolute minimum due to the enclosed spray booth. The total volume of pesticide is greatly reduced to a mere 200ml of Permethrin per 1000 saplings. Therefore, there is no need to dry the saplings, which can damage the roots. Secondly, the pesticide product is provided in the correct concentrations and can be emptied into the sprayer in an enclosed booth. Therefore, handling and mixing of the product is minimal. Finally, the highly volatile solvent evaporates quickly and the treated sapling is usually dry by the time it reaches the end of the conveyor belt (Heritage *et al.*, 1997a).

There are also disadvantages to the Electrodyn system apart from the problems posed by increased pesticide registration. Permethrin is due to be removed because of an EC directive (EEC 91/414), which requires manufacturers of all pesticides to provide extensive data with regards to performance, safety and environmental impact. Unfortunately many companies are unwilling to provide the data for older pesticides (which would involve some cost to them) because they have more profitable new products for which they have already obtained these data. Despite field trials showing that permethrin treated trees have improved protection over untreated trees (Heritage *et al.*, 1997a) it is unclear how this occurs. There have been few studies to see if this is a repellency effect (as permethrin can act as a repellent) or if it is due to the weevil's death by touching deposit from the foliage or perhaps by feeding on pesticide residue on the stem. Observations of some treatment systems show that many of the droplets do not appear to penetrate the foliage and do not reach the stem that needs to be protected. In contrast the dipping totally immerses the sapling and ensures 100% coverage of both the stem and foliage.

The Electrodyn system has been designed to work with Permethrin 12ED, an oil based pesticide formulation. Field trials have shown that plant vigour may be reduced, manifesting itself as reduced root growth potential (RGP), reduced first year increment or even death. The solvents (cyclohexane) in the permethrin formulation for the Electrodyn are particularly phytotoxic and this effect is exacerbated by the cold storage. The saplings must not be stored for longer than 14 days (Heritage *et al.*, 1997a). This limited period increases the need for a very efficient system of lifting-treating-planting. Frequently heavy periods of rain will prevent planting occurring and the saplings may be damaged if the period in cold storage has to be extended.

1.4.4 Applications at time of planting and post-planting

A recent development in post-planting treatment is carbosulfan slow release granules with carbosulfan carbamate pesticide. Carbosulfan is the only slow release insecticide approved for use in treating pine trees in the Forestry Commission (Heritage *et al.*, 1997b). This carbamate is usually applied to the soil and absorbed by the plant, resulting in systemic protection. It is not particularly soluble in water, therefore it avoids the usual problems of leaching. Unfortunately it has a short half-life and only lasts a few days in the soil and on plant tissues. However, the slow release formulation compensates for this problem.

Carbosulfan granules are made of a plastic polymer that slowly releases the carbosulfan active ingredient and also protects it from degradation. The granules are formulated and then placed in the potting hole at the time of planting. Unlike the other types of treatments, carbosulfan granules provide adequate protection for the first two growing seasons. This removes the need for a top-up spray in the second year (Heritage *et al.*, 1997b). This process is dynamic with the granules continuously releasing insecticide and the plant continually absorbing it and then breaking it down.

Specific applicators have been developed to add a fixed volume of granules (usually 10g) to the soil (Saunders, 1996). This greatly reduces the risk of operator contamination from the pesticide. This is very important as carbamates are toxic to fish, birds and mammals, including humans (Heritage *et al.*, 1997b).

Carbosulfan has a stomach action on *H. abietis* resulting in a small level of feeding before death occurs (Heritage *et al.*, 1997b). The application is reasonably specific because the pesticide is locked away in the granules and only insects feeding on the sapling will be poisoned. Below-ground-fauna may come in to contact with the soil region containing pesticide, and predators or scavengers feeding on dead *H. abietis* could pick up a lethal dose. However, carbosulfan breaks down very rapidly in the soil and is therefore only maintained in the environment when taken up by the plant (Heritage *et al.*, 1997b).

In field trials conducted within the Forestry Commission, carbosulfan granules provided similar protection to dipped plants, and significantly more protection than untreated plants (of which 80-89% in first and second years were killed). However, there were key differences. Namely, dipping with permethrin provided excellent protection in the first year with less than 5% of plants being killed, but in the second year protection dropped to an unacceptable 70%. Although carbosulfan granules are not as persistent as permethrin, the duration of protection seen in the field was due to the slow release polymer formulation. Thus, the carbosulfan granules maintained a high level of protection in the second year although there was an initial period of high damage in the first year compared to the permethrin treated saplings. This is partly due to a lag in protection until the plant takes up enough pesticide to give systemic protection. This typically takes about a month. Immediately after planting the saplings are vulnerable to damage and this was reflected in the Forestry Commission trials. To combat this the saplings were also treated with a permethrin spray at the time of planting, but this requires a doubling of the effort and cost to protect the trees (Heritage *et al.*, 1997b).

There have been no phytotoxicity trials on broadleaf species, but the long-term effects of the carbosulfan granules on Douglas Fir (*Pseudotsuga menziesii*) and Larcio pine (*Pinus laricio,* Poir.) have been investigated (Julien, 1998). This study concluded that there were no differences in the growth performance between treated and untreated or treated controls.

Further work is obviously needed in this area, but the use of pesticides in slow release formulations may well be banned by the PSD because of the risk to wildlife, including natural enemies and predators of *H. abietis* due to the concerns expressed by some environmentalists (Matthews, G.A., *Pers. Comm.*). The total area around the tree that is treated may affect future decisions. Carbosulfan granules and the released carbamates are confined to the root area of the sapling, which should reduce environmental risk. It remains to be seen whether carbosulfan granules are effective in the long term.

Frequently top-up sprays are used later in the second growing season in addition to the prophylactic measure used. This is often the case if the prophylactic treatment is only effective for a year (Heritage *et al.*, 1997b). This often correlates with the two peaks in population numbers of *H. abietis,* in early April and late summer (August-November). It is usually the case that changes in levels of damage occur very rapidly, even overnight. Therefore, treatment in response to damage is usually too late (Heritage *et al.*, 1997b). These treatments are primarily with permethrin (Permasect 25EC) used at a concentration of 0.4% or gamma HCH (an organochlorine), but this has had its licence revoked and all stocks must be used up within the next two years (Heritage *et al.*, 1997b); Matthews G.A., *Pers. Comm.*).

1.5 Physical control methods

There has been a variety of alternative control methods such as physical barriers that have been tried. Unfortunately many of these are either impractical, not effective enough, have an unfavourable impact on the tree or are too costly and time consuming to implement (Eidmann *et al.*, 1996). Such examples include the use of latex paint, which gave very effective protection and reduced feeding damage (Zumr & Stary, 1995). Unfortunately, there was little follow up of this research, presumably because the latex had to be applied by hand in the field. Additionally, the weevils were very quick to exploit the gaps left after poor application.

Physical barriers are exactly what they say and do not offer any level of repellency, as may be the case with pesticides and semiochemicals. Therefore, the weevil may be unforgiving in exploiting poorly applied devices. Eidmann *et al.* (1996) examined the use of physical barriers but only obtained 53% protection at the most heavily attacked sites. On further examination, it was found that correctly applied barriers gave excellent protection, but the high levels of mortality were caused by incorrectly applied barriers, particularly those that were applied in the field. Given that experimental technique is usually stricter and there are greater levels of care taken during an experiment, these results are hardly promising.

A variety of physical barriers have been tested, which range from PVC coated paper, thin Teflon coated PVC barriers (Eidmann *et al.*, 1996), plastic bags (the Struten), plastic fibres (the Berma) to ladies' stockings (the Strumpan) (Hagner & Jonsson, 1995). However, many of these barriers were not as effective as permethrin treatments and were considerably more expensive and labour intensive to apply (Eidmann *et al.*, 1996; Hagner & Jonsson, 1995). This is partly because it is especially difficult to design conveyor systems to apply physical barriers and therefore this process has to be done manually.

Recent developments in physical barrier design have addressed the issue of reduced plant growth and many of the barriers are designed to break away or degrade as the plant grows. These materials are usually biodegradable and therefore will have less impact on the environment than pesticide residues (Eidmann *et al.*, 1996). Unfortunately, there is often higher damage in the second year as the barriers start to degrade. The future development of physical barriers continues and if fast effective application methods coupled with effective second year protection can be achieved, it may be possible for this method to compete with the use of pesticides, particularly as increasing pressure mounts against the use of chemical control.

Current developments in chemistry have led to the discovery of ethylene vinyl acetate (EVA) as an insect barrier (Hoffman, M., *Pers. Comm.*). When sprayed EVA forms a candyfloss like netting but with fewer fibres. This has been tried and tested, significantly reducing egg laying and maggot infestation on onion and cabbage plants. The product gave up to an 85% reduction in infestation levels. This should ultimately lead to a reduction in damage by the larvae. Currently this product is too costly to produce for large-scale trials, but with future developments this barrier may be overcome. In addition, it is possible to formulate repellents into the barrier and perhaps increase protection (Hoffman, 2002). This product has not been tried for *H. abietis*, but may well be the sort of easily applied barrier that is sought after.

1.6 Biological control

In general, population control of *H. abietis* has not been extensively tested. This is obvious from the general lack of literature on the subject. Some biological control agents have been investigated for their suitability in the control of the Large Pine Weevil.

1.6.1 Fungi

One of the few references to the use of an entomopathogenic fungus is that of *Beauveria bassiana* Bals. (Fungi: Imperfecti) by Wegensteiner and Fuhrer (1988). The fungus appeared to cause 100% mortality in the weevils at the highest concentration at high humidity after 1-2 weeks. However, effective doses were in the range of 1-10 million conidia per insect. This is exceptionally high as typical locust applications of formulated products are around 100 conidia per insect (Bateman *et al.*, 1998). The fungus is recognisable in insects when the mycelia burst out of the insect cuticle and form white cotton wool like clusters (Plate 1.5).

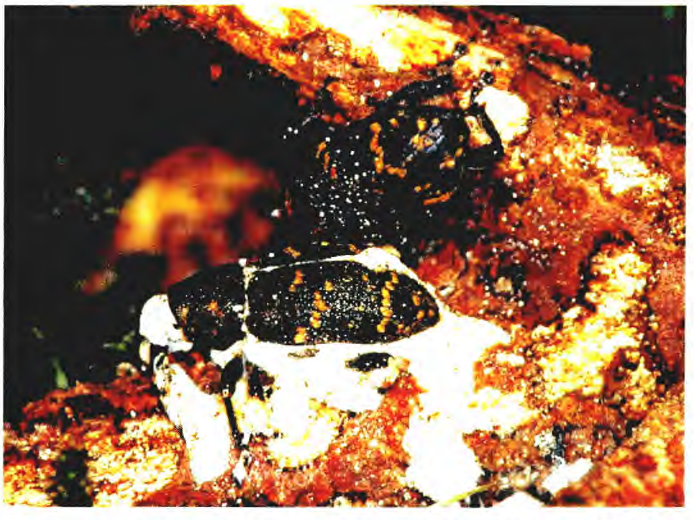


Plate 1.5 ***Hylobius abietis*** **infected with and killed by** ***Beauveria bassiana*** **entomopathogenic fungi.**

In addition to *B. bassiana*, there is also a larval specific entomopathogenic fungus, *Sporotrichum globuliferum* (Brixey, 1997). There are also several unidentified yeast and bacteria that can cause mortality in larvae, but whether these are specific to *H. abietis* is unknown.

1.6.2 Parasitoids and other microorganisms

There are several parasitoids and diseases that attack the two key life stages of *H. abietis*. The first, *Nosema hylobii,* N-SP (Nosematidae, Microsporidia) is a disease of the adult life stages of *H. abietis*. This is a protozoan parasite that invades the gut epithelial cells (Purrini, 1981). Along with two other species, *Gregarina hylobii* and *Ophhryocystis hylobii,* it causes a microbial infection resulting in the death of the weevil. No external symptoms were evident, although there was decreased activity and cessation of feeding (Purrini, 1981). Only the biology of the disease was looked at, not the efficacy as a population control method. Unfortunately, many of these diseases are only effective at high population densities when the disease can be easily transmitted (Matthews G.A., *Pers. Comm.*).

In total there are three species of parasitoid wasps that attack *H. abietis* and are indigenous to the UK; *Perilitus areolaris* Gerdin & Hedqvist, *Dolichomitus tuberculatus* (Geoffroy) and *Bracon hylobii* Ratzeburg (Henry & Day, 2000). *P. areolaris* (Hymenoptera, Braconidae) is a parasitoid wasp that preys on adult *H. abietis* (Stary and Soldan 1988). Studies of parasitoid wasps of *H. abietis* are mentioned as early as 1922 (Wulkner, 1922) and 1969 (Slysinski, 1969). There have been two closely related species mentioned frequently in the parasitoid literature, *P. areolaris* and *P. rutilus* that have probably been confused in their identification quite often (Stary and Soldan 1988). Neither of these two species are host specific, *P. areolaris* only parasitizes the Hylobiine family of weevils, whilst *P. rutilus* parasitises the Hylobiinae and the Brachyderinae families.

It has been shown that *P. areolaris* attacks the free-living adults, whilst another species, *B. hylobii*, parasitizes the buried larvae under the bark. *P. areolaris* is a polyembryonic parasitoid. Several larvae develop from a single egg inside the host, the larvae then migrate to the surface and form whitish cocoons outside the host individual (Stary and Soldan 1988). It is distributed across Sweden, Czechoslovakia, Poland and Western Germany.

There have been several hypotheses put forward for the apparent lack of parasitoids of *H. abietis*, one being that the relatively thick cuticle of *H. abietis* prevents parasitoids from penetrating their exoskeleton and also provides some protection from predators (Gerdin & Herqvist, 1984). This is an adaptation that all the Curculonidae possess. In favour of this theory, *Perilitus sp.* only parasitize Curculonidae species and a few other species that have similarly thick exoskeletons (Stary and Soldan 1988). This suggests that a co-evolutionary arms race may be taking place. Alternatively it is possible that parasitoids of *H. abietis* have frequently been overlooked. Firstly the parasitoid larvae emerge from the host and drop to the ground to pupate. Therefore, the parasitoids are only rarely seen in association with the weevil host. Secondly, adult *H. abietis* do not show any symptoms of infection other than cessation of feeding (Stary and Soldan 1988). There are other parasitoids that have been classified more recently, such as the ichneumonid wasp, *D. tuberculatus* that is again a parasitoid of adults (Stary and Soldan 1988).

The use of the parasitoid *B. hylobii* has been investigated within the Forestry Commission (Brixey, 1997). *B. hylobii* is a parasitic wasp specific to *H. abietis* that has been experimentally mass released but with little success. The Forestry Commission has mapped the level of parasitism that occurs naturally. It was shown that levels of parasitism ranged from 14.3% immediately after clear felling, up to 70% three years later. Unfortunately, the low initial dose rate allowed many of the larvae to escape parasitism and successfully emerge. At the end of the third year an average of 47% of larvae had been parasitized, but this was insufficient to alter feeding damage by the emerging adults. It is possible that supplementing the numbers may achieve better results (Brixey, 1997).

The potential of *B. hylobii* as a biological control agent has been further investigated and some of the reproductive behaviours also examined (Henry & Day, 2000). The reproductive strategy of a parasitoid can affect its success as a biological control agent and the allocation of reproductive resources is a major aspect of any strategy. This study identified that *B. hylobii* prefers larger *H. abietis* hosts and will deposit the greatest number of eggs into the first host found. The parasitoid then becomes increasingly choosy as the egg load decreases. The optimum temperature for reproduction was established, which lies between 10 °C and 20 °C. There was increased fecundity at higher temperatures, but reduced generation time and life expectancy (Henry & Day, 2000). However, an increase in fecundity may not necessarily be beneficial to the parasitoid larvae as they suffer increased competition from siblings and may themselves be smaller with lower fecundity (Waage & Ming, 1984).

Bracon hylobii were shown to reject *H. abietis* larvae less than two months old, but a typical larval stage will last for one to two years and they are therefore susceptible to parasitization for a substantial period of time (Henry & Day, 2000). Provision of additional food sources for the adult parasitoid can increase fecundity and longevity. This can be done by promoting nectar-producing plants or by deploying synthetic foods (Henry & Day, 2000).

1.6.3 Nematodes

There are two species of entomopathogenic nematodes that parasitize *H. abietis, Steinernema sp.* and *Dirhabdilaimus leukartis*. The use of entomopathogenic nematodes in forestry is subject to far fewer regulations than pesticides or entomopathogenic fungi, which makes them particularly attractive to the under-funded Forestry Commission. Indigenous species may even avoid the Wildlife and Countryside Act (1981) allowing very easy use (Brixey, 1997).

Entomopathogenic nematodes are able to locate larvae buried deep within the stump and infect them. Nematodes penetrate through the host cuticle or body openings. Death is actually caused by septicaemia, as bacteria carried by the nematode are toxic to the larva. Death can take as little as 48 hours. The bacteria rapidly multiply and the nematodes feed on the bacteria and degraded host tissues. The nematodes multiply and use up the remaining resources (host tissues), before around 10,000 juvenile nematodes burst fourth and go in search of their next victim (Brixey, 1997).

Extensive field trials by the Forestry Commission found that despite the efficacy in killing larvae, the nematodes did not infect enough larvae to reduce the population below the economic injury threshold. This was not through lack of success by the nematodes, but because there were so many more weevils than saplings, the population would need to be suppressed to an incredibly low level to achieve damage reduction. At this point the Forestry Commission decided to abandon the idea of substituting nematodes for pesticides (Brixey, 1997). However, nematodes are cheap and effective enough to form part of an Integrated Pest Management (IPM) program. Recently, the Forestry Commission has reviewed their nematode program and it has now developed a system that can separate the nematodes from their food media, which allows them to be concentrated into small volumes of water, making the application process much simpler and cheaper. Large-scale trials are currently underway (Heritage S.G., *Pers. Comm.*)

The use of entomopathogenic nematodes has been demonstrated with related species, such as *H. congener* (Eidt *et al.*, 1995). Damage done to seedlings was reduced to acceptable levels when the nematode *Steinernema carpocapsae* was applied to the roots of seedlings before planting. *H. congener* adults then became infected whilst feeding. This work was spurred by the initially encouraging results of work with the nematode and *H. abietis*. In a field trial the nematodes provided very good protection, reducing the damage below the economic injury threshold, which in this system prevented the need for replanting or topping up the restock site. Eidt *et al.* (1995) end with recommendations that this method be tested for *H. abietis*.

Population control of *H. abietis* with biological control agents has to date proved rather unsuccessful. There is huge potential for an integrated program that uses different pathogens to target each of the life cycles. Although no one agent is successful alone, it may be that they work synergistically and would greatly reduce the population level. Further work in this area is definitely needed, in particular research into the simultaneous use of several pathogens. Biological agents offer many advantages. In particular they have less environmental associated risk than pesticides, particularly higher organism agents when it is clear they do not harm animals or other wildlife (Dent, 1993). However, care should be taken when introducing exotic species. It may be that the first step in control is to use a combined pesticide and bio-control strategy forming an IPM program so that numbers of weevils can first be reduced before prophylactic treatment is stopped.

1.7 Silvicultural control

Hylobius abietis has been a pest since pre-Forestry Commission times and certainly before the invention and reliance on chemical control methods (Eidmann, 1985; Munro, 1928). Consequently, forest managers long ago developed methods that reduced the incidence of pest population outbreaks. In agriculture such practices are known as cultural control, however in forestry they are termed silvicultural control (Eidmann, 1985). Natural populations of *H. abietis* normally rely on storm-felled trees and death in older trees to expose stumps. Therefore, the ability to disperse over several miles and detect fallen trees is of a high importance. Modern forestry practices have created an artificial situation that encourages populations to build up to unnatural levels and cause widespread damage (Leather *et al.*, 1999).

By tailoring practices to resemble a more natural situation, mass population increases can be avoided. Foremost of these is the removal of breeding material. Colonisation of stumps is greater in clear-cut stands than in thinned areas (Eidmann, 1985). Presumably there is a lesser amount of host volatiles causing less immigration. Therefore, staggered felling in one site could result in lessened damage. Similarly removal of slash (the discarded side branches of felled trees), which is routinely raked into piles, will also reduce available breeding sites.

A common practice adopted by many foresters, is the 'wait and plant' method. This is based on the typical two year life cycle of *H. abietis* and therefore saplings are planted after three years (Orlander & Nilsson, 1999; Scott & King, 1974). This method has several drawbacks; firstly, three years of growth time is wasted resulting in an economic loss. Secondly, there is usually still a substantial population in the third year resulting in high levels of damage (Orlander *et al.*, 1997; von Sydow, 1997). There is also the additional competition with weeds and other ground vegetation (Orlander *et al.*, 1996) and possibly increased deer damage (von Sydow, 1997).

Manipulation of a clear fell site can alter *H. abietis* damage, in particular reducing the size or changing the aspect of the site. However, these practices are usually outside the ability of the forest manager to alter because of other more pressing demands.

Alternatively, shelter-wood, i.e. leaving a small proportion (150 of 2000 per hectare) of the mature stock unfelled has been investigated. The use of shelter-wood has been shown to reduce *H. abietis* feeding damage on planted conifers (von Sydow & Orlander, 1994). However, the mechanisms by which this occurs are largely not understood. It has been proposed that the trees may create a microclimate, affect ground vegetation or provide an alternative food source (Orlander *et al.*, 2000). The most likely explanation is that the trees provide additional food for *H. abietis* and therefore deflect the damage from planted saplings. Orlander *et al.* (2000) showed that this was not the case and that the damage done to the mature trees would not be enough to satiate *H. abietis*. Of all the silvicultural techniques, shelter-wood use is one of the more promising, but first it must be fully understood so that it can be properly implemented.

There are two other unrelated techniques that affect the alternative food sources of *H. abietis*. The removal of slash has been suggested as a method of reducing local populations by decreasing the available food supply. In addition, decaying slash liberates nitrogen, which in turn is used by the growing conifers. Selander & Immonen (1991) demonstrated that increased nitrogen levels of Scots pine leads to increased feeding damage, therefore, this may be another mechanism behind apparent decrease in damage due to slash removal.

The abundance of *H. abietis* has been studied in relation to a range of environmental factors (Orlander & Nilsson, 1999; Orlander *et al.*, 1997). In both of these studies the removal of slash provided mixed results. There was some increase in trap catches in the earlier study, but this was not related to seedling damage. It was hypothesised that this was due to increased mobility of the weevil. The later study showed a decrease in mortality of planted saplings. Unfortunately, the effects are often minimal and easily surpassed by other factors, whilst the work involved to remove slash can significantly increase planting costs.

Intercropping, or mixed planting as it is termed in forestry, may lead to decreased damage by *H. abietis*. Leather *et al.* (1994) showed that *H. abietis* exhibits a strong preference for Scots pine, but may also feed on other species, whilst the presence of some species (such as *Fraxinus excelsior* L., ash) can inhibit feeding. Other studies have shown similar results (Manlove *et al.*, 1997). Mixed planting frequently occurs with Birch (*Betula pendula* Roth.), but this is for both control and for public amenities (Morgan, 1999).

Recently, trials with neem have been carried out and showed that neem extract effectively reduced and prevented damage for several weeks, compared to the controls (Thacker *et al.*, 2002 Unpublished). Neem treated trees lost only 20% of their bark. However, it was not made clear whether there was any evidence of girdling occurring. The most effective protection was given by neat extract (100%), but this could possibly be quite costly for large-scale operations.

It is interesting to note that in clear fells with natural regeneration, the very young seedlings are particularly numerous and a large proportion escape predation and are too well established to suffer damage from *H. abietis* when they are at an attractive age. Contrary to this, planted saplings often suffer enormous levels of damage unless they are treated (Eidmann, 1985). Nystrand & Granstrom (2000) have since taken this further, listing all of the key predators of seedlings. They found that pitfall trap catches of *H. abietis* were not related to seedling predation. They stated that *H. abietis* did not appear to cause any significant damage to seedlings, but in fact a species of slug (*Arion subfuscus* Drap., Gastropoda: Pulmonata) was the key herbivore. However, *H. abietis* catches in pitfall traps are often an unreliable estimate of the population number and a poor indicator of potential damage that may be caused (Heritage S.G., *Pers. Comm.*).

Heritage and Moore (2001) summarised the Forestry Commission (UK) silvicultural practices that can affect the level of damage by *H. abietis* (Table 1.1). This information is used within the Forestry Commission to recommend the best practice for forest managers under a range of given conditions.

Table 1.1 **Site Characteristics that can affect damage by *H. abietis*. Taken from Heritage and Moore (2001).**

Previous crop	
Species	**Risk Level**
Lodgepole pine	Very High Risk
Other conifers	High Risk
Only broadleaves	Low Risk
Quality of crop before felling	**Effect on Damage**
Large quantities of conifer stumps or dead wood 0–6 yrs prior to felling	Increase
Large quantities of conifer stumps or dead wood 7+ yrs prior to felling	Neutral
Small quantities of conifer stumps or dead wood	Neutral
Felling	
Time of felling	
Felled between January and May	Neutral
Felled between June and December	Delay
Felling system	
Clear-felling total area larger than 1ha	Increase
Clear-felling total area smaller than 1ha	Neutral
Standing mature trees on site (i.e. strip felling, continuous cover or seed trees)	Decrease
Site location	
Isolation from other felling sites	
Less than 5 km from 0–4 year old conifer clear-fell	Increase
Between 5–11 km from 0–4 year old conifer clear-fell	Neutral
At least 11 km to nearest 0–4 year old conifer clear-fell	Decrease
Site preparation	
Stump treatments	
Stump removal decrease Felled after July and de-stumped before following May	Decrease
Stumps removed all other times	Neutral
Delayed planting (fallow periods)	
Planting 0–4 years after felling	Increase
Planting 5–6 years after felling	Neutral
Planting 7 or more years after felling	Decrease
Ground preparation	
Needle litter left undisturbed Exposed by mounding or screefing	Neutral
Brash treatments	
Brash raking	Neutral
Controlled burning of brash	Increase
Vegetation and weed control	
Woody plants surrounding planted stock No herbicide or spot treatments	Decrease
Woody plants surrounding planted stock treated with herbicide	Increase
Dense grass growth with or without herbicide treatment	Increase
Plant specification	
Plant species	
All conifer species	Increase
All broadleaf species	Neutral
Plant size	
Small plants	Increase
Large plants	Decrease

The age and size of the sapling can greatly affect *H. abietis* damage, with older larger saplings being better able to resist feeding damage (Selander, 1993). Additionally, adjustment of the planting time so that it does not coincide with the first weevil emergence can significantly reduce damage (Eidmann, 1985). Unfortunately these methods frequently reduce plant vigour or survival prospects because older saplings are less tolerant of transplanting. In addition the planting of older larger saplings can dramatically increase the costs of the planting operation (Heritage, S.G., *Pers. Comm.*).

Increased plant vigour can reduce damage, or increase the sapling's tolerance of damage and this can be brought about by scarification. This is the process of exposing mineral soil and removing the upper layer of humus (Selander, 1993). Several authors have found that scarification greatly reduces damage and mortality of saplings (Orlander & Nilsson, 1999; von Sydow, 1997), but this effect diminishes with age (von Sydow, 1997) and is seldom sufficient as the only method of control (Hannerz *et al.*, 2002). Additionally, accidental mixing of the humus with the mineral soil can nullify the reduction in damage and mortality (Orlander & Nilsson, 1999). There are several methods of scarification; patch scarification, disc trenching and mounding. Of these, mounding is the most effective as it has a more dramatic effect on soil temperature and decreases feeding damage the most. The causes of this reduction in damage are also not understood, but likely reasons are an increase in plant vigour and a reduction in activity of *H. abietis* on warm dry soil (Orlander & Nilsson, 1999).

Kindvall *et al.* (2000) videoed the movement of *H. abietis* on sandy mineral soil and humus. The weevils moved significantly faster over sandy soil than in the humus but did not turn back when moving into a sandy area. They concluded that *H. abietis* might find a sandy soil more hostile and less sheltered and therefore move faster on sand. The study did not appear to address the issue of the different texture of the two soil types and whether this was a physical reason for difference in speed, although both soils were flattened and equally moistened.

The range of factors that may affect *H. abietis* damage is extensive. Two of the most easily manipulated are scarification and the use of shelter-wood, both of which significantly reduce pine weevil damage. These systems provide a strong base for an

IPM program that could ultimately lead to the reduction in pesticide use. However, at present many countries are not in a position to withdraw pesticides unless faced with a ban on their use.

1.8 Aims and Objectives of the Project

Many of the current Forestry Commission methods of control face an uncertain future. In particular, the dipping system will soon be banned and the registration of permethrin will not be supported and therefore, it too will be phased out. This leaves only the Electrodyn system for treatment, for which the custom oil based formulation requires extensive registration and therefore the system can only slowly adapt to changing regulations. Currently, the Forestry Commission is not in a position to avoid the use of pesticides and replace them with silvicultural and biological control. Therefore, there is urgent need for a replacement treatment system.

The initial aim of this work is to design, build and evaluate an automated conveyor belt system for the treatment of young conifer saplings, which are to be transplanted into a clear-fell site. There is a range of criteria that are important to the design of such a system and these are carefully discussed in Chapter 2.

The second aspect of this work is to gain a thorough understanding of the mechanisms by which control is achieved with pesticides. Prophylactic treatment is comparatively rare in agriculture because of the unnecessary use of pesticides. However, in this system, prophylactic control actually minimises the environmental impact of the pesticides. It is believed that prophylactic use of pesticides by treating a food source may not result in knock down as may occur in normal systems. In addition, there is some speculative evidence to suggest that the pesticide does not kill the weevils. This section of work will aim to understand this system and draw conclusions with respect to a pest management system.

Chapter 2
The design and evaluation of an automated treatment unit to apply pesticides in low volumes to coniferous saplings

2.1 Introduction

This chapter details the construction of an automated treatment unit. There were several key aims intended for this system:

1. To build a prototype system that could apply a small set dose of pesticide to a specific area of a two-year-old conifer sapling, which will offer prophylactic protection for up to one year.
2. This system should have an advantage over other systems in that it will use "off-the-shelf" formulations of pesticide. Therefore, no special formulations will be required and approval for the system will be confined to an "off-label" approval. This will significantly reduce the cost in installing the system into current practice.
3. The system will maximise the percentage of pesticide sprayed onto the stem and reduce wastage by minimising pesticide that hits the foliage or misses the tree. The overall pesticide usage will be considerably less than the current dipping system.
4. The system will consider safety to be of the utmost importance. It will be completely self-contained and reduce potential operator exposure compared to the dipping system.

These key aims were established at the start of the project in the design brief. These aims are discussed in this chapter, whilst development of the system based on experimental work is discussed in Chapter 4. Certain sections contain experimental data to support the development of the system.

The following sections deal with the key components in the design of this system; the conveyor, nozzle application; framework and pump components.

2.2 The Dandrive Conveyor Sprayer system (DCS)

The design and construction of a conveyor belt system, such as the one required here, is a gradual process. Initial designs have to be taken further and developed both on paper and in the workshop. Development occurs throughout the entire design and construction process, not just in the early sketches on paper. In the construction of this system, it was found that it was necessary to completely redesign entire sections. Usually this was because the materials and parts available were not suitable for the initial design. At the start of the design stage, I was aware of such potential problems and the early design was intended to be as versatile as possible. One such aspect was the framework itself that held the conveyor and spray application equipment. This was built from Dexion® (right angle metal flats, commonly used for shelving) which could be pulled apart and put back together as many times as is necessary, allowing an infinite number of adjustments.

There are four key components in the design of this system that had to be designed separately and then integrated successfully at the end:

- The conveyor system – to hold and transport saplings
- The nozzle application system – to spray the saplings as they pass
- The framework and panelling – to support and shield
- The pesticide storage and pump system

In the following section, each of these four areas is discussed with the key criteria set out at the beginning. The following chapter examines aspects of safety in a range of tests. These were not intended to be definitive, but to bring various aspects of the systems use into context. In addition, a full range of technical drawings and design sketches can be found in Appendix 1.

2.2.1 The conveyor system

Key Criteria The conveyor belts must transport saplings measuring 20-40cm in height, past a series of nozzles. The saplings must be correctly held in place at all times so that they pick up the maximum proportion of the intended dose of insecticide. The conveyor belts must not damage the roots. In conjunction with the spray application equipment, they must prevent the roots from getting wet. The conveyor belts must not pose any risk to human operators.

The Electrodyn system (Plate 2.1) currently uses a flat conveyor belt composed of several cords, somewhat similar to that of a supermarket conveyor belt. These cords allow the pesticide to be sprayed onto both sides of the plant, with the plant laying flat in a horizontal position. This is only possible because the Electrodyn has a unique ability to electro-statically charge droplets, which are then capable of flying upwards against gravity (Matthews, 2000). After examining this system, it was decided that it would be less suitable for a pair of hydraulic nozzles, which would need to be positioned above and below the tree. There would be potential changes in spray cone characteristics when a nozzle is aimed straight up and these could be detrimental to the working of the system. In addition, soil from the saplings could fall onto the nozzles and block them.

Plate 2.1 **The Electrodyn conveyor sprayer, clearly showing the supermarket style bed and cord type belts.**

Early studies (Appendix 2) and unpublished reports from the Forestry Commission indicated that the prototype system would need to present the lower 15cm of stem to the nozzles without blocking the spray. Therefore, the trees would need to be hung vertically and sprayed from the sides. By hanging the trees upside down, the grappling mechanism could hold onto the roots and not the foliage and thus compromise treatment. A range of systems was investigated involving the use of various claws, hooks and chains to hold the plant. Most of these systems required saplings to be put into pre-set positions which may be difficult for the operator to do accurately time after time. In addition, they would not be particularly gentle in their handling of the saplings. After further design work it was envisioned that a pair of conveyor belts in a vertical plane turning in opposite directions would be capable of moving a sapling forwards in the central channel, as shown in Figure 2.1.

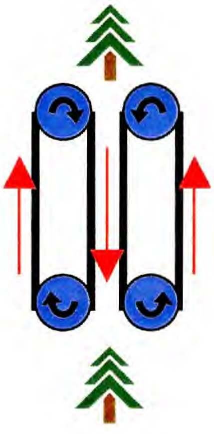


Figure 2.1 Initial conveyor belt design.

The early concept involved the use of two parallel belts, which would move counter clockwise and clockwise. This moves the central canal in one direction. Small saplings can be supported in this gap by friction alone, thus being gently moved past the spray application equipment.

The effects of gravity and friction should hold the trees upside down in a stable position. However, trees held the correct way up may be less stable and more likely to fall over. This design was followed through to the production stage and the prototype was built, as discussed below.

The two belts used on the system are approximately 0.8cm thick, 260cm long and 10cm high, set 3cm apart. This completely encases the roots, preventing any loose roots from being trapped in the pulley system. The original conveyor belt design formed a kite shape, (Figure 2.2). However, compared to the simpler proposed layout (Figure 2.1, above), this design used several more pullies and a substantially longer timing belt and would cost significantly more. The kite pattern has the advantage that the trees would be dropped to the ground in the centre of the conveyor. This keeps the roots and the sapling well away from the electric motor and final set of pulleys, thus ensuring that they do not get damaged or jammed in the machinery. Despite the advantages, it was decided to use the cheaper option, at least for the prototype.

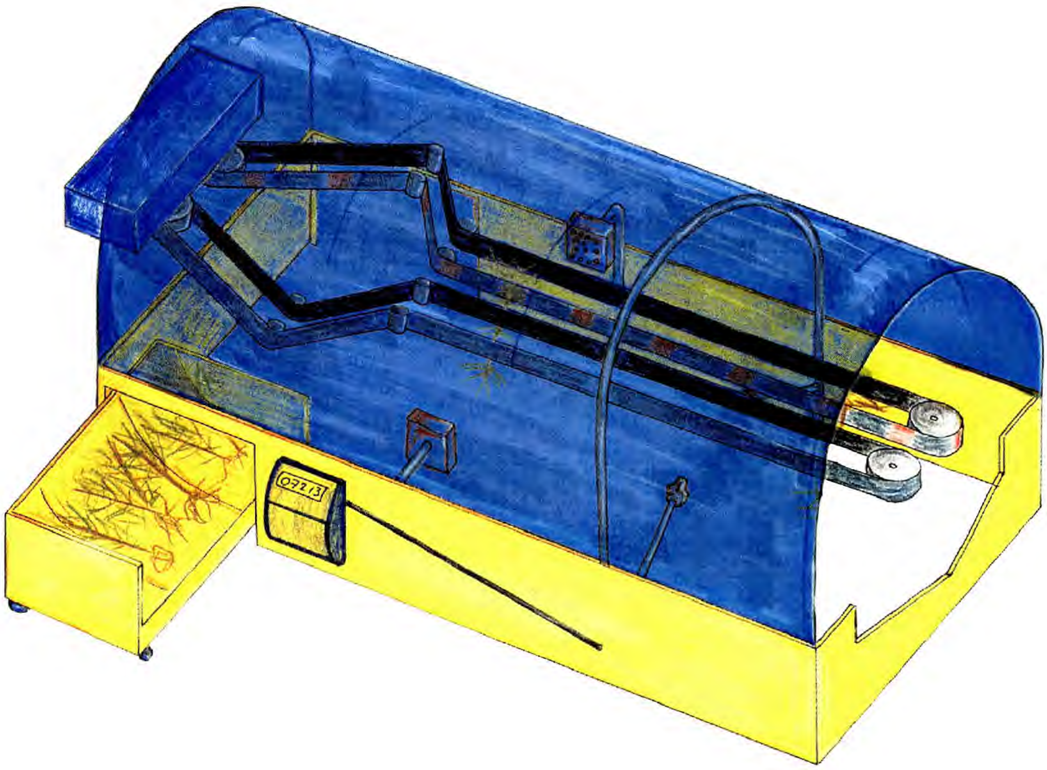


Figure 2.2 **The Dandrive Sprayer Conveyor (DCS). The system will treat pine tree saplings with pesticides, protecting them against weevil damage. It aims to replace the systems that are currently being used. The black conveyor belts can be seen running the length of the system, they form a kite shape, which allows the saplings to fall out of their grip in the centre of the conveyor, clear of the machinery (in the dark blue box).**

The conveyor belt consists of 6 timing pulleys that are toothed and will grip onto a timing belt without slipping (Plate 2.2). The timing pulleys are 10cm high with a diameter of 13cm. These dimensions were chosen as they were available in pre-made sizes and are best suited to the timing belt. Each of the timing belts is 2.6m long, which will transport the sapling about 1.2m (allowing for the circumference of the pulley). The pulleys are mounted on stainless steel shafts, which are supported by pillow block bearings. These bearings, as well as rotating along a radial axis, can be set to support the shaft at angles other than perpendicular to the horizon. This allows for a greater margin of error in the engineering.

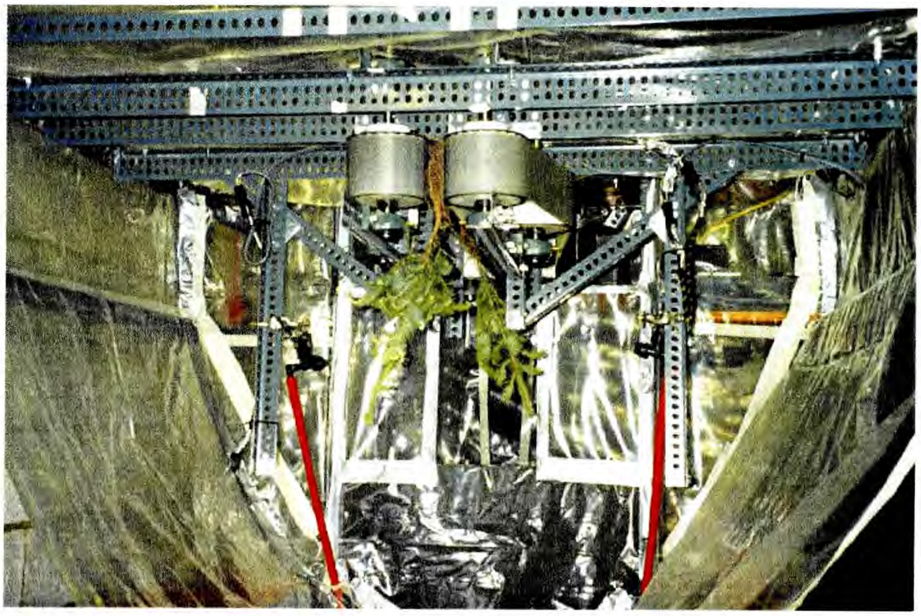


Plate 2.2 **A view of the pulleys and timing belts trapping Sitka spruce saplings. This photograph does not show the EPDM sponge that was later added in.**

The system is driven by a 0.5 hp electric motor that although humidity proof, is mounted above the conveyor belt so that it is removed and protected from spray drift (Plate 2.3). The motor is connected to two crossed helical axis gearboxes, one left handed and one right handed, creating the opposite rotations seen in the belts. The gearboxes were chosen because they change the plane of drive by 90° and they are cheaper than the heavier duty bevel type gearboxes. The gearboxes are mounted on to aluminium mounts, which have been milled so that the face of the gearbox sits snugly on top. It was important that these mounts held the central axis of the gear boxes at exactly the same height as that of the electric motor so that the drive shaft would be exactly horizontal. If this was not achieved, the life of the gearboxes and electric motor may be substantially reduced as the shaft is forced to rotate at an awkward angle. It was later decided that in subsequent modifications to the Dandrive, a universal coupling should be positioned between the motor and the gears to absorb any of the vibration produced by misalignment.

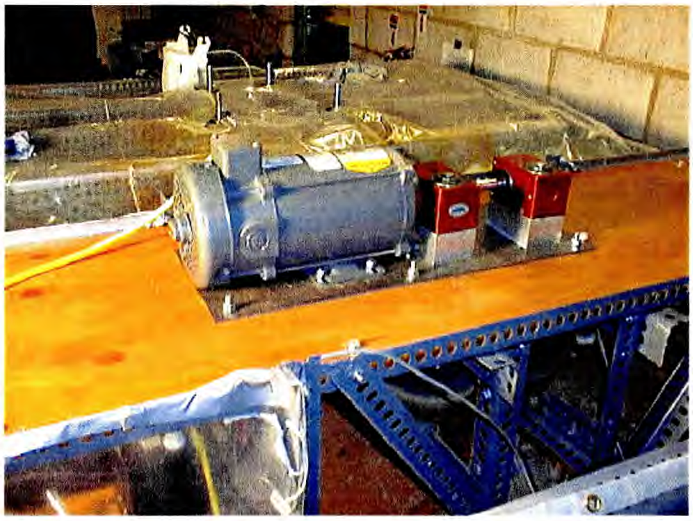


Plate 2.3 **The electric motor and gearboxes (supported by aluminium mounts), which sits above the pulleys and spray apparatus. This arrangement prevents the risk of any spray dripping into the motor and will have a lower humidity due to better air movement, therefore reducing the risk of rust.**

The gearboxes are of the one-third-reduction type gearbox, which reduces the speed of the engine by a third (Plate 2.3). The intended operating speed of the conveyor is between 12-20m min^{-1} and so the timing pulleys are required to turn at an average speed of 500rpm, therefore the motor would operate at 1500rpm. After production it was noted that the motor was running at about 20% capacity and the conveyor operated as expected. It was believed that the conveyor could operate at speeds as fast as 25m min^{-1}. At this speed the saplings would traverse the conveyor's length in just over 3 seconds, compared to 4.4 seconds at the intended operating speed of $20mmin^{-1}$.

Other than speed, the force at which the electric motor turns the conveyor belt is also of importance. If the operator should happen to get their arm stuck between the belts, they could quite easily become stuck as the conveyor pulls it with such force. This force can be measured and is known as torque. One mechanism for preventing such injuries would be for the system to be able to distinguish between a human arm and small saplings. Saplings only weigh a few grams and will provide next to no resistance, yet the human arm is attached to the human body, which weighs 60-80kg and will provide considerable resistance. In industry a slipping clutch is often used. This is a type of coupling that when a certain resistance is exceeded (torque) it spins freely disengaging the drive from the rest of the machinery. This prevents damage to the machinery if some of the parts become blocked or jammed.

This system could be applied to the conveyor belt system if it was decided that the twin conveyors pose considerable risk to a human operator. To implement this, the torque on the system needs to be calculated so that the correct size can be ordered. This system can also be connected to the electric motor, causing it to shut down once the coupling has disengaged and even sound an alarm alerting others of possible accidents.

The conveyor belts are made of tough plastic polyurethane, with steel tensioning members. The timing pulleys, after consideration of the weight, were ordered in Delrin®, a tough plastic approximately one-sixth the weight of steel. The distance between the two belts had to be carefully decided, maximising grip and minimising damage to the sapling. On testing, it was found that the saplings roots had some resistance in them to being squashed, therefore, if the gap between the belts was correct most trees would be held between the belts without falling. It was found that there was large diversity in the root structure of different saplings and many of the smaller saplings could not be held, whilst others were only held if very carefully positioned. This would be a significant drawback to the speed and efficiency with which the system could be used. By adding a layer of sponge or other similar material the gap between the belts could be reduced, the grip would be increased and there would be some give in the material to prevent crushing of the saplings.

Several materials were tested, namely polyester, LD45 (a low density sponge, like the material used in wrist rests for keyboards) and EPDM (an expanded polymer sponge). EPDM was found to have both the softer qualities of polyester and the water fastness and durability of the LD45. In addition, it has excellent acid and alkali resistance, but may be degraded by chlorinated solvents. This gave a good balance between gentle handling of the plants and a long operating life. Samples of different thicknesses were obtained and tested and the 4mm thick size was found to be the most suitable. The EPDM was therefore obtained in rolls 10cm wide, 4mm thick and 200cm long. The EPDM is backed with a peelable adhesive allowing it to be easily removed after each pesticide trial and disposed of. The EPDM protected conveyor belts can be seen in Plate 2.4

Plate 2.4 **EPDM (expanded sponge) is used on the surface of the timing belts to minimise slippage of the smaller saplings whilst still being exceptionally durable and waterproof.**

After continual use the conveyor belts and sponge could become wet enough to pose a hazard to the operator. If this happens, then absorbent sponge rollers or plastic 'windscreen wiper type blades' can be fitted so that the pesticide is wiped off, maintaining a dry surface at the end where the saplings are fed in. A second alternative would be the use of automated tree feeders, removing the operator further from the risk of exposure and also maximising tree loading speed.

2.2.2 Recent developments to the system since the initial construction

This section and similar sections at the end of other design units, detail improvements that have been made and could be made on future models. Ideas for these improvements were largely generated from the extended use of the DCS system during the field trial. Many helpful comments were provided by the Forestry Commission staff who used the DCS system during the trial.

Several restrictions to the design of the system have been encountered due to the imposed financial budget. It is for this reason that some aspects of the system were inadequate during the field trial. One such aspect was the conveyor belts. It was decided that extending the length of the conveyor belts by at least 2-3m, the operators could be further distanced from the spray and resulting drift. In addition, the belts should also be much deeper, the current 10cm depth was acceptable for many of the smaller saplings, but there were a significant number of trees that had very long roots. These were difficult to feed in to the conveyor belts without exposing some of the roots to the spray. Increasing the depth of the belts to 20cm would be sufficient. This will also prevent the roots from becoming tangled at the exit (Plate 2.5). However, the early kite design may also prevent tangling and be a more effective way of ensuring that the roots do not get caught up.

Plate 2.5 **Many of the plant roots become tangled at the exit of the conveyor belts, which quickly causes pile ups. In some cases the trees may be dragged back along the conveyor into the spray zone.**

The EPDM on the conveyor belts worked well at trapping the majority of saplings. Unfortunately, there were two small groups of trees that still caused problems. Firstly, very small saplings with few roots still fell out of the belts. Secondly, some exceptionally large saplings (some were over 70cm tall) had very large root masses, whilst others had huge clumps of mud on the root mass. These trees became trapped at the exit forcing the conveyor to jam. One way of preventing both of these failures would be to use a double sponge layer. A very thick layer (approximately 3cm) of polyester or similar material would line the belts. On top would be a 4mm layer of EPDM. This sponge sandwich would completely close the gap between the two belts, but instead of crushing the larger saplings, the very soft nature of the polyester would prevent this. In addition, the durability of the system would be maintained by enclosing the polyester with EPDM (Figure 2.3).

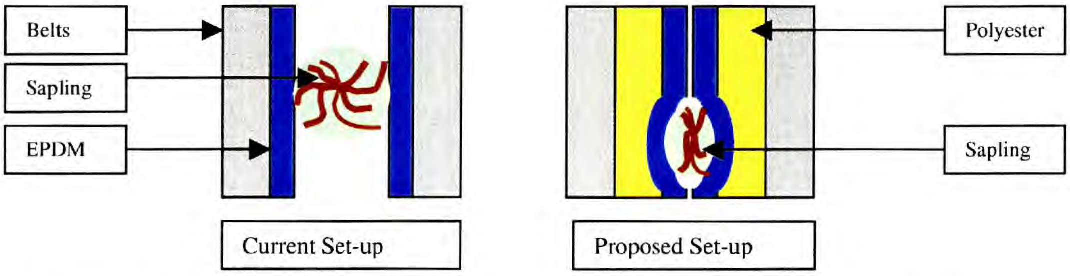


Figure 2.3 **Diagram illustrating the polyester/EPDM sandwich effectively trapping a Sitka spruce sapling.**

Originally it was decided that operators would find the exact placement of trees at regular intervals too difficult. However, it may be useful to indicate the rate at which trees should be placed in the system to maintain a daily output and to ensure adequate tree spacing. Red vertical bars could be included to indicate ideal tree spacing.

Finally, the entire conveyor belt frame should be raised, so that the workers do not have to squat whilst feeding in trees. Details of human ergonomics should be collected when deciding on the optimal height.

2.2.2 The pesticide application component

Key criteria This component must be capable of applying small doses of pesticide to the lower 15cm of stem on the sapling. The dose will range from 0.5ml to 4ml per nozzle. More importantly, the application component must be triggered to spray only when a sapling is in the spray area. This will reduce the volume of pesticide wasted. A third criterion is that the component must be very adjustable to allow for different nozzles and positions to be used.

Early in the development process, a system was found that met the above criteria. This was a solenoid nozzle apparatus that is capable of opening and closing the nozzles by electronic signal. It also has the added feature of pulsing, whereby the nozzles open and close rapidly many times each second (5-20hz) effectively reducing the volume of water sprayed (Giles, 1997). This facility can reduce the volume to 25% of the original value and more importantly it does not affect normal spray spectra (for example, droplet size, spray angle, flow rate). Unfortunately, commercially available control boards suitable for the DCS system were not available.

In collaboration with Dennis Wildman (Imperial College), two circuit boards were produced to give the application component a range of settings (Plate 2.6). Initially the nozzles were required to activate as a sapling approached the nozzles. Once triggered, the nozzles would spray for a fixed length of time and then switch off (referred to as spray time or duration). Spray times could range from 0.1–3 seconds. For the future prototype digital readouts would be preferable.

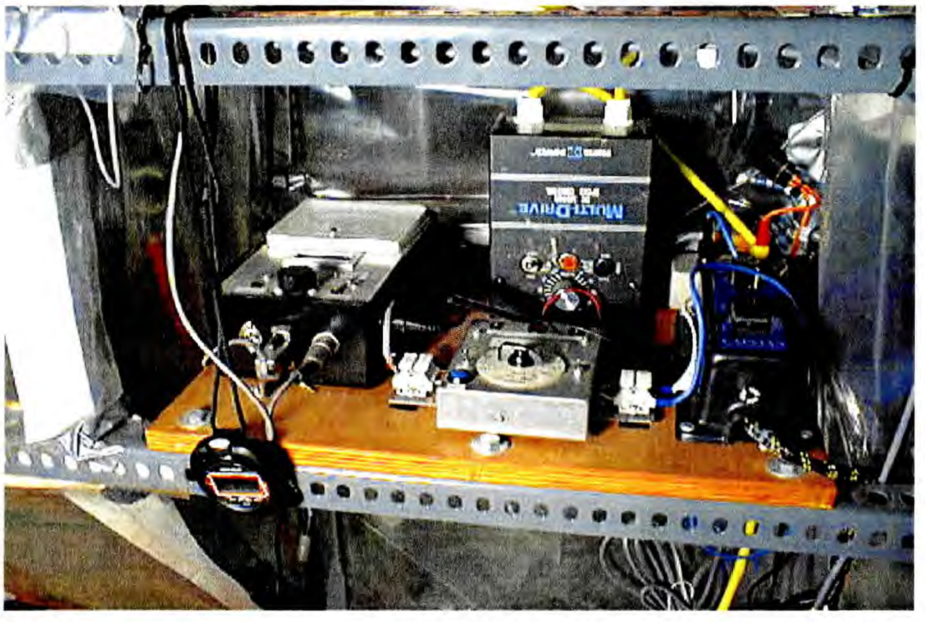


Plate 2.6 **The series of electronics that controls the DCS system. The two boxes in the front left control the timing of the spray components and were constructed by Dennis Wildman. The black box at the front right controls the pulsing of the nozzles and was purchased from Capstan AG systems, Inc. The box standing on end in the centre controls the speed of the conveyor belts and was purchased from Mick Jarvis, Kent Rewinds.**

The second circuit board controlled the trigger system. Initially an infrared detector was not used because of possible problems such as accurate triggering when a diffuse object such as a sapling moves past. Secondly, accumulation of dirt or water could prevent it from working correctly. Despite these reservations an infrared detector seemed preferable to other methods. Two other designs were investigated. A metal bar could be used, which closes a circuit when it is moved, thus triggering the nozzles. Unfortunately this only has a life of about 100,000 uses. Considering that the intended output is 36,000 trees a day, it would not last a week! The second option was for a magnetic switch to be used. Several magnets would be placed on the conveyor belt, which would close a circuit when passing near a receiver and trigger the nozzles. The drawback with this system is that the saplings have to be positioned in exact places next to the magnets. Although possible, it requires greater operator skill and the system could still be triggered even if trees are not present. Lastly, a laser could have been used, which has a much narrower beam than infra-red (which spreads to a diameter of 5cm over 50cm distance) and would be more accurate and consistent. However, lasers can cause damage to the retina and it would have to be carefully shielded so that it is not directed towards an operator.

After construction and early testing the infra-red detector proved to be quite reliable, successfully triggering the nozzles to fire for every sapling. Problems arose when individual stems were used, lacking the bushy foliage, resulting in some of the thinner twigs escaping detection. This is a result of the beam thickness being wider than the stem; therefore the beam is never actually blocked. During normal use, the thickness of the infrared beam should be narrow enough that all trees successfully trigger the system. Another problem arose with the second half of the system. To prevent the detector getting wet, it needs to be positioned away from the spray cone. This leaves a gap of several centimetres that the sapling will traverse through, not getting wet, whilst the nozzles are still spraying. A delay component was designed by Dennis Wildman that stopped the nozzles spraying for a set (adjustable) period after the detector was triggered. This was quite difficult to accurately calibrate due to the wide range of tree sizes. It was found that some trees arrived too late whilst others arrived too soon, which was in part due to different parts of the sapling, such as the stem, small or large branches or roots hanging down ahead of the sapling, triggering the detector. To compensate, the nozzles have to be set to spray slightly longer than required, i.e. starting and stopping before and after the sapling has entered or left the spray cone. With further refinement the trigger system can be improved and aimed accurately at the stem, reducing the volume of spray wasted.

In addition to the trigger and delay components, two other devices were added. The first was an electronic counter to record the number of times the nozzles spray. The second device was a speedometer to measure the speed of the conveyor belts. This is important for calibrating the volume of spray that is deposited as the saplings pass. Increasing the speed decreases the deposition. The speedometer uses a small flywheel that is turned by the conveyor belts, generating a current. This current is linked to an analogue display that is calibrated to display the speed in metres per minute.

One last feature of the spray application component is the infinite adjustability and ability to spray the same volume onto the plant with several different combinations of settings. For this reason, several tables were produced which list the different theoretical depositions for different conveyor speeds, pulse times and nozzle flow rates. These can be found in Appendix 3.

2.2.3 Recent developments to the spray application component since the development

Development of the spray application component is the focus of Chapter 4. A key area is the refinement of the targeting system, with the aim of increasing deposit and decreasing volume sprayed. Numerous different settings were tested ranging from nozzle type to spray pressure and conveyor belt speed. With regard to the development of the mechanics of the component, only two points are of relevance here.

During the field trial it was found that the infra red trigger was not as effective as early tests showed. After extended use dirt and mud started to obscure the sensor preventing some trees from being sprayed. The sensor was moved to a more shielded position a further 15cm away from the nozzles in an attempt to keep it cleaner and drier. In addition, the sensor was periodically cleaned to prevent the problem from manifesting. However, the continued problem of some trees tripping the sensor at the wrong time continued. This was remedied by setting the delay period to zero, but on the converse side this increased pesticide usage by 50%.

Future modifications to this component are required. Use of a compound sensor which would rely on signals from three infra red beams may prevent horizontal branches and loose roots from blocking the beam and activating the trigger. The three sensors would be positioned in a vertical line, so that only the tree would block all three beams simultaneously.

In chapter 4, different nozzle positions are investigated. It was found that positioning the nozzles in front of the oncoming tree (slightly offset at an angle) increased the reliability of the sensor (Plate 2.7). This was because the spray was directed at the oncoming tree, rather than perpendicular to it. Therefore a tree arriving at the spray cone slightly ahead or behind schedule would still be sprayed, the only difference being that the droplets travel slightly further.

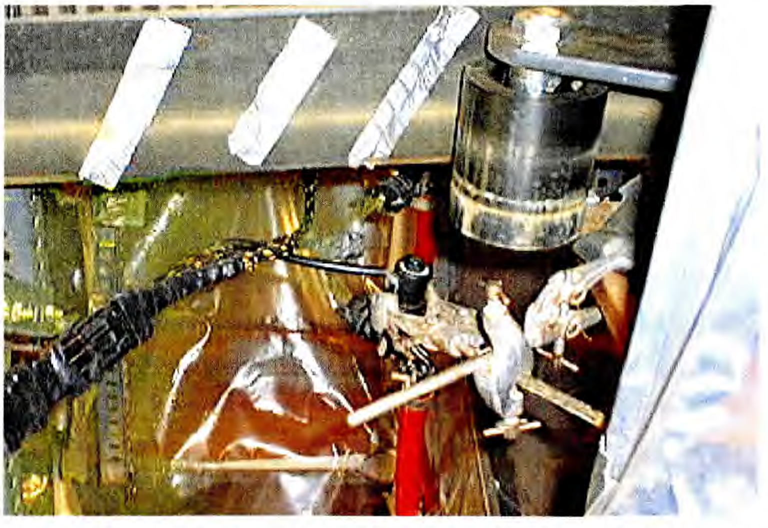


Plate 2.7 **Nozzles positioned in a 'Frontal' position so that they are aimed at the oncoming tree (as indicated by arrows), rather than perpendicular to it. This arrangement had a much great efficacy of deposit**

Finally, the tree counter and speedometer did not work as planned during the trial. The tree counter was activated every time the infra red beam was blocked and not every time the nozzles sprayed. This led to counts substantially above the actual number of trees used as an individual tree (roots and side branches) triggered the counter several times. This does not happen with the nozzles because the system has no 'memory' and will not start spraying again until the delay and spray period has expired. This is also the reason for having a minimum space between trees. A simple modification linking the counter to this part of the system would ensure that the counter only counts when the nozzles spray and cannot count again until they have finished.

The speedometer failed to work because it was difficult to maintain enough friction of the flywheel on the conveyor belts. It was decided that future speedometers should be linked physically to one of the drive components. One such device would be a small pair of magnets on the framework and on a pulley, such as those on bicycle speedometers.

2.2.4 Experimental data to support development of the spray application component

During early use of the DCS system a discrepancy between the calculated spray volume and the actual spray volume was noted during use of the pulsing component. One cause of this was highlighted as the difference in nozzle size used and that intended for the normal application of the component. The nozzles used here are approximately eight to sixteen times smaller than the intended nozzle size that would be used on a boom sprayer in the USA. This causes a problem when the volume of the chamber between the solenoid valve and the nozzle orifice is considered. This space is quite large relative to the output of the nozzle and resulted in the nozzle spraying for approximately 0.5-1 seconds longer than intended. This spray was 'powered' by the residual pressure in the system. However, as the pressure fell the normal spray cone collapsed into a thin stream. This problem was remedied by placing a small plastic cylinder into the chamber before the nozzle, occupying a large proportion of the space. Additionally, a much finer filter but larger filter was used. The combined effect was that most of the chamber space was used up and any small particles that may block the solenoid valve open would be filtered. This largely cured the problem and the nozzle stopped spraying much more quickly. Unfortunately, some discrepancy still remained. This was considered not to be a problem as long as it could be calibrated. The next section discusses the calibration curve produced between expected and actual sprayed volume. It also details an experiment comparing the addition of a small electronic component, which should shut off the nozzles even quicker after the power is cut.

2.2.4.1 Use of a Zener-diode arrangement to prevent back EMF and calibration of the DCS at different pulse settings

A Zener diode arrangement is a group of three diodes in parallel to the rest of the circuit. In the diagram below (Figure 2.4), a basic circuit is shown with a power supply, a contact (to open and close the circuit on command) and the solenoid nozzles. The solenoid nozzles operate by switching on and off an electromagnetic field very quickly, thereby opening and closing a valve.

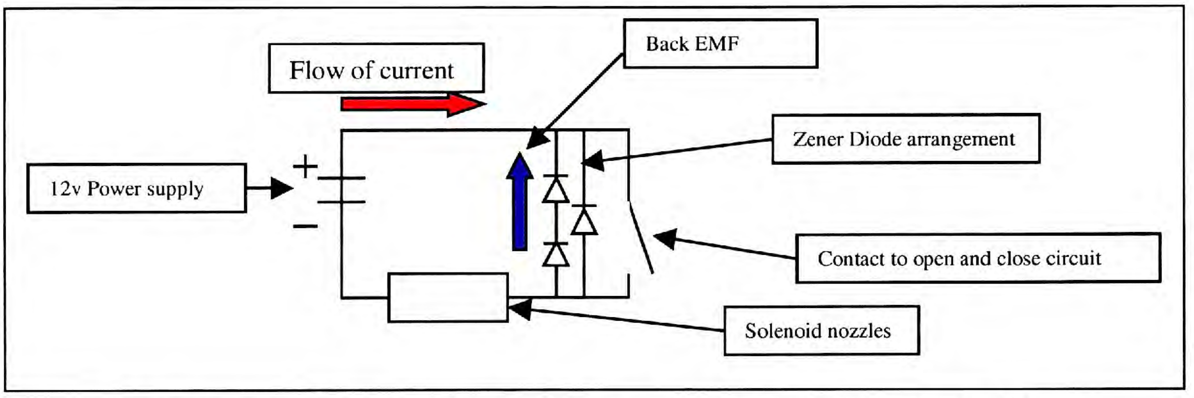


Figure 2.4 **Use of a Zener diode arrangement to prevent back EMF discharge from the Solenoid nozzles. See text for full explanation.**

As the contact opens (after a preset spray time), the power in the circuit is cut and the electromagnetic field in the solenoid nozzles stops. However, energy cannot be created or destroyed. Therefore the physical magnetic field is converted back to a current. The current cannot be dispersed easily because the contacts in the circuit are open. Therefore, by adding an arrangement of diodes (which allow the flow of current in only one direction) the current can dissipate more quickly. This has the advantage that the nozzles close much more suddenly after the contact opens. It is possible that this has been the cause of the discrepancies in output volumes that have been identified.

2.2.4.2 Materials and methods

For this trial, two nozzles were tested; AI (air induction) and FE, a flat fan nozzle. In addition, the FE nozzle was tested in conjunction with the Zener diode arrangement. For each nozzle type, water was collected from one of the nozzles in a measuring cylinder. A previous experiment showed that there was no difference in the output of either member of the pair. The nozzles were set to spray for three different time periods; 0.2 seconds, 0.4 seconds and 1 second. At each of these settings three replicates were taken for each of three pulse settings, 100%, 50% and 25%. Pulse settings represent the proportion of time that the nozzles are open. For each replicate the volume collected was from several sprays, which varied depending on output and the average taken.

2.2.4.3 Results and analysis - Use of the Zener diode arrangement

Analysis showed there to be no significant differences in the output of the nozzles with and without the inclusion of the Zener diode arrangement. The results from the two groups were almost identical although duration affected the volume sprayed (Figure 2.5).

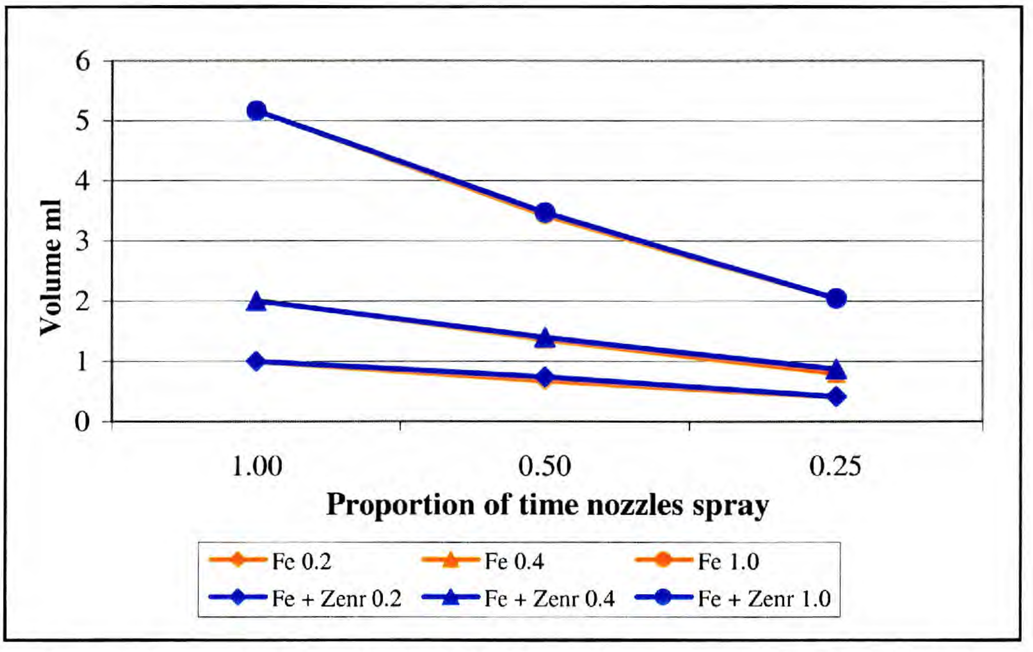


Figure 2.5 **Output volume of the FE nozzle in conjunction with the pulsing system with and without the Zener diode arrangement. The legend shows the nozzle and duration of each spray.**

2.2.4.4 Results and analysis - Comparison of FE and AI output volumes with the expected output

This section aims to examine and quantify the discrepancies between the volume sprayed and the expected volume. It also aims to compare the FE and AI nozzles, which might be affected differently by changes in the pulse settings.

Estimated outputs were calculated based on the volume sprayed at the 100% pulse setting and then divided by two and four to obtain the expected output for 50% and 25% pulse settings, respectively. These were plotted as percentages for observed and expected values (Figure 2.6 and Figure 2.7). It can be seen for both nozzles that the observed values are greater than that expected. For both nozzles there is an interaction between the pulse setting and the time that the nozzle sprayed for ($P<0.001$, $F=8.07$, $df_{1,18}$). This indicates that the discrepancy does not change in a linear manner for different spray durations.

There were no interactions between nozzle and either pulse setting or spray duration. However there was a more complex three-way interaction between pulse setting, spray duration and nozzle type ($P=0.067$, $F= 12.21$, $df=1$, $n=18$). There was also an interaction between pulse setting and spray duration ($P<0.001$, $F=6.23$ $df=1$, $n=18$).

In addition to these complex interactions, Figure 2.6 and Figure 2.7 show that the volume sprayed for any setting at the two pulse settings of 50% and 25% are less than would be expected. For example, at typical operating spray durations of 0.4 seconds, the reduction is only 61% instead of the expected 75% reduction.

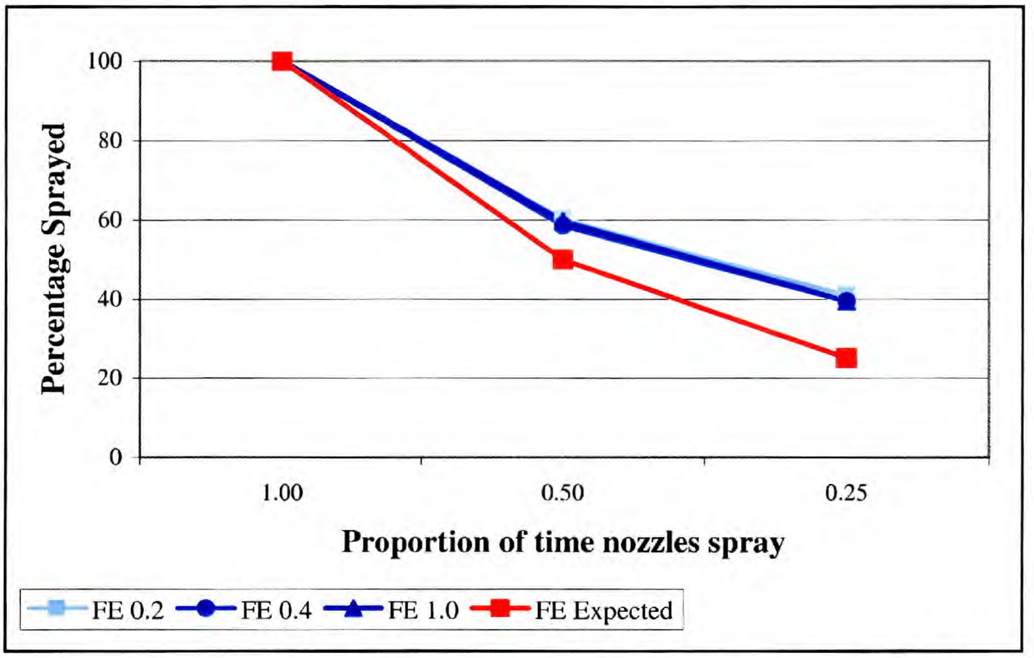


Figure 2.6 **Comparison of the observed (blue) and expected (red) volume outputs of the FE nozzle at different pulse settings and three different spray durations. The system sprayed more than expected on all occasions.**

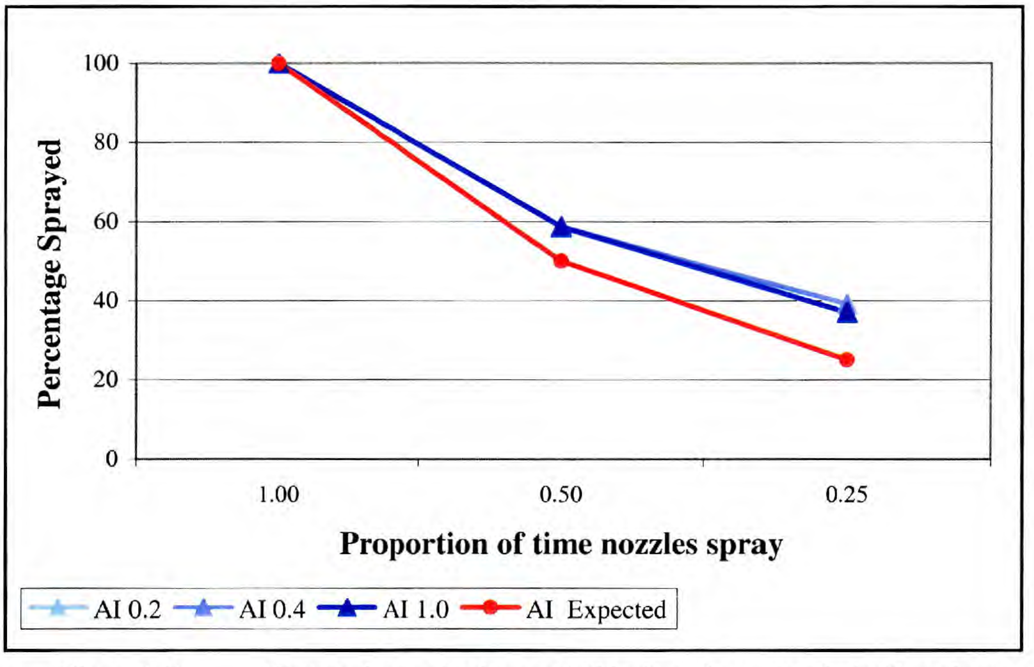


Figure 2.7 **Comparison of observed (blue) and expected (red) volume outputs of the AI nozzle at different pulse settings and three different spray durations. The system sprayed more than expected on all occasions.**

2.2.4.5 Conclusion

This experiment has highlighted the need to recalibrate the installed pulsing system. The use of the Zener diode arrangement failed to reduce the discrepancy between the observed and expected flow rates. However, this does not mean that the Zener diode arrangement is not working at all, merely that the affects are quite small and not noticeable within the range of this experiment.

The results indicate that the AI nozzle behaves slightly more as expected than the FE nozzle, presumably due to its larger flow rate, for which the system was designed. At the minimum volume, the AI nozzle gave an average (of the three durations) of 39.4% compared to 39.9% for the FE nozzle.

The interactions between spray duration and pulse and the three-way interaction between spray duration, pulse and nozzle indicate that calibrations may need to be made for each set spray duration, as the results may not easily be extrapolated. However, an alternative explanation and probably the main reason for the interactions with spray duration may have stemmed from the methodology. Without digital dials repeatedly selecting a spray duration using the analogue dial may have yielded small differences. Coupled with the identical values of each replicate, significance could well be placed where it is not warranted, particularly for the interaction terms. When examining the data, the percentage reductions for each of the three spray durations are all remarkably similar. Therefore the cause is more likely to be due to the accuracy of the dials, rather than some complex set of variables producing the highly significant interaction terms.

These data have redefined the lower limits that the DCS system is capable of spraying a sapling. This data must now be used in conjunction with calculated efficacy values (Chapter 4) to determine how much and how little can be deposited onto the saplings.

2.2.5 The framework and panelling

Key Criteria The framework must support and safely house the conveyor belt and the application system. Therefore, it must be strong and water proof and must survive the rigours of sapling treatment. It must be possible to construct in Silwood Park with the facilities available and cost must be kept to a minimum. The framework must be adjustable to allow for design modifications. The panels must be transparent, removable and prevent operator contamination with pesticide.

Two separate framework designs were generated; one was a concept model in which aesthetics were important (Figure 2.1). The second was for the prototype design (Plate 2.8). As there were no welding facilities available at Silwood Park, it was decided that a metal framework would be difficult to build. Instead Dexion framework, commonly used for shelving was obtained from STS storage (Slough, Berkshire). It is also known as slotted angle because it is a metal flat bent at right angles giving it great strength. It has a series of holes along the entire length to fix bolts through. Therefore, it is immensely adjustable and strong frames can be built. It is not completely ideal for housing two conveyor belts, which are under tension, but by triangulation and reinforcement of the key stress points it proved adequate for the prototype.

Plate 2.8 DCS system

In this view of the DCS system, the Dexion framework and some of the panels can be seen. The protective panel by the entrance has been removed for this photograph. The panels at the entrance end can be seen to taper into a central canal which houses a drain pipe that will direct pesticide residue away from the operator into a safe container in the centre of the system.

Several redesigns were necessary after construction commenced so that the framework was stronger and did not interfere with the function of the belts. For example, the edges of the pillow bearings would have prevented trees passing down the central canal. They had to be reoriented so that they did not do this, therefore the framework had to be changed accordingly.

In addition to triangulating key areas, six shelves were incorporated into the design. This gave space to house the electric motor and gearboxes (top shelf which needed to be strong and well supported), the electronics, the two belt tensioners, and two large shelves running the full length of the machine, which provided some workspace. The shelves effectively triangulate the areas that they are attached to, providing extra strength. These shelves were made from ¾ inch plywood that was treated with a wood stain to protect them from moisture.

Standard 8mm nuts and bolts were used to fasten all joints. However, some parts such as the areas around the bearings and the electric motor are prone to vibration. For these joints 'Nylock' type nuts were used, these have a rubber seal that prevent the nuts from coming undone.

It was intended to use PVC or acrylic/perspex panels on the final machine. These will provide protection from the pesticides, PVC will not absorb pesticides, and also provide the operators with a view of the interior workings. Other materials were investigated such as polycarbonate and fibreglass. Unfortunately polycarbonate will crack on contact with water unless it is protected and fibreglass is not transparent at the required thicknesses. For the final system, a choice will be made between PVC, which is strong and Perspex, which has better optical qualities. Other important aspects will be investigated such as chemical resistance, strength, flexibility, impact strength and cost, when the final machine is built.

These materials are too expensive to use for the prototype design and would be difficult to modify as the design is developed. Therefore clear plastic polyethylene sheeting was used. This is easy to cut, extremely cheap and is not permeable to water and just like more expensive materials, it will contain all the pesticide vapour.

However, it may absorb some of the pesticide and therefore care will have to be taken to dispose of the panels after pesticide use in the field trial.

2.2.7 Recent developments to the framework and panelling since the initial development

During the field trial it was found that there was evidence of pesticide vapour and maybe droplets escaping from the system. This could pose a risk to operators, exposing them to pesticides. Two main modifications are required on the system before further pesticide usage. The first change is to incorporate an extractor fan system, which is discussed in Chapter 5. The second change is to modify the current panels so that the majority of the vapour is contained within a sub-section of the conveyor.

Currently the conveyor has a small opening at each end allowing trees to enter and exit. The nozzles are in the centre of the system. Towards the exit end there is also a second panel that protects the electronics and the pulleys under the motor. By placing a second set of panels towards the entrance end, a significant proportion of the spray is retained in a central section.

In Chapter 4, different nozzle positions are described. One such position involves aiming the nozzles towards the oncoming trees. Unfortunately, this also involves aiming the nozzles toward the entrance opening. Therefore the panels will need to be repositioned, as the consequence of this re-aiming is that the excess spray lands much nearer the operator. If this evaporates, it could cause problems for the operator. Combining improved modifications to the panels with an increased conveyor belt length will decrease any risk to the operator whilst maintaining the improved efficacy of the frontal position discussed in Chapter 4.

2.2.8 The pesticide storage, pump, collection and recycling system

Key Criteria This system must safely pump pesticides to the nozzles where it will be sprayed. The pesticide must be stored in a suitable container, which will not leak. Any residue that is produced must be collected and funnelled to a suitable collection container. If possible this residue should be recycled.

There are two main methods of pumping aqueous pesticide from a container to the nozzles. The first that has been used for the majority of the testing is a diaphragm pump. It does not require any pressurisation of the storage container and therefore is less likely to cause an explosion if used incorrectly. The key advantage is that the pesticide is continually recycled and therefore settlement does not occur. Unfortunately, the diaphragm pump can be quite noisy and become irritating after several hours of use. This could largely be reduced by housing the pump away from the DCS system and using sound proofing material.

Alternatively, a compression system combined with a compression tank could be used. A large air compressor situated at Silwood Park (Ascot, Berkshire) pumps compressed air to several outlets inside the building. A compression tank can be connected to this system and maintained at a constant pressure. However, as the pesticide is not recycled, settlement of the suspended pesticide (for wettable powder formulations such as the α-cypermethrin intended for use in the trial) is quite likely to occur. It is also not as practical for use in large trials, as it has to be agitated regularly, although it is ideal for testing as it is much quieter and the pressure can be more accurately controlled.

When the compression system is not in use an alternative tank has to be used to store the pesticide. A 60l spray tank was obtained for this purpose from Team Sprayers (Ely, Cambridgeshire). It was mounted on a frame that has long metal struts running the length of the base to allow the tank and frame to be easily pushed under and pulled out from the DCS system. It is not possible to compress this tank, so it must be used in conjunction with the diaphragm pump.

The panels on the sides of the conveyor taper in to the centre at the base, allowing the pesticide residue that misses the sapling to drain in to a gully (Plate 2.8). The gully leads into a piece of house drainpipe that is angled down towards a second container via an outlet. The residue runs along the drain and collects in the container. The container is situated at the end of the treatment zone so that pesticide drains away from the operator.

Even after short uses this gully accumulates dirt, needles and sometimes saplings. This would further contaminate the pesticide waste making it more difficult to filter. In a modification to this system, not yet installed, three gullies will be used. The outer two collect pesticide residue as it runs off the side panels. These are not directly below the trees and should remain fairly clean. The central gully will collect pesticide that may drip off the saplings (although this should be minimal) which is likely to be dirtied by the mud on the sapling.

Once the pesticide is collected inside a suitable container, recycling is a simple matter. There are two options available. Firstly, the pesticide could be taken away and filtered in a separate unit, possibly examined to ensure it is still at the correct concentration (a fluorescent marker dye may aid in this process). Secondly, a recycling unit could filter the residue and return the cleaned pesticide into the original tank. In order to first establish the parameters that a recycling unit would encounter, such as volume of waste and level of contamination and reduce cost, a recycling unit was not installed on the prototype. One drawback to the use of a recycling unit may be that many pesticides will bind very strongly to soil and become inactivated. It may be difficult to assess to what extent this has occurred. Therefore, future development should focus on preventing the spray from becoming contaminated and focus on methods to capture clean spray and reduce the total volume of waste.

The DCS system was designed to run on mains electricity (240V AC). Most of the systems that were purchased have AC plugs. However, the electric motor may run on DC current, but it has an AC-DC converter. The nozzle system also requires DC current and this has been rigged with a 'switch mode' AC-DC converter. The benefit in using AC is that everyday 12/24V batteries (such as a car battery) would need to be recharged daily with the demands of this system.

2.2.8 Developments made to the pump and storage system after initial development

This system has remained largely unchanged after the initial development. During the trial it was found to be more convenient to use the air compressor to pump the pesticide to the nozzles. A future system should also aim to use an air compressor, but with an automatic agitator built into the system so that suspended particles do not settle to the bottom of the tank over time. This could be either a magnetic stirrer or some of the pesticide can be pumped back into the top of the tank to cause surface agitation and re-suspend the particles.

To improve safety a closed mixing unit should be added. This would be a simple device where a new bottle of pesticide is placed upside down into a small booth. This will be lever operated and a hollow tube will pierce the lid and drain the bottle into a pre-measured volume of water. The agitator can then be activated to adequately mix the pesticide. Similar systems have been seen on the Electrodyn conveyor system and could be easily adapted for this application.

2.3 Summary

Key areas of the DCS system have been identified and described. This allows future developmental work to be carefully structured. Experimental data to support some of the developments were given. At the end of this period of work, some of the improvements described above have actually been added to the system, but due to lack of funding a second field trial was not possible and therefore the effects of the modifications could only be shown on a physical and not a biological basis.

Chapter 3
Field Assessment of the DCS system

3.1 Introduction

On completion of the initial design and production of the Dandrive Conveyor Sprayer system (DCS), it was used to treat forest saplings for the Forestry Commission in April 2001. These were incorporated into a large-scale field trial. The aim of the field trial was to compare the level of plant protection offered by the DCS system with two formulations of a pyrethroid insecticide and also compared to the current Electrodyn and dipping treatments. This section examines the efficacy of plant protection in a field situation, when numbers of *Hylobius abietis* were very high. In addition, it is also necessary to examine any potentially negative effects of treatment by the DCS system. The main area of concern was plant phytotoxicity with the use of different pesticides and after the plants had been exposed to periods of cold storage. This following section, therefore, has three experimental trials each examining a different aspect of the potential for commercial use of the DCS system.

3.2 Background

It has been reported (Heritage, 1996) that unless transplanted saplings are protected by plant protection products, an average of 50% of the stock will be lost in the first two years through damage by *H. abietis* and *Hylastes sp.*. When densities of these two insects, particularly *H. abietis*, are high enough, 100% mortality may occur. Losses of only 10% require replanting to ensure that minimum planting thresholds are achieved (Evans H.F., *Pers. Comm.*). Replanting is a major expense and uses up valuable foresters' time and therefore methods that reduce mortality below this threshold are usually employed. Chapter 1 reviewed the current methods of protecting plants, such as dipping and the Electrodyn system.

These methods currently use the pyrethroid insecticide permethrin, but the insecticide is likely to be withdrawn from use in agriculture in any EC country after 2003 as manufacturers have not supported its re-registration under an EC directive (EC 91/414). In addition the Pesticide Safety Directorate (PSD) announced that the dipping of plants in permethrin would not be allowed after June 2000 (Heritage S.G., *Pers. Comm.*). However a key feature of the DCS system is that it was designed to use off-the-shelf formulations, thereby reducing registration time and increasing its adaptability to any future change in pesticide registration requirements.

In 1999/2000 the Forestry Commission conducted trials comparing the efficacy of a range of insecticides for possible use in forestry (Heritage S.G., *Pers. Comm.*). Potential pesticides for use in forestry must be long lasting in the field so that protection is provided all year round. Pesticides that break down rapidly in the environment, such as dimethoate (an organophosphate) would not be particularly suitable (Dent, 1993). The Forestry Commission trials found that two formulations of a pyrethroid; ι-cyhalothrin (Hallmark MC, Syngenta) and α-cypermethrin (Contest WG, Aventis) were particularly effective at providing protection from *H. abietis*. The trials discussed here examine the use of ι-cyhalothrin in conjunction with the DCS system. There are three sections; efficacy of ι-cyhalothrin to protect forest plants from damage by *H. abietis*, the efficacy of five doses of ι-cyhalothrin (dose response) and the effects of cold storage on the phytotoxicity of ι-cyhalothrin applied by the DCS system.

3.3 Materials and methods

3.3.1 Measurement of damage

To ensure that all Forestry Commission trials are assessed equally, a Standard Operating Procedure (SOP) has been drafted for the measurement of damage to transplanted saplings (all relevant SOPs are detailed in Appendix 4). Often these trials are very large, therefore the level of assessment has to be kept to a minimum to reduce overall assessment time. Assessments were carried out after *H. abietis* activity had ceased, typically in late October to early November, but finished before the end of March when feeding damage is likely to resume.

The assessment was based on a four point scale; A – No evidence of damage; B – Slight damage, which can cover a large area, but must not involve girdling of the stem (plant likely to survive); C – Severe damage, may only cover a small area but stem is girdled (plant is unlikely to survive or is already dead); D – plant dead but not due to *H. abietis* damage; X – plant missing (Figure 3.1).

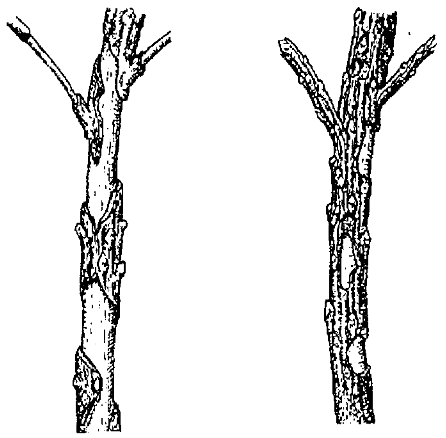


Figure 3.1 **Assessment of damage to plants caused by *H. abietis*. Severe damage (left) can be small and localised but involves girdling of the bark. Slight damage (right) may involve extensive damage but does not involve girdling. It is the presence of girdling that determines whether the sapling will die or recover.**

It was important that all of the trees were thoroughly examined, especially those in loose soil in which socketing may occur (build up of loose windblown soil around the base of stem). *H. abietis* may feed below ground in these circumstances and the damage is not so obvious.

3.3.2 Selection of field sites

Testing of new insecticides and application procedures must be carried out under the highest insect pressure to ensure that the results are a reliable worst case scenario under all conditions. A range of factors can affect *H. abietis* abundance and therefore these must be considered and kept constant in any field trial. These factors are listed in SOP 104 (Appendix 4e). However, some factors are of particular interest. Field sites must previously have held a coniferous crop that was felled between 10 and 21 months prior to the planting of the experimental trees. This is to ensure that a resident population of emerging *H. abietis* are present. Ground preparation, surrounding vegetation and weeds are all known to reduce *H. abietis* feeding pressure and should be manipulated so that pressure is increased. Finally sapling quality, size and spacing (in particular from the edge of the forest) should all be kept constant. The field sites used were found across the UK and are detailed in Table 3.1.

Table 3.1 **Details of the field sites used in the Efficacy trials**

Site	Field Site	Location	Chapter 4 Forest District	Grid Ref.	Elev. masl	Soil Type
A	Newton	Culbokie	Inverness	NH609586	110	Humus iron podsol
B	Cairnbaan	Fire Tower Road, Lochgilphead	West Argyll	NR 867912	115	Peaty gley
C	Cairnbaan	F1, Lochgilphead	West Argyll	NR 878906	100	Deep peat
D	Bush	Glen Finlet	Tay	NO 244647	360	Upland brown earth
E	Bush	Loch Ard	Aberfoyle	NS 476956	140	Peaty brown earth
F	Bush	Wauchope	Borders	NT 602029	360	Deep peat/peaty gley
G	Mabie	Lauriston	Galloway	NX 658667	180	Peaty gley
H	Mabie	Kirkland	Ae	NY 018913	340	Flushed gley
I	Mabie	The Coomb	Kielder	NY 765915	330	Flushed gley
J	Wykeham	Harwood Dale	North York Moors	SE 974975	130	Peaty Gley
K	Wykeham	Langdale	North York Moors	SE 909955	220	Iron pan
L	Fineshade	Roudham	East Anglia	TL 932 878	30	Sand
M	Talybont	Clocaenog	Llanrwst	SJ 009 555	445	Upland brown earth
N	Talybont	Allens estate	Coed Y Cymoedd	ST 032 951	320	Upland brown earth

3.3.3 Layout of plants and species selection

Sitka Spruce (*Picea* sitchensis) was used for all the trials to allow comparisons with previous Forestry Commission trials. Between each experimental treatment a buffer row of plants was planted to reduce possible edge effects between treatments. All saplings were planted within 14 days of treatment unless specified otherwise (for example, the cold storage trial) to reduce phytotoxic damage by the pesticide.

3.3.4 Use of the DCS system

Use of the DCS system has been drafted as a SOP (Appendix 4), which details use of pesticides in the system, mechanical operation and details on how to change the nozzles and settings. For this series of trials, a common group of settings were used to maximise pesticide deposit on the plants and also to increase reliability.

The delay period, which prevents the nozzles from spraying before the sapling reaches the spray cone, was set to zero. At that stage of development the trigger system was not accurate enough to ensure that all trees would be accurately targeted whilst they were in the spray cone. Thus, removal of the delay period ensured that for the purpose of this trial, all would be adequately treated. Two Tp6501E flat fan even spray nozzles (FE) were used, each with a flow rate of 0.36 l/min at the 2.5 bar operating pressure, applying 12 ml per tree. The nozzles were positioned at right angles to the line of tree movement (side position). The conveyor belt was running at 20 m/min. This was checked and recalibrated between treatments to ensure the correct speed was maintained.

All saplings were handled with care during the trial as root growth potential can easily be damaged through negligent handling. The saplings were allowed to warm to at least 5 °C before treatment. The roots of the saplings provided were often extremely tangled and required an operator to separate and bunch them so that the second operator could feed them in to the DCS system more quickly and efficiently. Saplings were treated in batches of 200 to ensure that no more saplings were removed from the humid co-extruded bags than was necessary. The roots of the saplings were kept covered by co-extruded bags as much as was possible, both before (during preparation) and after (before packing) treatment to minimise desiccation of the roots.

All of the DCS treatments were carried out in IPARC, Silwood Park (Ascot, Berks). Pesticide suspensions were mixed immediately before the trials and once the saplings were treated, they were placed in a co-extruded polythene bag (black on one side, white on the outside) with the tops exposed and left to air dry for 30 minutes. At the end of the day they were dispatched to a cold storage facility (2 °C, high humidity). Saplings were planted within ten days of treatment.

3.3.5 The efficacy of ι-cyhalothrin sprayed by the DCS system to protect forest plants from damage by *Hylobius abietis*

A total of 3600 Sitka spruce were divided amongst 15 field sites (each 0.15 hectares in UK) with six experimental treatments and two blocks per site. Each treatment contained 20 saplings planted as a 5x4 grid at 2m spacing. The experimental treatments are listed in Table 3.2.

Table 3.2 **Treatments for the DCS efficacy field trial (PPP01007).**

Spray system	Active Ingredient	Formulation	Concentration % Active Ingredient
DCS	ι-cyhalothrin	Hallmark	0.4
DCS	ι-cyhalothrin	Hallmark	0.8
DCS	ι-cyhalothrin	Hallmark	1.6
Electrodyn	Permethrin	Permasect 12ED	12
Dip	Permethrin	Permasect 25EC	0.8
DCS	Water	Control	0
Dip	Water	Control	0

The experiment was carried out at the beginning of April 2001, slightly later than the recommended deadline, due to Foot and Mouth Outbreaks. All saplings were checked in November 2001. Results were analysed using Anova. Further details including site locations can be found in Appendix 5.

3.3.6 The efficacy of five doses of ι-cyhalothrin applied using the DCS system

A total of 1500 Sitka spruce (*P. sitchensis*) were distributed between 6 experimental treatments, each with five replicates on two field sites. Each replicate contained 25 saplings planted in a 5x5 block with 2m spacing. The six treatments are detailed in Table 3.3.

Table 3.3 **Details of the concentrations of pesticide sprayed on to Sitka spruce saplings with the DCS system in a dose response trial.**

Spray system	Active Ingredient	Concentration % Active Ingredient
DCS	Water	0
DCS	ι-cyhalothrin	0.1
DCS	ι-cyhalothrin	0.5
DCS	ι-cyhalothrin	1.0
DCS	ι-cyhalothrin	1.5
DCS	ι-cyhalothrin	2.0

The two field sites (Braemore and Wauchope) each had different aspects. Braemore was previously planted with larch and had abnormally high populations of *H. abietis*, whilst Wauchope had been planted with Sitka spruce and had more normal populations. The mortality and damage to the saplings was assessed in November – December 2001 by the methods described previously.

3.3.7 The effect of cold storage on the phytotoxicity of ι-cyhalothrin

A total of 660 Sitka spruce (*P. sitchensis*) were distributed between four treatments (Water control, DCS 0.4%, 0.8% and 1.6%) and three storage treatments (0, 4 and 8 weeks). For each combination there were 12 saplings planted at 0.3m spacing. The treatments were randomised within each block and planted on one field site.

For this trial the plants were treated in the DCS system as described previously, but were sealed in co-extruded bags within 15 minutes of treatment and sent to a cold storage facility (2 °C, high humidity). The plants were kept there for the specified length of time (0, 4 or 8 weeks) and then planted. Therefore the saplings were planted at different times of the year. This trial measured the survival and growth increment of the saplings from planting until November 2002. This gave an indication of the phytotoxicity of the pesticide and cold storage. The results were analysed using Anova.

3.4 Results and analysis

The assessment of the saplings was carried out in one of two ways. For the efficacy trial and the dose response trial, the survival probability was assessed, whilst for the phytotoxicity trial the annual growth increment was recorded.

The collected results varied greatly across the different field sites, especially for the efficacy trial. However, the trends between treatments were similar and averages across the sites gave favourable results for the DCS system compared to the Electrodyn and dipping systems.

This chapter has been carried out in collaboration with the Forestry Commission and the data were initially analysed by FC using angular transformation and Anova. However, for the purpose of this results section, the data have been re-analysed using GLM in S-Plus and discussed from the viewpoints of replanting thresholds and attempts to explain the occurrence of damage on treated saplings.

3.4.1 The efficacy of ι-cyhalothrin sprayed by the DCS system to protect forest plants from damage by *Hylobius abietis*

This was the key trial in this series of DCS field evaluations and aimed to show if there was a greater level of protection offered by the DCS compared to other treatment systems. Unfortunately, there were no significant differences between any of the pesticide treatments (Figure 3.1). However, there were significant differences between the two control treatments and all of the pesticide treatments (GLM + Tukey post hoc test, for all contrasts; $P<0.001$, $F=13.52$, $df=1$, $n=182$).

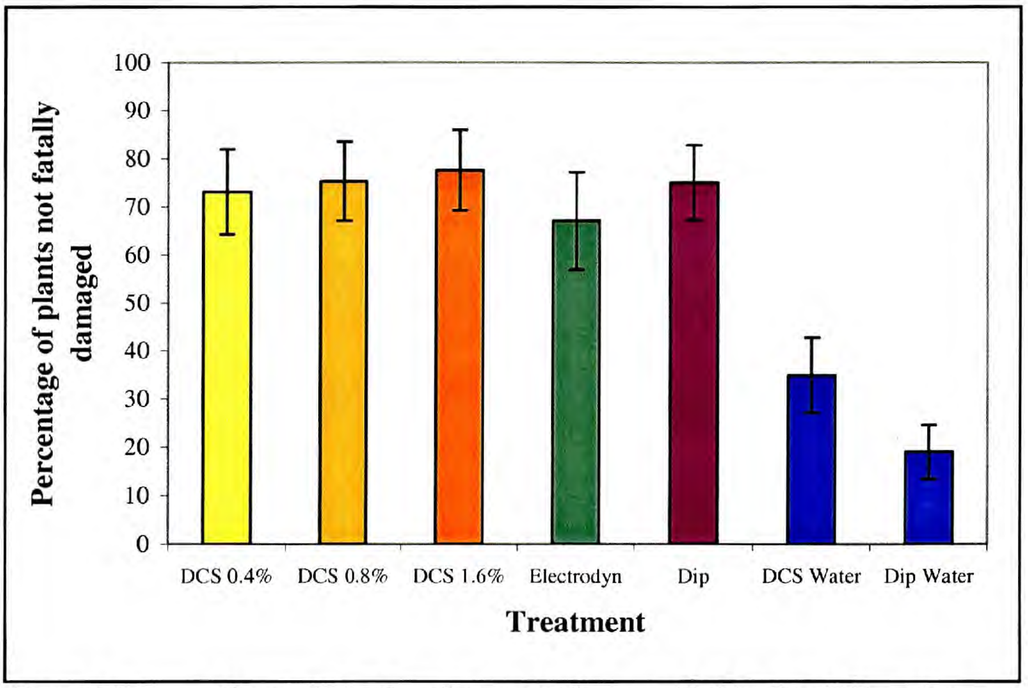


Figure 3.1 **Comparison of sapling protection provided by the DCS system at three pesticide concentrations compared to the standard Electrodyn and dipping treatments. All pesticide treatments were significantly different from the two control treatments (GLM + Tukey post hoc test, for all contrasts; $P<0.001$, $F=13.52$, $df=1$, $n=182$). There were no significant differences between any of the pesticide treatments.**

The data in Figure 3.1 are an amalgamation of those collected from 13 field sites (two sites were discarded due to Foot and Mouth restrictions), each with two replicates per site. There was no significant difference between the two replicates at each site. Of the 13 sites, two were not analysed individually due to the high number of 100% data points (recorded as no damage). Despite the low level of replication, analyses were carried out for each field site.

At two sites (The Comb and Roudham) there was a significant difference within the pesticide treatments. Unfortunately each of these sites showed conflicting results, one with dipping significantly better, one with the DCS significantly better. Therefore little conclusive information can be drawn from these individual analyses.

Unusually there were three sites in which there was no significant difference between the DCS water treatment and some of the pesticide treatments. At two of these sites there was a high level of protection from the dipped water controls, indicating a low

H. abietis population. Additionally, there was an overall significant difference between the DCS water control and the dipping water control.

At one site (Roudham) the DCS treated controls had a higher level of protection than both the dipping and the Electrodyn treatments. The dipping controls at this site suffered 100% mortality indicating a high weevil population. Overall damage to the dipping controls ranged from 50% to 100% fatally damaged plants.

Data were also collected on the number of slightly damaged plants, fatally damaged plants and undamaged plants. The results here have been presented only the number not fatally damaged (number undamaged + number slightly damaged). However each of these different sets of measurements have been analysed individually and generally the same patterns were evident for each set. However, there were two exceptions. The number of slightly damaged plants was significantly different between the two control treatments. Secondly, the number of fatally damaged plants was significantly different between the dipping and Electrodyn treatments. It is interesting to note that the number of slightly damaged plants interacted with the number of fatally damaged plants. Very high fatal damage usually resulted in a low number of slightly damaged saplings, which was independent from the number of undamaged saplings. This may explain the difference seen between the two control treatments.

Despite the lack of significant difference between the pesticide treatments, due mainly to the large variance between sites, it is interesting to note that the increase in concentrations for the DCS system gave increased protection as would be expected, although these are not significantly different. In addition, the highest doses (which applied volumes of active ingredient equal to (0.8%) and double (1.6%) that of the Electrodyn) gave nearly 10% more protection than the Electrodyn treatment, whilst giving very similar levels of protection to the dipping treatment.

3.4.2 The efficacy of five doses of ι-cyhalothrin applied using the DCS system

This trial aimed to identify the optimal concentration of pesticide for control of *H. abietis*. Two field sites were used and there was a significant difference between the proportions of fatally damaged plants between the two sites (GLM, Chi^2, $P<0.001$, df=1, n=60). However, there was no significant interaction between the two sites, therefore only the intercepts of the model differed.

GLM analysis showed that the dose had a significant effect on the fit of the model (GLM, Chi, $P<0.05$, df=1, n=60). The dose response curves for each site, Figure 3.2, indicate that at Braemore, the best protection was achieved at the highest two doses. However the mean proportion of fatally damaged plants was actually lower at the 2% concentration than the 1.5% concentration. The damage at Wauchope was generally very low and therefore even the lowest pesticide doses gave very good protection. At this site difference in protection was found between the 0.5% to 2% spray concentrations.

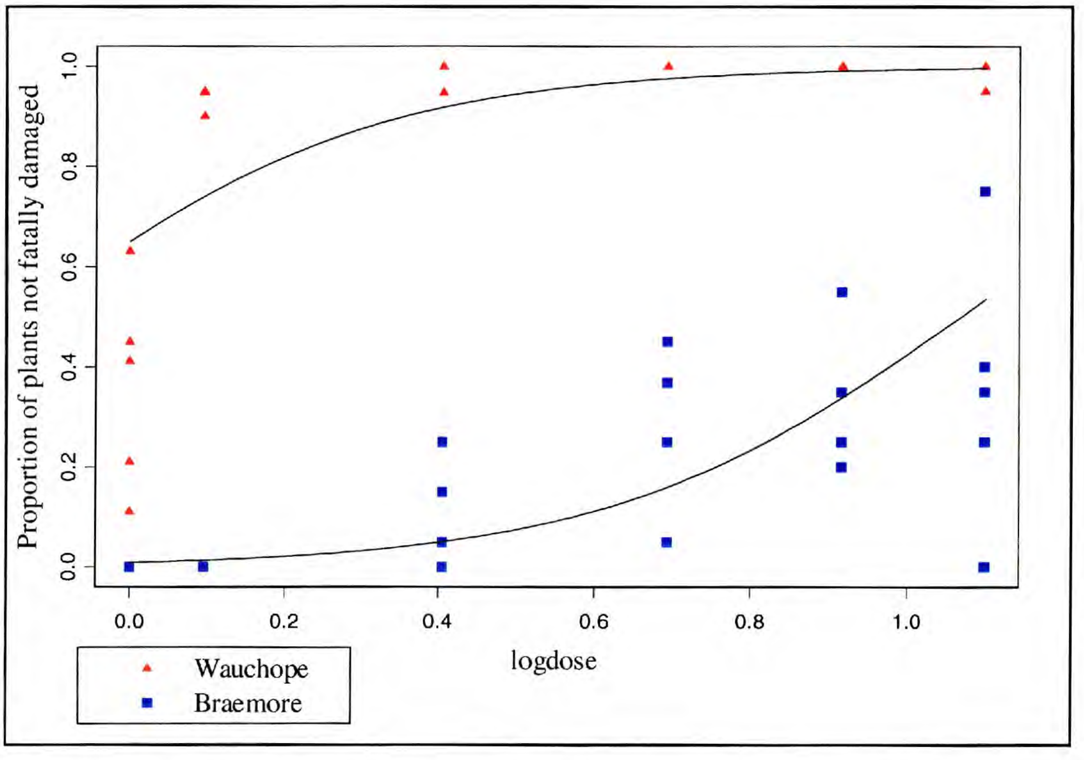


Figure 3.2 **Dose response series showing the proportion of plants not fatally damaged at 0, 0.1, 0.5, 1.0, 1.5 & 2% (left to right) concentrations of ι-cyhalothrin at two field sites. All saplings were treated using the DCS system. The most effective concentration was 1.5% ι-cyhalothrin, as the highest concentration (2%) did not give substantially increased protection at the Braemore site.**

3.4.3 The effect of cold storage on the phytotoxicity of ι-cyhalothrin

Cold storage significantly increased the growth of the Sitka spruce (*Picea sitchensis*) transplants with increasing storage length ($P<0.001$, $F=30.35$, df=1, n=720, Figure 3.3). This can be seen for all three pesticide treatments. However, the water control does not show this pattern.

There was no effect of pesticide treatment on the growth increment of Sitka spruce saplings ($P=0.93$, $F=0.15$, df=3, n=720). However, there was a significant interaction between storage and treatment ($P<0.001$, $F=6.84$, df=3, n=720). This is likely to be due to the different trend shown by the DCS water control.

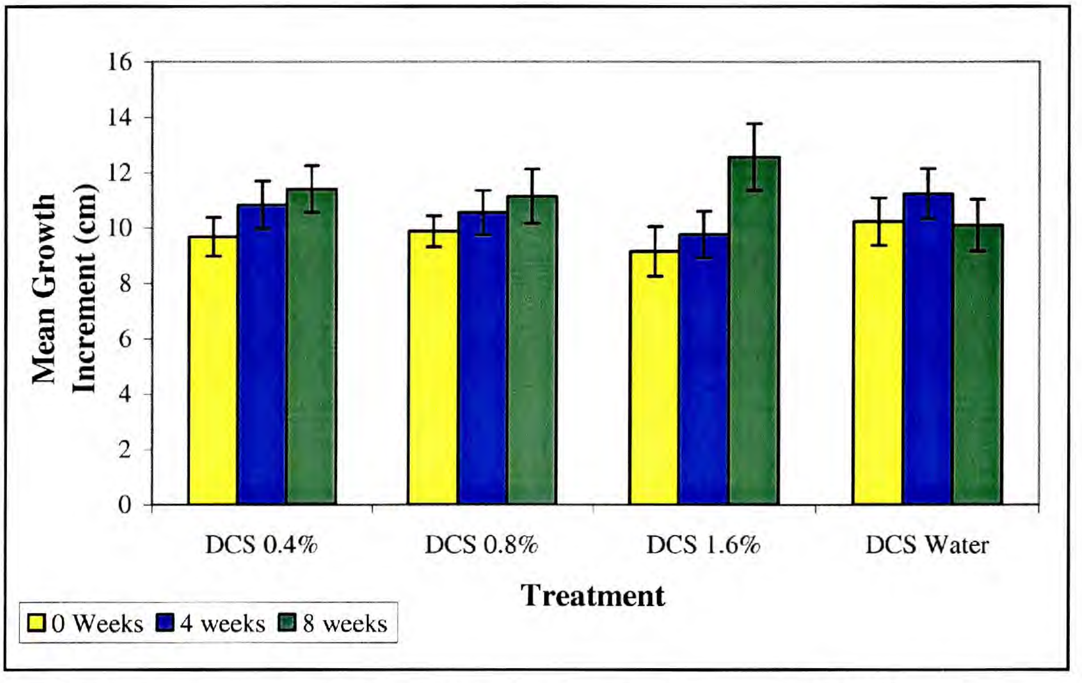


Figure 3.3 **Effects of cold storage on the growth increment of Sitka spruce saplings. There was a significant effect of storage ($P<0.001$, $F=30.35$, df=1, n=720) but no effect of pesticide treatment ($P=0.93$, $F=0.15$, df=3, n=720).**

3.5 Discussion

The collected data were initially very promising with the DCS system providing protection to a higher mean proportion of saplings than either the dipping or the Electrodyn treatments. Unfortunately, GLM analysis showed that there were no significant differences between any of the pesticide treatments with any of the systems. However, the data shows that the DCS system has performed at an equal level to those systems that have been in development for several years.

There was an increase in protection with the three concentrations of pesticide sprayed by the DCS system. There were no significant differences between these concentrations, but the dose response trial, which was better replicated, showed significant differences between treatments. It was estimated that the most practical pesticide concentration to use was the 1.5% concentration as there was no gain in protection with the highest concentration of 2%.

The pesticide was not phytotoxic to the sapling even after it had been in cold storage for up to eight weeks. In fact, plants kept in cold storage for longer had a significantly greater annual growth increment. This effect has been noted before for permethrin, which causes an increase in growth of the plants after cold storage and this agrees with the results, which only shows that treated trees had better growth with cold storage (Heritage S.G., *Pers. Comm.*). This is in contrast to the work by Kohmann (1999), who found that whole tree treatment with the permethrin formulation Gori 920 significantly reduced the frost tolerance of *P. abies*. In addition, the treated trees suffered reduced leader growth and increased mortality over two years. It was concluded that the responsible agent was the emulsifier used in the formulation, ethoxylated nonylphenol, and not the permethrin.

Therefore, it is also possible that the saplings planted later had more favourable (i.e. wetter) conditions and were able to make up for the shorter growing season, rather than a beneficial effect of the pesticide. Unfortunately the data for the dipping controls were unavailable, so it is not possible to compare whether the DCS system reduces annual growth.

The efficacy test was set up with low replication at each site, but over many sites. Unfortunately, it is because of this low replication and high 'between site' variability that has made it very difficult to distinguish any effects between treatments. The fact that the three DCS treatments show a steady increase in protection with increasing pesticide concentrations indicates that perhaps there are differences between the DCS and the other treatments.

This experimental design was used to ensure that sufficient *H. abietis* damage would occur whilst managing limited resources. With this in mind, it would have been better to record *H. abietis* population densities at each field site. The field site data could then have been converted into a continuous variable, therefore freeing up a number of degrees of freedom and giving the statistical model more 'resolving power'.

When examining data for individual field sites, it became clear that the damage was quite variable even within one block. It was unusual for the controls to have significantly more protection than the Electrodyn and dipping treatments, but this was the case for at least one site. With only two replicates at each site, it is not surprising that there is such large variation. In particular, the LSD values calculated by the Forestry Commission required two data points to be different by up to 40% and rarely less than 20%. However, the overall LSD was only 5% and therefore allowed much smaller differences to be distinguished significantly.

A second drawback in the experimental design concerned the way that trees were selected for the Electrodyn system. One of the author's concerns was that the electrostatically charged droplets would not be able to penetrate the foliage onto the stem, therefore some bushier saplings might suffer increased damage. Unfortunately it has been difficult to find evidence for this, until now. SOP104 details the procedure for evaluating plant protection products (Appendix 4). One section describes optimal plant size for plant protection trials and states that for Electrodyn trials, trees should be carefully selected to avoid those with bushy lower stems as bushy stems can prevent the droplets being deposited on the stem. In contrast, many of the trees provided for the DCS trial far exceeded usual height specifications of 25-40cm and were often either less than 10cm or greater than 70cm. In addition, these trees were heavily covered in mud, which is known to bind to and inactivate many pesticides.

This information indicates that the real levels of protection provided by the Electrodyn are probably unknown, only the 'best case scenarios'. In contrast, it seems that a 'worst case scenario' was generated for the DCS system, particularly with the increase in efficacy that could be achieved in future trials as discussed in Chapter 4.

It is interesting to note though, that DCS controls treated with water had better protection than water dipped controls. This effect has been noted in other Forestry Commission trials where water treated controls suffer less damage than untreated controls. This is possibly due to increased plant vigour after treatment, rather than a repellent effect of water. In part this could be due to a detrimental effect imparted onto the saplings by the vigorous drying procedure that dipped plants are subject to.

In summary, the DCS system gave a high level of protection to Sitka spruce saplings planted in the field, even under high insect pressure. The level of protection was always substantially more than control treatments without pesticide protection. Unfortunately, the level of protection was often not enough to prevent the need for replanting, except at the few sites where 100% protection occurred. Future work should aim to examine the effects of the modifications discussed in Chapter 4, in relation to the biological efficacy in the field.

Chapter 4
Development and Assessment of the Dandrive Conveyor system (DCS)

4.1 Introduction

Following the Forestry Commission field trials of the Dandrive Conveyor Sprayer System (DCS), improvements and modifications to the system were required to improve system performance and efficacy. This chapter deals with the changes to the working settings that will affect efficacy, such as nozzle type and positioning, spray duration and spray pressure. Generally, changes to the system settings can fall into three broad categories; physical changes to the nozzle type and positioning, changes that will affect droplet spectra and flow rate and finally changes at the tree surface that will affect deposition.

Changes in these categories may all have an effect on deposition. Firstly, it is important to redefine the deposition that is important in this system. Since *Hylobius abietis* damage is restricted to the root collar and lower stem and not to side branches and needles (Appendix 2), it is this area that needs to be targeted. However, other work (Chapter 6) has shown that the nature of protection may be by repulsion and not pesticide uptake so that exact targeting would not be so essential. Throughout this section, the deposition on both the stem and whole tree has been measured.

4.1.1 Physical changes to the nozzle type and positioning

For the majority of these trials, two flat fan type nozzles were used, which have very different droplet spectra. One of these nozzles, the air induction nozzle (AI) has the unique feature that it draws in air through a small orifice on the side of the nozzle (Matthews, 2000). The air is mixed into the droplets, creating a higher proportion of larger droplets than could be otherwise achieved with the same volume of water. The larger droplets have the beneficial properties typically associated with large droplets, such as low drift risk, without the increase in volume (Piggot & Matthews, 1999). The second nozzle was a standard low volume narrow angle flat fan nozzle (FE). Initial trials tested the possible use of other nozzles (Appendix 6). In addition, different spray positions were used, spraying head onto the direction of the tree, perpendicular to the tree and a combination of the two. It was found that different nozzle positions have implications for reliability as well as deposition.

4.1.2 Changes that affect flow rate

The flow rate of the nozzle, which is ultimately responsible for the volume sprayed per tree, can be varied by altering the spray pressure, adjustment of the pulsing control system and increasing nozzle aperture. Adjustments to the operating pressure though, have more than just an effect on flow rate. For example, an increase in pressure will increase the spray angle as the droplets have greater velocity on exiting the orifice. In addition, an increase in pressure will produce smaller droplets and there is a greater risk of the very fine droplets remaining airborne and being inhaled (Matthews, 1992). Sometimes these side effects can be used advantageously, for example, when farmers want to use larger droplets at the field margin to reduce the risk of drift (Matthews, 2000).

For each nozzle type there is usually a range of nozzles that have different flow rates, but droplet size and spectra are similar. In the DCS system, the volume sprayed must be kept to a minimum, especially as each tree is sprayed by two nozzles, therefore the type of nozzles was selected on the basis of their low flow rate. The use of nozzles with even lower flow rates has a strong risk of blockage from debris in the pesticide solution. The blockages are usually incomplete and many trees may be only partially treated before the problem is noticed. Therefore, it is important to first choose a reliable nozzle and second a low flow rate.

A final method of altering the flow rate is by using a built in system on the DCS that rapidly opens and closes the nozzles several times a second. This can reduce volume by up to 75% and still maintain the required spray pressure and droplet spectra. Previous systems that have attempted to do this have not maintained these criteria (Giles, 1997). This system is referred to as the Pulsing Control System.

4.1.3 Changes at the tree surface that will affect deposition

Deposition can be increased by increasing the ratio of pesticide on the stem to the whole plant. This can be achieved by accurately targeting the stem and avoiding the foliage. This targeting can be quite difficult because in many specimens there is a dense layer of needles and branches surrounding the stem that traps pesticide droplets. The Electrodyn system, in theory, is able to overcome this problem because the droplets are electrostatically charged and are attracted to the sapling (Matthews, 2000). Consequently the volume of pesticide solution that misses the tree is minimised. However, the pointed tips of leaves, particularly the very pointy needles of coniferous trees can reduce deposition through a gaseous discharge that flows from the point to the spray cloud, thereby neutralising the attraction between tree and droplet (Coffee, 1971). Whether this would result in increased deposition deeper within the tree or not is unclear.

There are additional methods of increasing droplet deposition. Firstly, by increasing the length of time that the tree is in the spray cone, i.e. by slowing the conveyor belts down, will lead to an increase in deposition. Secondly it may be possible to use an adjuvant such as agral. This is a surfactant and will aid in the dispersion of the droplets once they are on the tree. This could stop splash-back or bounce off and it could make the tree less 'wet' and safer to handle (Matthews, 2000). However, many of the commercially formulated pesticides contain a proportion of surface-active ingredients which aid in mixing with water and wetting of the leaves (Matthews, 1992). The use of a surfactant is discussed in Chapter 5.

4.2 Materials & methods

4.2.1 Fluorescent dye recovery and general spray methods

For some trials the trees supplied were very muddy and were rinsed and dried before use. This prevents contamination of the wash. Under the pressure, nozzle and timing settings of the experiment, 0.25g l^{-1} of Sodium fluorescein (Sigma, a brilliant green fluorescent dye) is sprayed onto each tree using the DCS system. This procedure was used to measure the volume of deposit on the sapling for all the following experiments. Unless stated, trials used both the Tp6501E (FE, Spraying Systems) nozzle and the air induction nozzle (AI, Lurmark). Two-year-old (1+1) Sitka spruce saplings were used for all trials. The numbers in brackets indicate that the tree was grown for one year in a nursery, then transplanted and grown on for a further year before dispatch to the forest for planting (Morgan, 1999). The trees are then dried indoors under low lighting, as the dye is photosensitive. This was typically for one hour depending on the room temperature. Care was taken not to cross contaminate the trees or to shake off any of the droplets.

Once dry, the roots were cut off and discarded and the basal 15cm of the stem removed. The side branches and needles were removed and put together with the remaining foliage from the top portion into 8x12cm resealable plastic bags. Into these, 500ml of water was added. The bags were then shaken for 30 minutes.

In addition to the treated trees, 100μl of stock solution was applied to the stem of a control sapling. This was left to dry with the other trees and then the dissected stem was put into a resealable bag with 500ml of water as described and similarly shaken. This was used to calibrate the spectrofluorimeter.

Following washing, a 3ml sample of the wash solution was taken from each bag and put into a cuvette. The fluorescence of this was then recorded using a spectrofluorimeter. Full details of the spectrofluorimeter operating protocol can be found in Appendix 7.

4.2.2 Effects of varied pulsing length

The Sitka spruce saplings were treated at a speed of 20m min^{-1}, a spray pulse of 0.56 seconds and 2.5 bar spray pressure. There were three treatments for this trial; 100%, 50% and 25% spray periods. Each treatment contained ten replicates.

4.2.3 Effects of nozzle position

The Sitka spruce saplings were treated at a speed of 20m min^{-1}; a spray pulse of 0.56 seconds and 2.5 bar spray pressure. There were three treatments for this trial; spraying from the front (frontal position), spraying perpendicular to the line of motion (side) and one nozzle at the front and one to the side (front/side). The last treatment was included, as many of the trees had been flattened during packaging so that the foliage was in one plane. Therefore, there was a greater chance that one nozzle could penetrate to the stem with little impedance. The rest of the experiment followed the same format as above. Each treatment contained ten replicates.

4.2.4 Effects of pressure

The Sitka spruce saplings were treated at a speed of 20m min^{-1} and a spray pulse of 0.56 seconds. There were three treatments for this trial; 2 bar, 2.5 bar and 3 bar. The latter two pressures equates to an increase of volume sprayed by 12.5% and 21% respectively. The rest of the experiment followed the standard format. Each treatment contained ten replicates.

4.2.5 Effects of speed

Two separate experiments were carried out for this trial. Firstly, only the FE nozzle was used but included the pulsing facility as a treatment and two different speeds (20m min^{-1} and 24m min^{-1}). This experiment aimed not only to examine the effects of speed but also any interaction that it may have with the pulsing system.

In the second trial, both nozzles were tested and this aimed to compare the effects of speed only. Therefore, three speeds were used; 16m min^{-1}, 20 m min^{-1} and 23 m min^{-1}. The rest of each of these trials followed exactly the same format as the previous trials.

4.2.6 Effects of nozzle position and spray duration

The Sitka spruce saplings were treated at a speed of 20m min^{-1} and at 2.5 bar pressure. There were a total of 12 treatments for this trial; two positions (front and side); two nozzle types (AI and FE) and three spray durations (0.2, 0.6 and 0.9 seconds). The last two settings, 0.6 and 0.9 seconds, are both longer than the length of time it took the sapling to pass through the spray cone, which is 0.56 seconds. Each treatment contained 20 replicates and a total of 240 saplings were treated.

4.2.7 General use of the Dandrive Conveyor System (DCS).

A Standard Operating Protocol (SOP) for use of the DCS system with and without pesticides was drafted, which also covers safety procedures and outlines hazards. Throughout the trials the nozzles were positioned at a distance that would spray an area approximately 15cm high from the root collar towards the top of the tree. This was calculated using trigonometry as the spray angle was also known. The roots were protected from spray as they were enclosed between the conveyor belts. This nozzle position was used as it gave the maximum volume of deposit on the target area, but did not allow the spray to disperse any more than it needed to. However, when the nozzles were positioned in the frontal position as described above, the distance of the nozzles from the sapling was constantly changing. In this position the AI nozzle was preferable as it was available in the 120° angle giving greater coverage at closer distances. Unfortunately, it is not available in the 60° angle to enable direct comparisons with the FE nozzle.

4.2.8 Droplet Sizing

The Volume Mean Diameter (VMD) is a measure of the average diameter that 50% of the droplets occupy by volume can be measured in the Malvern apparatus (Matthews, 2000). The Malvern uses laser diffraction measurements to calculate the size of droplets. The size of droplets was measured for the AI and FE nozzle at different parts of the spray fan. These data can be found in Appendix 8.

4.3 Results and Analysis

The following section consists of data from twelve small trials. To increase the reliability of the results, the data from all of these experiments have been grouped, log transformed and analysed together using Anova. This has led to some settings having greater reliability than others, for example, 20m min^{-1}, 2.5 bar pressure and 100% pulsing were included in all experiments whilst the other settings were not. However the experiment detailed in section 4.2.6 was in response to the earlier experiments to confirm the findings and therefore was analysed separately.

Each of the figures in the following sections displays the deposition on both the stem and foliage as a measure of the deposition per 2ml sprayed. Therefore, the differences in flow rate of the two nozzles and different pressures were taken into account. The unit of 2ml was chosen, as this is a volume that would ideally be sprayed onto the sapling, therefore it is sensible to indicate the level of deposition with this setting. Deposition on the stem was always a smaller fraction of the deposition on the foliage. Ideally this volume needs to be as high as possible, whilst the deposition on the foliage should be low in comparison. However, as these experiments only manipulate one setting at a time, an increase on the foliage generally indicates a more efficient setting.

4.3.1 Effects of nozzle type

Throughout the series of trials two different nozzles were used, the FE flat-fan and the AI nozzle. It was known that the flow rate and spray angle of the AI nozzle was greater than that of the FE nozzle by approximately 30% and 60° respectively. When the deposition per 2ml (DUE) is considered, the AI nozzle deposited significantly more than the FE nozzle ($P<0.001$, $F=134.26$, $df=1$, $n=841$, Table 4.1), but only for deposition on the stem of the sapling. There was no significant difference between nozzle type and foliage deposit.

Table 4.1 **Mean volume (ml) of deposit and deposit per 2ml (DUE) sprayed for two nozzles on two plant parts.**

Plant part	AI Nozzle		FE Nozzle	
	Deposit	DUE	Deposit	DUE
Foliage	**1.27**	0.18	**0.92**	0.17
Stem	**0.23**	0.032	**0.15**	0.027

4.3.2 Effects of pulsing control

The effect of pulsing has been included as a factor in several of the following sections. In each of these experiments, the pulsing control has been shown to effectively reduce the volume sprayed ($P<0.001$, $F=624.30$, $df=2$, $n=84$, Figure 4.1).

However, it is clear that the reduction in volume is less than that indicated by the settings of the pulsing control, i.e. for a 75% reduction on the stem, only a reduction of 63% is observed. This discrepancy occurred throughout the majority of the trials and may be responsible for the significant interaction terms in parts of the minimal adequate model. For example, there was a significant interaction between nozzle type and pulse duration ($P<0.005$, $F=8.365$, $df=1$, $n=841$), indicating that there was a difference in the way that the two nozzles behaved with the different pulse settings. In this case, the AI nozzle showed results closer to the expected values.

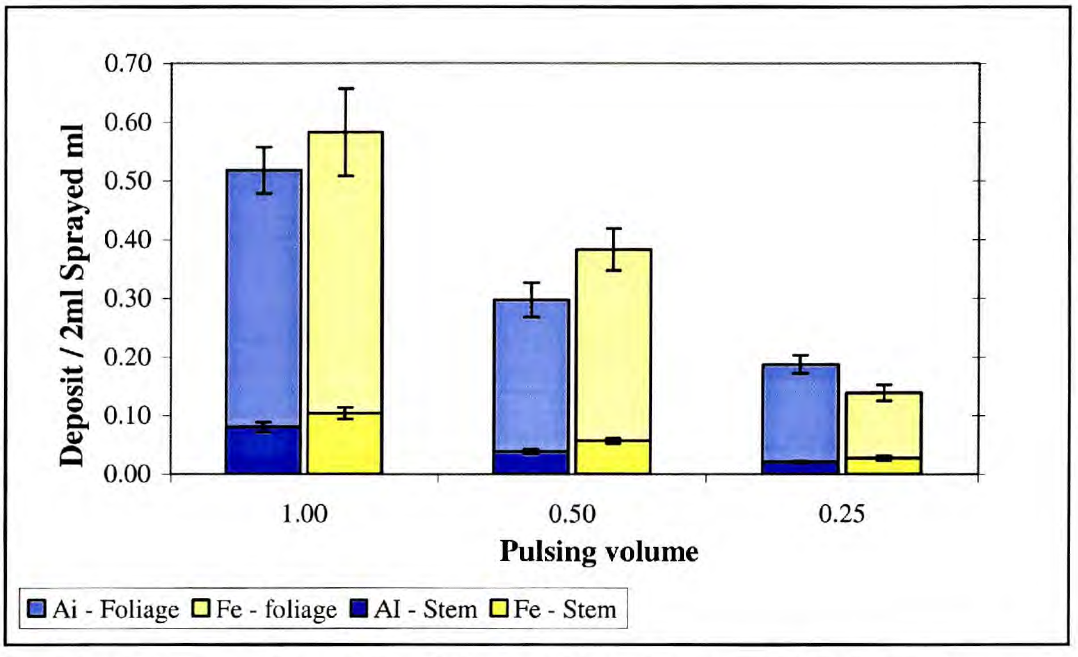


Figure 4.1 **Comparison of the deposition of dye onto Sitka Spruce stems and foliage with two nozzles at three different pulse settings. The deposition has been expressed as deposit per 2ml sprayed by the nozzles. The effects of pulse duration were highly significant ($P<0.001$, $F=624.30$, $df=2$, $n=841$).**

4.3.3 Effects of nozzle position

The effects of nozzle positioning on the deposition of dye onto Sitka spruce saplings were shown to be highly significant ($P<0.001$, $F=6.15$ (stem) $F=43.59$ (foliage) $df=2$, $n=841$) for deposition on the stem and the foliage. However, there was also a significant interaction between nozzle position and plant part ($P<0.001$, $F=19.24$, $df=2$, $n=841$), hence the non-significant main effects for position when foliage and stem were analysed together. This was because the highest and lowest deposition on foliage and stem occurred when different positions were used. Whilst the front position was very poor at whole tree coverage, it deposited the most on the stem (Figure 4.2a). In contrast, spraying from the side produced the exact opposite set of depositions (Figure 4.1). Spraying from a combination of the two positions (front/side) gave results similar to a front position but with lower deposition on the stem and slightly higher deposition on the foliage (Figure 4.2b).

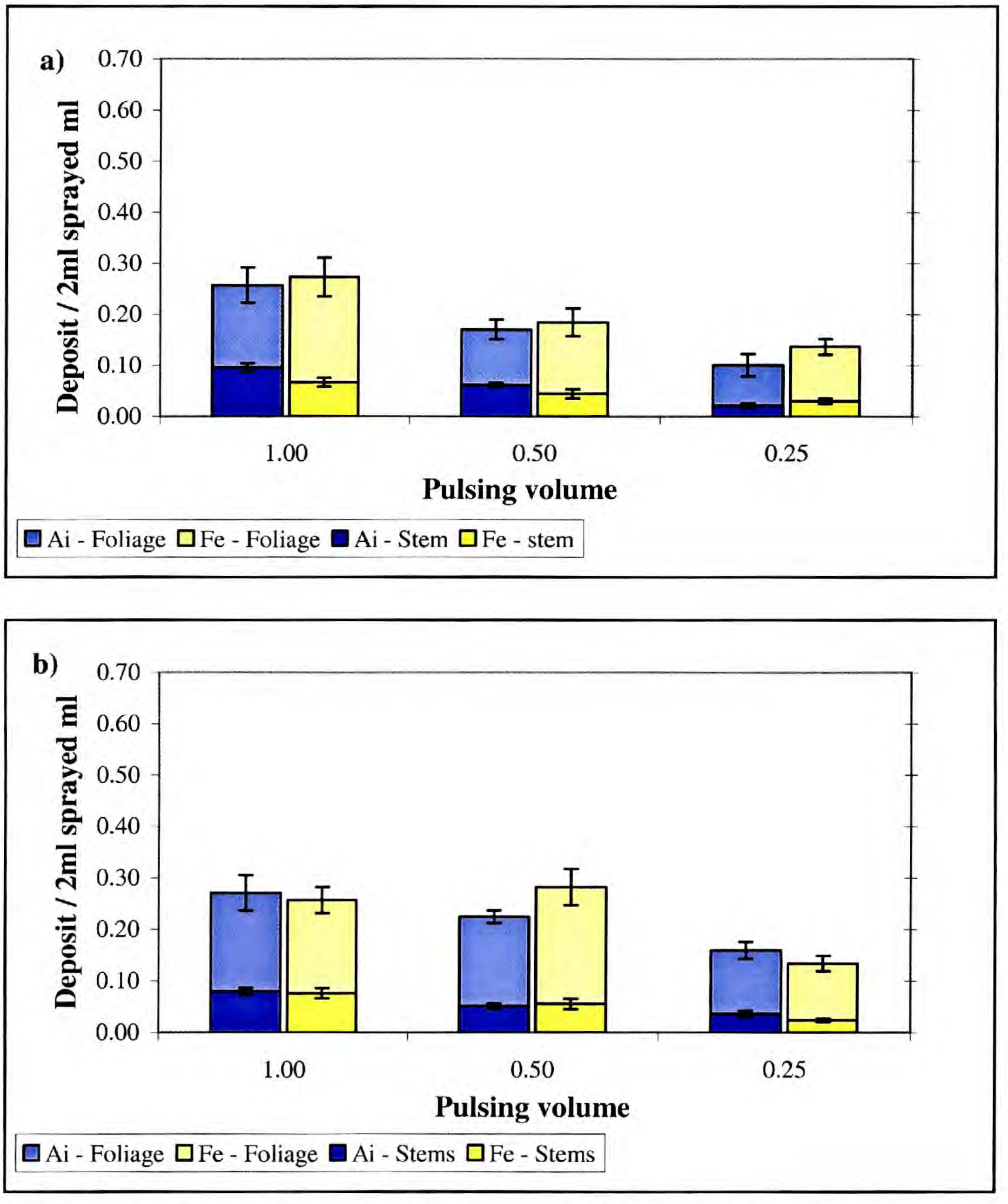


Figure 4.2 **Comparison of the deposition of dye onto Sitka Spruce stems and foliage with two nozzles at three different pulse settings when sprayed with the nozzles in a front (a) and front/side (b) position. Deposit is shown as deposit per 2ml sprayed by the nozzles. The effects of nozzle position were shown to be highly significant ($P<0.001$, $F=6.15$ (stem) $F=43.59$ (foliage), $df=2$, $n=841$).**

4.3.4 Effects of pressure

Despite the increase in flow rate induced by the three different pressure settings, there were no apparent differences in deposition (P=NS, F=0.70, df=2, n=841). The increase in flow rate caused by a 50% increase in pressure was 21%. This would indicate that an increase of 0.5ml for deposition on the foliage would be observed. The total increase in deposition was only 0.1ml (Figure 4.3).

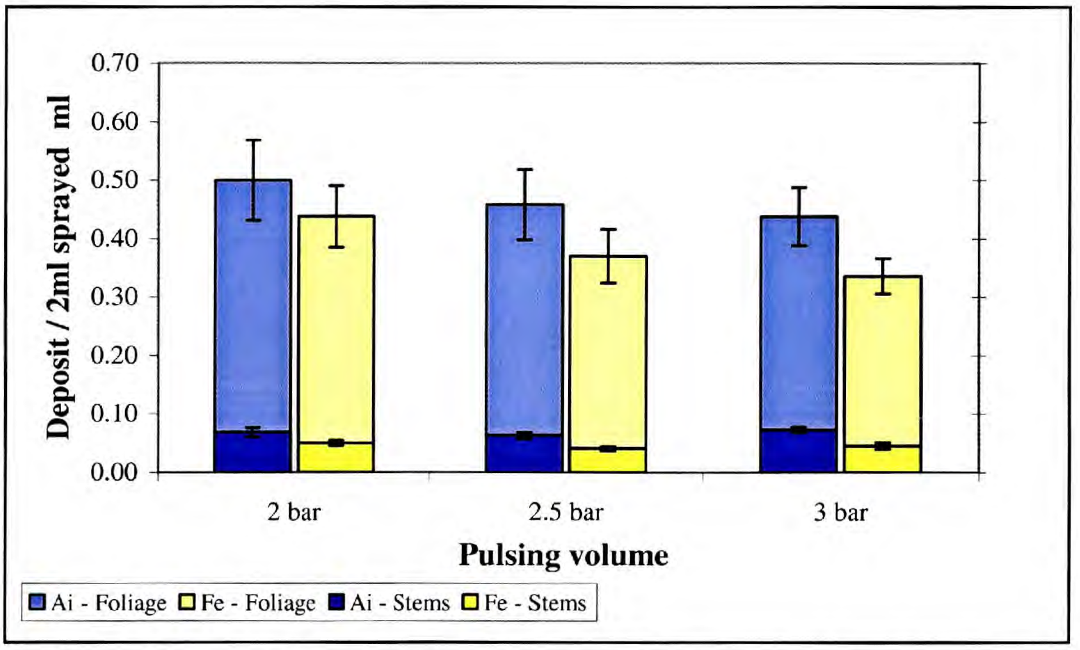


Figure 4.3 **Comparison of the deposition of dye onto Sitka Spruce stems and foliage with two nozzles at three different pressures. All trees were sprayed from the side. Deposit is shown as deposit per 2ml sprayed by the nozzles. There was no significant difference in the deposition on either the stem or foliage.**

4.3.5 Effects of speed

The penultimate trial examined the effects of operating the conveyor belts at different speeds. Four speeds in total were examined in two experiments. Initial analysis showed that speed was marginally significant (P=0.069, F=2.68, df=1, n=841). However, the setting of 23m min^{-1} appeared to be higher than expected, particularly for the AI nozzle (Figure 4.4). When analysed, data from both experiments were included (second data set not shown), the higher speed of 24m min^{-1} was in line with the expected volume of deposit. Therefore, the 23m min^{-1} treatment was removed and the data then showed a significant trend (P<0.05, F=6.40, df=1, n=841). This removal also highlighted an interaction between pulse duration and speed.

The removal of this treatment was warranted because it was expected that volume of the deposit would decrease if the saplings were in the spray cone for a shorter period of time and there were no substantial reasons for an increase in deposit. There was however, no significant difference in the effects of speed on stem deposit (P=NS, F=1.05, df=1, n=841) as can be seen in Figure 4.4.

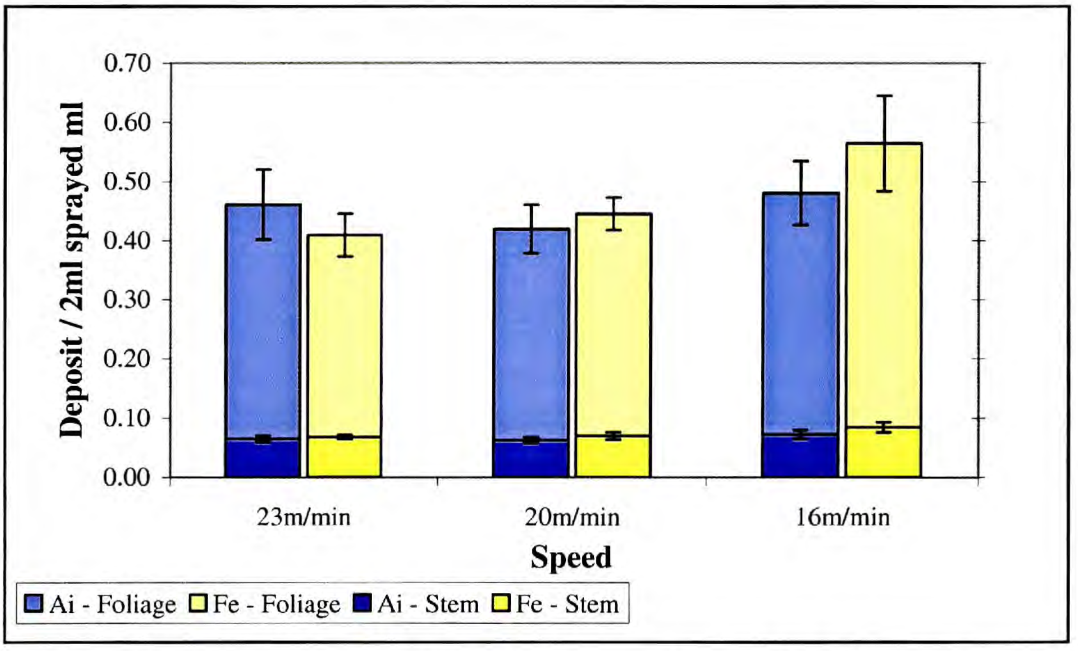


Figure 4.4 **Comparison of the deposition of dye onto Sitka Spruce stems and foliage with two nozzles at three conveyor belt operating speeds. Deposit is shown as deposit per 2ml sprayed by the nozzles. All trees were sprayed from the side. An increase in speed was shown to significantly decrease deposition (P<0.05, F=6.40, df=1, n=841) for the foliage only.**

4.3.6 Effects of nozzle position and spray duration

This trial was analysed individually and not part of the grouped data from the previous experiments. Previous data have shown that there may be some effect of changing the position of the nozzles. This trial aimed to highlight these effects.

The effects of position were found to be highly significant (P<0.05, F=5.21, (foliage) and P<0.001, F=47.50 (stem), df=1, n=229, Figure 4.5). It was noted during the experiment that saplings treated from the front nozzle position with the AI nozzle were substantially wetter than those with other treatments.

In addition to the effects of position, length of spray pulse showed a significant effect as was to be expected ($P<0.001$, $F=136.99$ (foliage), $P<0.001$, $F=149.43$ (stem), df=1, n=229). However, Figure 4.5 shows that only FE front and AI side treatments emulated the expected pattern for foliage deposition. The AI nozzle positioned at the front position consistently showed the same pattern of deposition on stems and foliage.

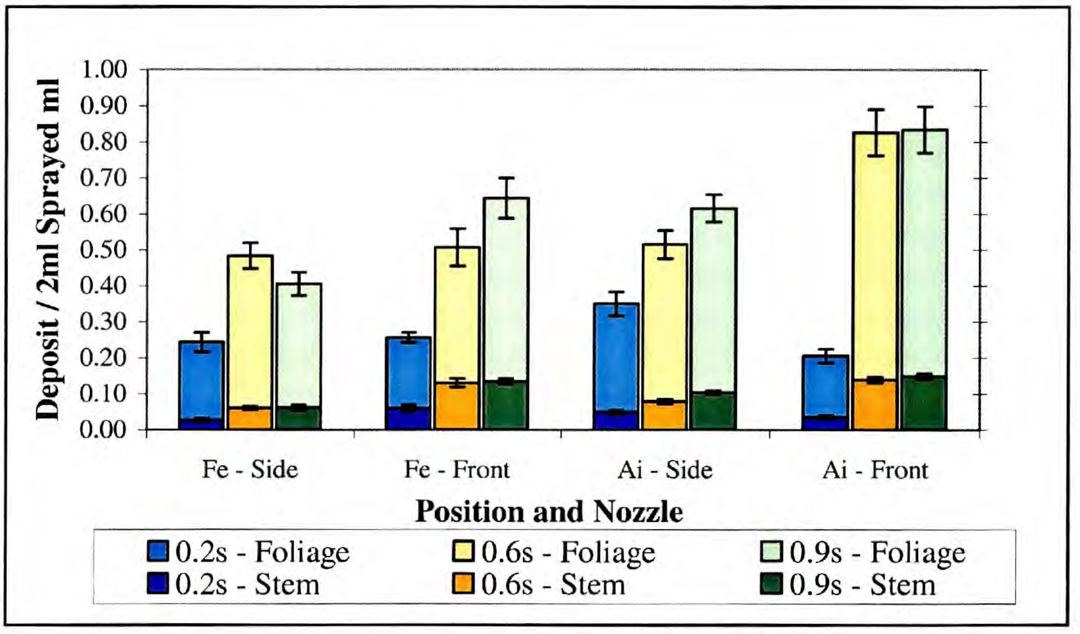


Figure 4.5 **Comparison of dye deposition on both the foliage and central stem of Sitka spruce saplings. Deposit is shown as deposit per 2ml sprayed by the nozzles. Duration of spray was highly significant ($P<0.001$, $F=136.99$ (foliage), $P<0.001$, $F=149.43$ (stem), df=1, n=229). Variation of the position was also significant ($P<0.05$, $F=5.21$, (foliage) and $P<0.001$, $F=47.50$ (stem), df=1, n=229). Log transformations were taken to normalise the data.**

The volume of deposit on the stem was higher when the nozzles were positioned in front of the oncoming tree. However, for both nozzles the longest spray period did not substantially increase the volume of deposit on the stem compared to the middle spray period.

Volumes of deposit within this experiment tended to be slightly higher than the previous experiments with a total deposit of 3.5ml for the AI nozzle from the front. A combination of factors may be responsible for the difference and are discussed later.

4.4 Discussion

The analysis of the data has revealed some interesting trends that were not otherwise apparent. In particular, grouping the data from several experiments has helped to increase the reliability of the results.

Firstly, it is interesting that whilst there was no significant difference between the two nozzles for foliar deposit, there was a significant difference in stem deposit. An increase in stem deposit without an increase in foliar deposit is very promising as it reduces risk of droplet transfer to the operator's gloves whilst increases the volume of pesticide on the target site for *H.abietis*. This difference is presumably due to the difference in droplet structure. The AI nozzle in use here has a small droplet range (span ~1.5) and less than 2% of droplets are smaller than 102μm diameter. Droplets smaller than this threshold are those that tend to remain airborne and are prone to drift, whilst even smaller droplets (~20μm diameter) are light enough to be inhaled and stay in the lungs (Matthews, 1992).

Droplet sizing measurements with the Malvern (Appendix 8) indicate that the Volume Mean Diameter (VMD) for the AI nozzle is 654μm whilst the FE nozzle produces a VMD of approximately 270μm. The values for the AI nozzle are similar to that found by Piggot & Matthews, (1999). In addition, the measurements have shown that there are approximately 0.5% of droplets below 32μm for the AI nozzle but 1.0% for the FE nozzle. Also there are up to 13% of droplets below 102μm for the FE nozzle but between 2% (Piggot & Matthews, 1999) and 5% (Appendix 8) for the AI nozzle. Therefore, the AI nozzle has a larger VMD and a lower proportion of the very small droplets than the FE nozzle. A difference in velocity of the droplets between the two nozzles, combined with the reduced possibility of the bounce off by the AI nozzle due to the air cushioning effect will lead to improved deposition (Matthews, 1992).

The pulsing system has been shown to be capable of reducing the volume of liquid sprayed to 40% of the original volume (Chapter 2, Figures 2.7 and 2.8). This is substantially higher than the 25% predicted by the manufacturers and is largely a consequence of the very low volume nozzles in use. However, it may be possible for a system to be built suitable for the low volumes in use on the DCS system. The experiments here showed that the deposition on the tree might be reduced to 36% of the original volume for the AI nozzle and 24% for the FE nozzle. This discrepancy is likely to be an artefact of the analysis since the sprayed volume for the FE nozzle at 25% pulsing is 1.62ml and the deposit is 0.45ml. Therefore, this difference in reduction between sprayed and deposit probably indicates that there is a loss in efficacy at the lowest pulse settings, when we consider that 40% is sprayed but only 24% is deposited. From this analysis it appears that the AI nozzle suffers less loss of efficacy.

The effect of pressure was not significant, which was somewhat surprising. There is a 21% increase in flow rate between 2 bar and 3 bar pressure. This would equate to nearly a 0.5ml increase in deposit for the AI nozzle, but only a quarter of this value was observed. Therefore, an antagonistic factor may be reducing the increased deposit in response to the increase in pressure. This may be increased bounce-off by droplets or increased production of smaller volume droplets, which may not deposit on the bark surface as readily. The AI nozzle had a higher deposit than expected by the increased flow rate alone, therefore, the larger VMD may explain this effect.

Due to the lack of difference shown, final choice of operating pressure should remain from a practical viewpoint. Therefore, a lower pressure (2 bar) will be chosen to avoid potential production of droplets less than 102μm in size. Any future experiments should continue to be at 2.5 bar so that experiments can be easily compared. An advantage of using the lower pressure setting is that the expected output of the system will be lower and therefore it will be easier to comply with maximum 'field rate' volumes imposed by the PSD.

The effect of speed was significant, but there is a clear trade off between deposition and total output of the system. The results presented indicated that the best deposition occurred at the lowest speeds, as expected. In this trial, the trees were within the spray cone for a minimum of 0.56 seconds even for the fastest speeds. Therefore the results are not attributable to trees only collecting a smaller proportion of the spray due to less time in the spray cone.

Future work should examine the limiting factors in tree handling and processing. The most obvious factor is speed of the conveyor belts, but operator handling time and tree preparation are equally important. If conveyor belt operating speed is not the limiting factor, the speed can be reduced, thereby increasing deposition and efficacy without affecting daily output.

The effect of position was examined twice and has been a factor in some of the experiments in Chapter 3. The initial trial showed significant interactions between position and plant part. There was a very good ratio of stem deposit compared to foliage deposit for the front position. The second experiment confirmed that the front position is very effective at increasing the deposit. In fact, the highest recorded deposits have been recorded from this position. This effect is particularly pronounced for the AI nozzle. It is believed that the earlier experiment had lower deposit from the front due to the difficulty in accurately positioning the nozzles and timing the spray pulse. Another advantage of this position is that it will minimise the number of times that the triggering of the spray pulse and arrival of the tree into the spray cone do not coincide. As the nozzles are aimed forwards, the tree will still receive a dose of pesticide even if the tree is not exactly in the correct position.

In contrast, the side position frequently mis-sprays passing trees, especially those that have long side branches or roots dangling in front of the tree. This effect was difficult to measure in a small experiment as the occurrence was fairly low and mostly resulted in a partial spray rather than a complete miss. However, this effect became evident when the differences between 0.6 second and 0.9 second spray were examined. The sapling only took 0.56 seconds to pass through the spray cone, but in most cases an increase in deposit was seen with the increase in spray pulse duration. A longer spray pulse duration will increase the chance that the sapling is sprayed for the entire time it is in the spray cone by allowing a margin of error.

Both nozzles showed an increase in efficacy with the frontal nozzle position and values as high as 41% (AI) and 32% (FE) were achieved on the foliage. Stem deposit was also increased and values of 7% (AI) and 6.5% (FE) were achieved.

This series of trials has examined the volume of deposit onto different parts of the tree. However, it has so far not examined the overall efficacy of the system. Data taken from the effects of pulsing trial, showed that the AI nozzle achieved a 26% level of efficacy for a 0.6 second spray pulse from the side. Similar values were observed in the trial effects of position trial. In addition, there was approximately a 4% level of deposit onto the stem, again repeated in both trials. However, the FE nozzle was less consistent across the two trials with foliage deposit ranging from 24-29% and stem deposit ranging from 3-5.5%.

The other trials generally showed similar levels of efficacy and volume of deposit, although they were sometimes lower. The variation observed was in part due to the variation in tree morphology but also in the difficulty in accurately targeting the trees. This is obviously a key problem and an area that will be addressed. Steps have already been taken to reduce the variation, one of which is to spray the saplings from the front, as this is believed to increase the reliability of the triggering system. Future developmental work will focus on a more accurate trigger, but also a more precise way of aiming the nozzles will be greatly beneficial.

In summary, it is believed that to achieve a low volume of deposit in conjunction with a high efficacy, that trees should be treated from the front with an AI nozzle at 2 bar pressure and at a speed of 16m min^{-1} and a spray pulse of 0.4 seconds. This combination gives not only the best deposition, but decreases the volume of pesticide used, maximises efficacy and increases reliability.

Chapter 5
Operator Safety

5.1 Introduction

The Dandrive Conveyor Sprayer system (DCS) combines heavy-duty mechanical equipment with pesticides in an enclosed environment. This is a situation where safety issues should be of foremost concern to the operator. The two large timing belts have the potential to trap and injure the operator's arm or hand, whilst the levels of pesticide spray droplets or vapour in the air may pose a risk to other aspects of the operator's health. It is of importance, therefore, that the risk the system poses to an operator is quantified before it is put into large-scale use.

The risks of injury through mechanical incident can be virtually eliminated with the use of some simple components. For example, the use of a slipping clutch, which is connected to an alarm and the electric motor (Section 2.2.1), will ensure that a trapped limb is quickly registered by the system resulting in an automatic shutdown.

There are also less obvious, but potentially longer term effects that the system may have on an operator's well being, such as being forced to stand in an awkward posture for long periods. For example, the operator may have to stoop if they cannot clearly see their hand actions. Alternatively, a shorter person may have to work with their arms at an uncomfortable height. These potential health risks can be minimised by ensuring that the design of future systems are based on human ergonomics and not on material structure and availability, as was the case with the prototype. Automatic tree feeders could potentially make the system safer still by reducing the involvement of the operator with the system.

The main element of risk in this system concerns the use of pesticides. Exposure to pesticides can occur very easily at a range of levels. For example at the operator level, users may touch their hair or face with contaminated gloves. Improper protective clothing may result in pesticide residue building up on personal clothing, and finally there is the risk that the system does not safely contain the pesticide during spraying or mixing. Therefore, the safety emphasis should be on reducing exposure by maximising containment of the pesticide, coupled with good operating procedures so that contact with treated surfaces is kept to a minimum.

Previous work has shown that dermal exposure can depend on a variety of factors, from the pesticide type, to the volume of dislodgeable residue (Brouwer *et al.*, 1992) and finally even the size and surface type of the hand (Matthews, 2001). This study demonstrated that the maximum volume of water that can be retained on the skin is no more than 2.5ml as the water will only form a thin film on the skin. In a second experiment up to 10ml could be retained if droplets were not allowed to run off. Another study has demonstrated that around 6ml may be retained on the hand, although this experiment only used one subject (Velsar, 1984).

Flower culture in glasshouses involves handling procedures very similar to the DCS system. Flowers are hand-cut and collected into bundles held in the arms, therefore pesticide exposure is confined mainly to the arms and hands (Brouwer *et al.*, 1992). In this system, the volume of dislodgeable residue was strongly correlated to dermal exposure. In particular, use of a low volume mister instead of standard high volume sprays reduced dermal exposure. Therefore, nozzle selection for the DCS should consider these factors.

With these precautions and procedures in mind, this section attempts to quantify the risk of exposure and assess the volume of pesticide that may escape the system. It is not meant as a series of experiments so that the system may pass a safety test. It is instead intended to admit to the shortcomings and risks, quantifying them so that they can be dealt with in future models. In particular, the use of an extractor fan system is examined in relation to exposure risk and effects on tree deposit.

5.2 Use of an extractor fan system to remove excess airborne droplets

A centrifugal fan was installed into the DCS system to expel air via ducting, thereby reducing the risk of operator exposure. Although most of the excess pesticide spray (that which misses the tree) lands on the plastic panel opposite the nozzle, there are a small proportion of droplets that remain airborne and could potentially drift out of either end of the conveyor. This is most likely to be those that are below 102µm in diameter (Piggot & Matthews, 1999). This was considered to be a greater problem for the FE flat fan nozzle, which has a much higher proportion of small droplets than the air induction nozzle (Appendix 8 and Section 4.4). Although nozzles of the same flow rate were used, the AI nozzle produces a coarser spray and as the angle was much wider (120° compared to 60°), there was effectively more space between droplets. Therefore, when the two nozzles are aimed at each other most droplets are involved in nothing more than high speed passes. Contrary to this, the FE nozzle has a much finer spacing between the very small droplets with the consequence that when the nozzles are aimed at each other, the opposing flow of spray creates turbulence causing the smallest droplets to form a spray cloud. A proportion of the spray then drifts to either end of the conveyor rather than in the intended direction. This problem is increased when the nozzles are aimed towards the oncoming tree, rather than perpendicularly to it, unfortunately this was also when the best tree deposits were achieved (see Chapter 4).

In the Electrodyn system, an overhead extractor hood was fitted to minimise exposure to the oil droplets. However, a hood in this position may not be the most suitable solution in the DCS system, because positioning the hood above the nozzles may affect the deposition. It was not possible to install such an expensive part on the prototype, so a centrifugal fan was connected via a splitter junction and to two sections of ducting, which served as a dual inlet. The inlets were positioned either side of the conveyor belts near the front opening.

5.2.1 Materials and methods

A series of quantitative observations were made of these phenomena in action. Two methods were used to record the relative amounts of droplet escape, by photography and by the use of water sensitive paper. The escape was classed as droplets leaving the small opening at the front of the conveyor. This is directly in front of the position where an operator's upper body would be, so is of particular importance. In addition, these measurements were made with the extraction system installed with the openings to the vents at different places inside the conveyor system.

To assess the extent to which the droplets might escape through the opening of the DCS system where trees are fed in, a piece of water sensitive paper was held in front of the opening in the front panel as described by Turner and Huntingdon (1970). Each piece of water sensitive paper was held in position for the duration of three operations, each lasting 0.9 seconds. This is the equivalent of treating approximately 7 saplings.

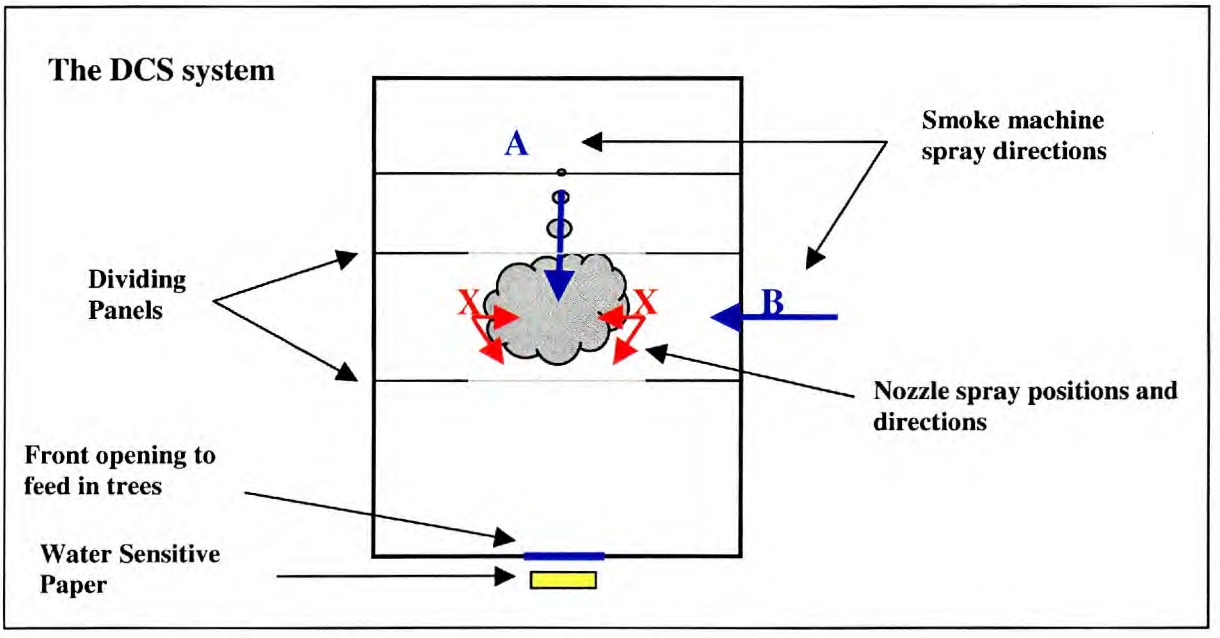


Figure 5.1 **An outline of the DCS system indicating the direction of the smoke and fluorescent spray when examining the system for containment. The extractor fan is not shown.**

A second test was also carried out using smoke generated from a special effects smoke machine (Atari). Although the smoke was lighter than many of the droplets, it was easy to visualise and supplemented the previous experiment (Figure 5.1). The extractor fan ducting was positioned in a range of positions and the smoke patterns were recorded with and without the extractor fan in use.

5.2.2 Results and analysis

Figure 5.2 and Figure 5.3 show the results obtained from the two nozzles. The most striking difference was that between the FE and AI nozzles. The FE nozzle consistently produced clouds of droplets that escaped the system, whilst the AI nozzle resulted in only a few larger droplets escaping. Secondly, the use of the extractor fan had a far greater effect on the escape of droplets produced by the FE than it did for the AI nozzle. However, in some cases the extractor fan seemed to make the level of escaped droplets substantially worse such as in Figure 5.2, cell E and Figure 5.3 cell I.

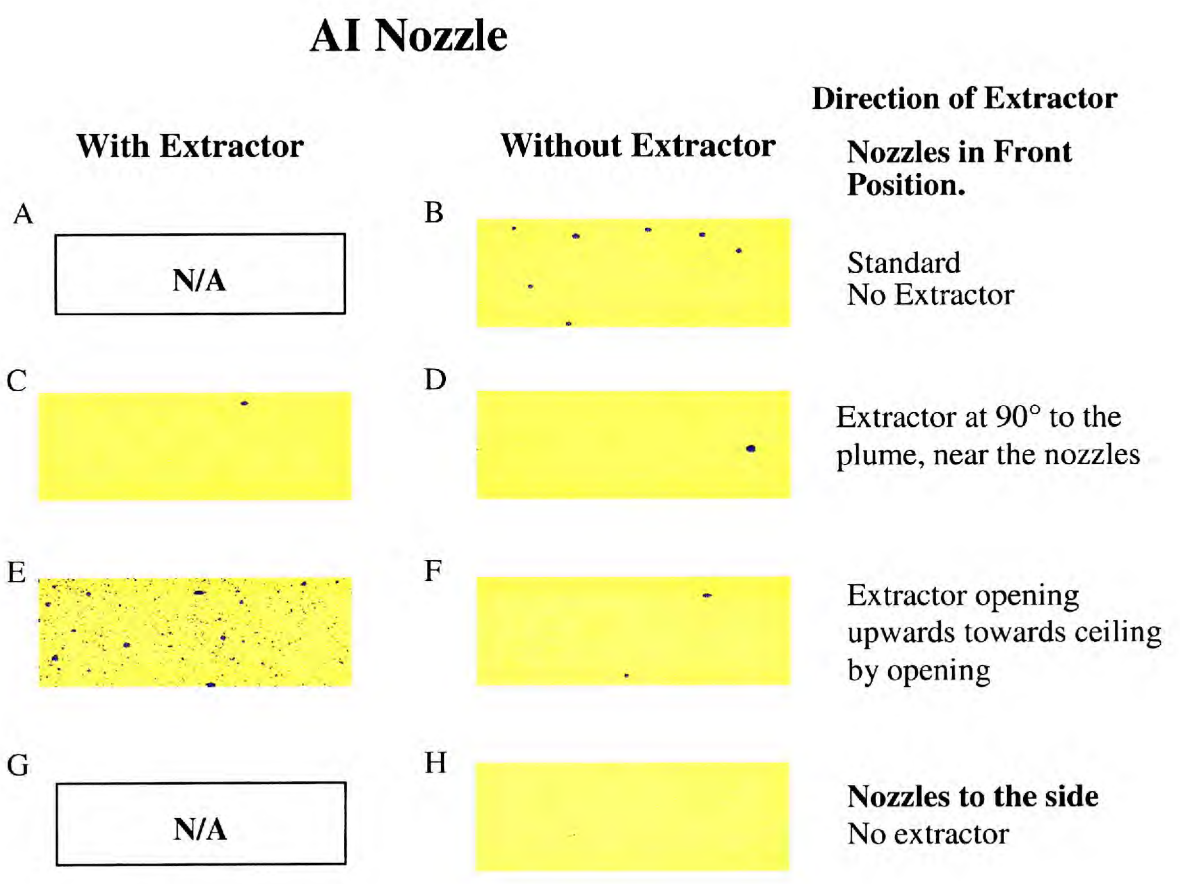


Figure 5.2 **Use of water sensitive paper to quantify the volume of escaping droplets with the AI nozzle. In the majority of cases there was very little droplet escape and this was marginally reduced by the use of the extractor fan. However, positioning of the extractor fan below the entrance (E) caused an increase in droplet escape.**

Figure 5.2 shows that there was a low level of large droplets escaping through the opening of the conveyor for the AI nozzle with no extractor fan in place (cell B). Addition of the extraction system reduced this volume even when it was turned off (cell D). This was most likely due to the ducting which was positioned either side of the central canal pointing at right angles to the line of tree movement. It was believed that the ducting acted as a shield and it was for this reason that little difference could be seen between the two cells C and D.

FE Nozzle

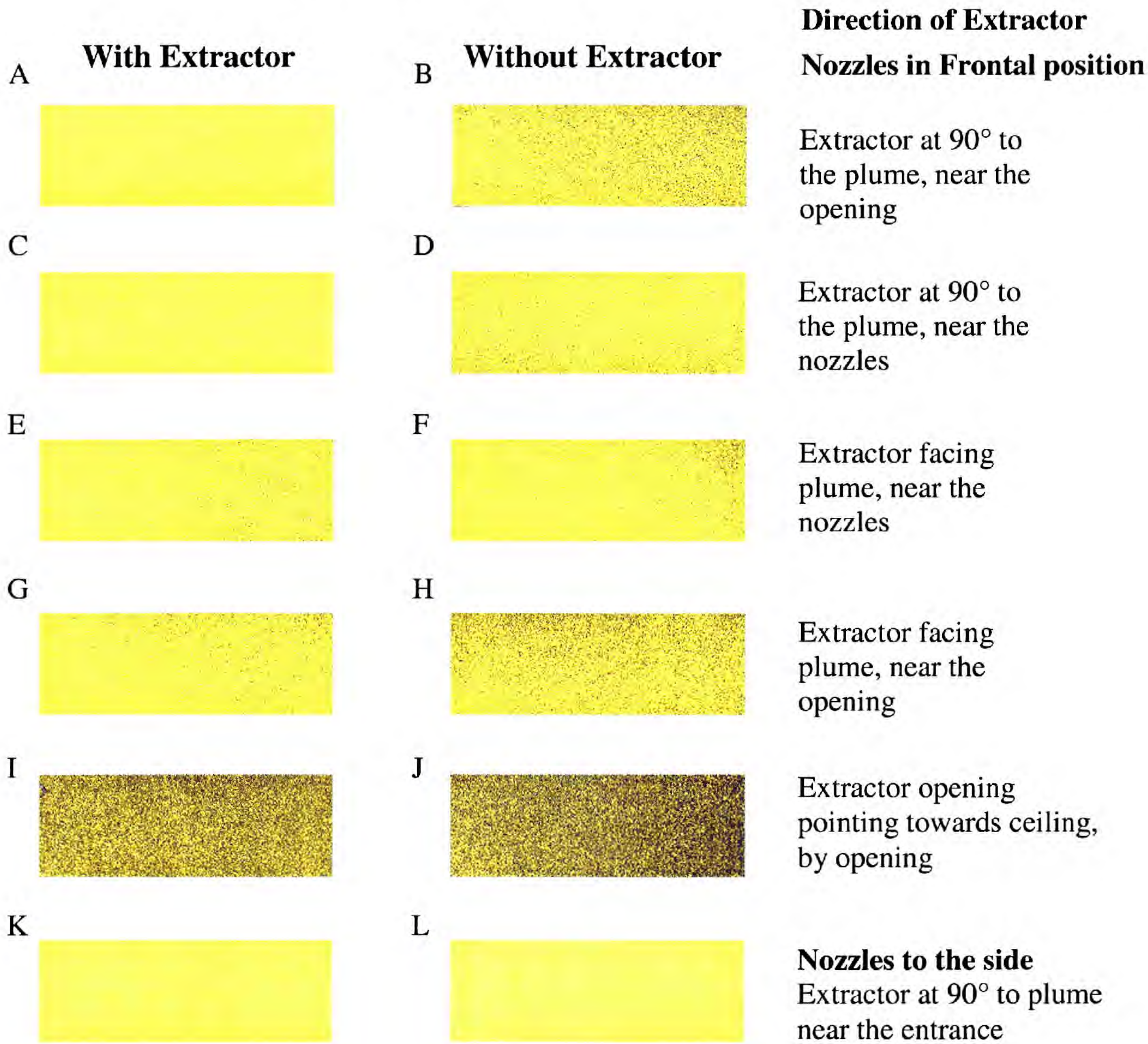


Figure 5.3 **Use of water sensitive paper to quantify the volume of droplets escaping from the opening on the DCS system with the FE nozzle. In the majority of cases there was substantial droplet escape, but this was greatly reduced by the use of the extractor fan. However, positioning of the extractor fan below the entrance (cell I) caused an increase in droplet escape.**

The ducting openings were also positioned directly below the opening pointing upwards in an attempt to pull escaping droplets downwards into a collision trajectory with the panels or into the ducting itself. However, it appeared to have the opposite effect for both nozzles and it is probable that the air currents drew airborne droplets towards the opening but did not hamper their escape. This effect was especially evident with the FE nozzle (Figure 5.3, cells I and J). It was also clear that the positioning of the ducting in this position, even when the extractor was switched off, enhanced escape. However, examination of the methods used and order of treatments highlighted the probable cause. The measurement for the 'with extractor' was taken first and the 'without extractor' second. When a second replicate was taken for the 'without extractor' several minutes later, the deposition on the water sensitive paper was very similar to that of the other 'without extractor' readings. Therefore, it appeared that the air currents inside the DCS system were maintained for a few minutes after the extractor fan was switched off.

The final measurement taken for the AI nozzle was with the nozzles positioned at right angles to the line of tree movement. Only 'without extractor' results are shown (Figure 5.2, cell H), but there was no escape of droplets in either case. Therefore, modification of position remains a very important aspect of reduction in risk of exposure.

The FE nozzle was tested more extensively because of the more dramatic results. Unsurprisingly, it was immediately obvious that the smaller droplets were more easily affected by the air currents. The ducting was positioned in a total of four positions, near and far from the entrance and at right angles or head on to the spray cloud.

Distance from the entrance (near or far) appeared to have an effect on the level of droplets escaping the system (Figure 5.3, cells A and C). When the ducting was placed head-on to the oncoming cloud near the entrance, there was a greater reduction in escape than when placed nearer the nozzles, albeit a slight one. It may be that positioning the extractor near the entrance drew all of the droplets away from the opening, but nearer to the nozzles some droplets got past the extractor as they had a higher velocity and could then escape. However, this is of little relevance when the volume of droplets escaping was considered, which was still too high. It is probable that the flow of air drew as many droplets towards the opening as it drew in to the ducting. Therefore there was little difference seen with and without the extractor fan in operation.

The alternative position (at 90° to the plume) yielded excellent results and although little difference could be seen in the scanned images above, the least escape occurred when the ducting was nearest the opening (Figure 5.3 cells A-D). In fact, there was no escape at all seen in any of the replicates for cell A. The difference between the two distances may be best explained by the velocity of the droplets, which was greater nearer the nozzles and therefore allowed them to escape the pull of the extractor fan. In addition, when the ducting was near the opening, the extractor fan very effectively sucked in the droplets or deflected their trajectories substantially so that they would not escape through the opening.

Finally, the FE nozzles (not the extractor fan) were positioned at right angles to the conveyor belts, as described for the AI nozzles. Even though there was a substantial reduction in escaped droplets, there was still a substantial volume escaping. In this position though, the nozzles gave the cloud no direction, as it would have with the Front position and equal levels may have escaped from the rear end. When the extractor fan was positioned at right angles to the conveyor belts (the best position from above), the escape of droplets was negligible.

The use of smoke generated by a smoke machine revealed many of the patterns described above. Although the smoke particles were much lighter than most droplets, many of those produced by the FE nozzle in particular, were small enough to remain airborne for several hours (Matthews, 2000). The movement of the smoke was recorded with and without the extractor fan in use (Figure 5.4). When the extractor fan was not in use the smoke quickly filled the interior of the DCS system. When the extractor was aimed directly towards the entrance, large clouds of smoke escaped through the front opening. This effect was reduced if the smoke machine sprayed at right angles to the conveyor belts, which is the equivalent of the nozzles spraying from the side position.

The use of smoke to demonstrate air movements gave similar results to the use of water sensitive paper when the extractor fan was in use. Positioning of the extractor fan at the front of the conveyor with the opening of the ducts pointing to the ceiling only served to accelerate the smoke towards the exit (Cell A). Very little smoke was drawn in to the ducts and large clouds escaped through the front opening.

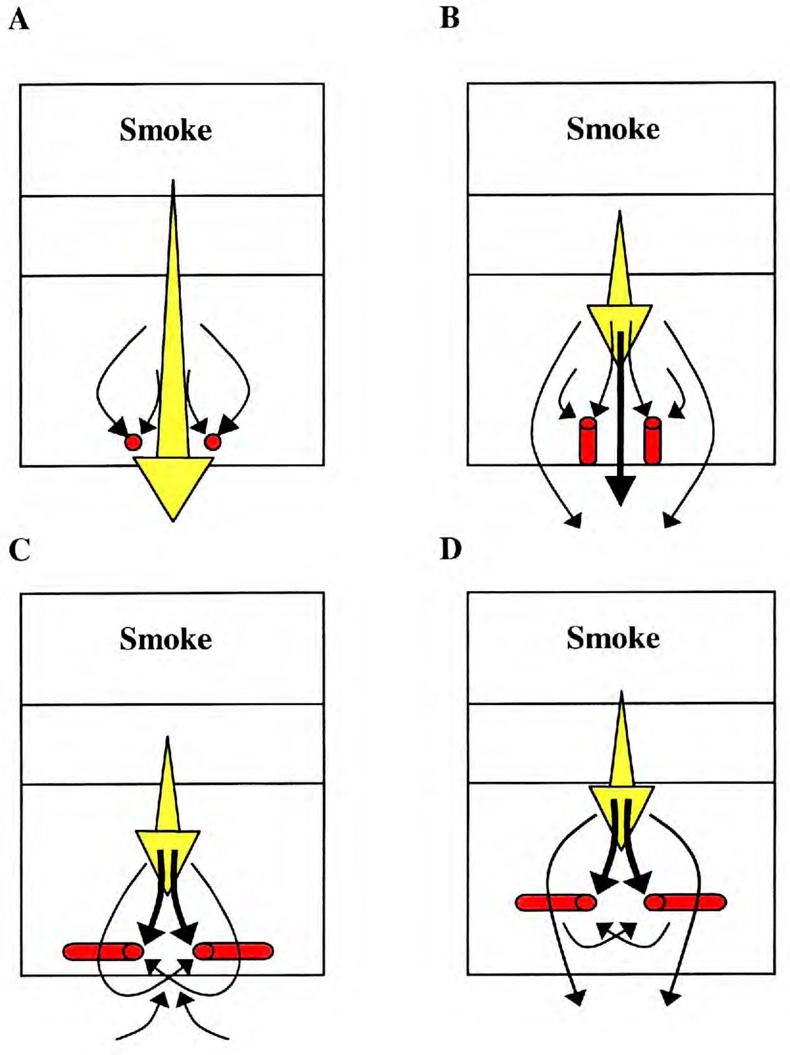


Figure 5.4 **Air currents revealed by use of smoke. The thickness of the arrows gives an indication of the approximate volume of smoke. The length of the yellow arrows gives an indication of the approximate acceleration of the smoke. The extractor fan position is shown by the red cylinders and circles. See text for full explanation.**

Positioning the extractor fan ducts head-on to the smoke plume was also quite ineffective (Cell B). Although some smoke was drawn into the ducts, substantial amounts moved either around the ducts or between the two of them. Once past the ducting the smoke escaped through the opening.

The final position placed the ducting at right angles to the smoke plume and was far more effective than any of the other positions. Cells C and D reveal the air currents that are probably responsible for the low leakage of both smoke and spray. Positioning of the ducting further away from source (Cell C) was more effective than when it was close to the source (Cell D). This was because the smoke was retained by the front panel. However, it was not able to escape through the opening because this area was within the range of 'pull' of the extractor fan. It was also seen that smoke outside the DCS system was drawn into the interior, indicating a slightly negative pressure inside. This would also help to contain both the spray and smoke. In contrast, when the ducting was closer to the source some of the smoke was able to get past the pull of the extractor and was not impeded by the front panel. It was then free to escape from the entrance.

5.2.2 Summary

A particular set-up of nozzles or positioning of the nozzles may give enhanced deposit, but this is not necessarily confined to the tree, which is unfortunate when a trade-off is required between maximising the deposit and the safety of operators. The DCS set-up must first respect the safety aspect and secondly the deposit. However, it has been shown that careful positioning of an extractor fan with a pair of ducts can reduce that risk to near zero, as demonstrated through the use of both smoke and spray. This mainly applies to the use of the FE nozzle, as the AI nozzle was not so easily influenced by the extractor system, presumably due to the larger droplet size (Appendix 8). Therefore, there is a choice regarding the reliance of an extractor fan system to remove the droplets produced by the FE nozzle, or the relatively safer use of the AI nozzle. It must be remembered that the AI nozzle is used to reduce drift in the field, but it also appears effective in this situation. If safety issues are equal when comparing the AI nozzle with the FE nozzle and the extractor fan, then the maximization of deposit should be considered as the next most important factor.

5.3 Potential detrimental effects of the extractor fan on deposition

The extractor fan was shown to be extremely effective at reducing the levels of escaping droplets produced by the FE nozzle. However, the effects it had on the larger droplets produced by the AI nozzle were less noticeable. Unfortunately, the extractor fan may have some negative effects on deposition, particularly for the very small droplets that are most prone to drift. A small trial was set up to test for any differences in deposit that may arise from the use of the extractor fan.

5.3.1 Materials and methods

The DCS system was used to spray 80 two-year-old (1+1) Sitka spruce with a solution of 0.25g l^{-1} sodium fluorescein from a front nozzle position at 2.5 bar. The Sitka spruce were divided into four treatments for two nozzles (air induction (AI) and flat fan (FE)) and additionally with the extractor fan on and off. The extractor fan was positioned near the entrance at right angles to the line of tree movement as this was shown to be the most effective position in the previous experiment. A few minutes between treatments was allowed to give the air currents time to establish or dissipate.

The whole treated tree was then subjected to the wash procedure described in Appendix 7. Samples of the wash solution were then analysed in a spectrofluorimeter and the results analysed using Anova in S-plus.

5.3.2 Results and analysis

There was a significant decrease in the volume of deposit with use of the extractor fan ($P<0.05$, df=1, F=5.8, n=40). In addition, the nozzle type and plant part had a significant effect on the deposit ($P<0.001$, df=1, F=24.8, n=40 and $P<0.001$, df=1, F=114.7, n=40, respectively). This followed similar trends to the results found in Chapter 4. There was also a strong interaction between nozzle and plant part ($P<0.001$, df=1, F=13.55, n=40).

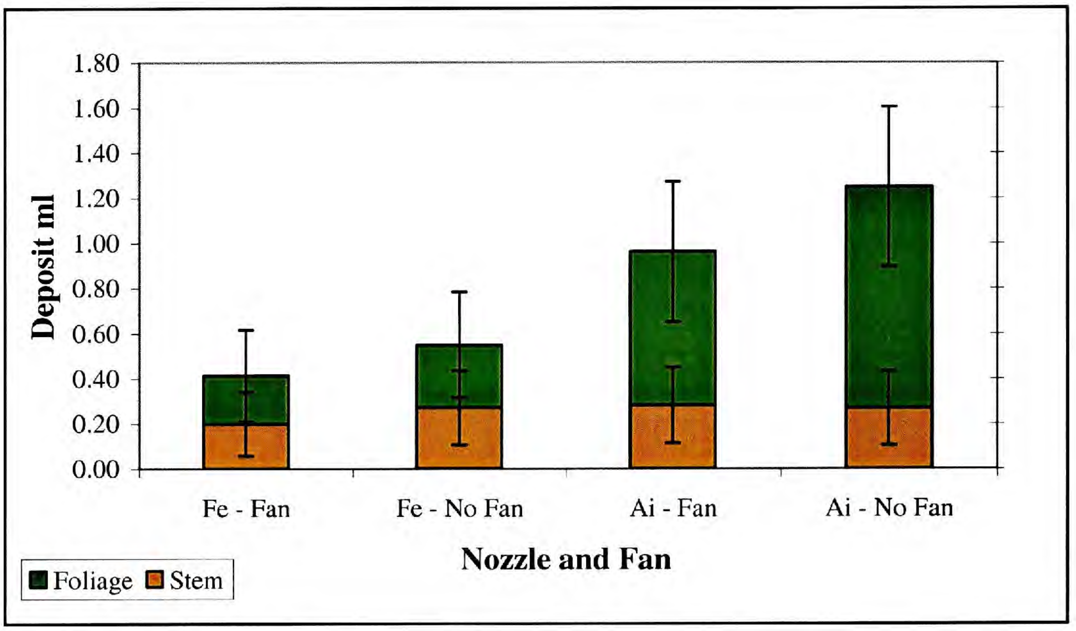


Figure 5.5 **Effects of the extractor fan positioned near the entrance of the DCS system on the deposit of dye onto Sitka spruce saplings. There was a significant effect of both nozzle ($P<0.001$, df=1, $F=24.8$, n=40) and use of the extractor fan ($P<0.001$, df=1, $F=114.7$, n=40).**

The use of the extractor fan decreased deposit by approximately 0.2ml on the foliage for both nozzles (Figure 5.5). However, for the stem the deposit was only reduced for the FE nozzle and increased for the AI nozzle.

5.4 Potential pesticide exposure from treated trees

Removing potential escape of pesticide by use of an extractor fan system is one important way of decreasing the risk of operator contamination. However, there is still an inherent risk in the handling of the treated trees as they exit the system. For the Electrodyn system, this is less of a problem as only 0.2ml of pesticide is applied to each tree, but for the DCS as much as 3ml may be applied. However, the Electrodyn applies pesticide at a concentration of 25% and any dermal contact would be more likely to cause side effects such as a local allergic reaction. The Electrodyn treated trees are very nearly dry after being treated and therefore the dislodgeable residue is reduced and the risk of pesticide transfer onto the operator's gloves is minimal.

At the other extreme, the dipping process completely soaks the trees. However, the trees are not handled until they have been dried, unlike the very early dipping systems that involved dipping bundles of trees by hand. In this process, the operator would pick up substantial pesticide residue on their gloves. Instead exposure from this system may occur through splashing as the trees are lowered in to the vat of pesticide.

The operator of the DCS system will be wearing protective gloves, a rubber apron, coveralls and possibly a facemask in accordance with the Pesticide Safety Directorate specifications (Heritage *et al.*, 1997a). However, currently there is a greater concern to reducing reliance on personal protective clothing and installing engineered control methods. Stringent protective measures must therefore be incorporated into the DCS system. In conjunction with these safety measures, it is also necessary to precisely quantify the volume of pesticide that an operator may pick up on their gloves in the routine handling of treated trees. This next section examines this aspect by measuring the volume of a fluorescent marker on the gloves.

5.4.1 Materials and methods

This section is comprised of three parts, using similar methods. These three parts were:

- Relationship between number of trees handled and contamination
- Contamination in conjunction with surfactant
- Operator handling efforts to reduce contamination

5.4.1.1 Relationship between number of trees handled and contamination

One hundred two-year-old (1+1) Sitka spruce were used. Each of these were passed through the DCS system and sprayed only once with a sodium fluorescein solution (0.25g l^{-1}) using either a FE or AI nozzle at 2.5 bar with the nozzles in a front position. Trees were treated in batches of 20 and a portion of the 20 trees were handled and put to one side. This was repeated for each of the five treatments; 1, 3, 5, 10, 20 and 50 handlings per batch. Ideally new trees would have been treated and handled for each treatment, but this would require over 200 trees and the resources were not available.

A tree was handled by grasping it firmly above the root collar in the sprayed zone by an operator wearing a latex glove. The glove was then carefully removed and placed into a plastic resealable (22.5cm x 15cm) bag. Water (250ml) was added and the bag shaken for 20 minutes in an automatic shaker. A sample of the wash solution was then removed and the fluorescence measured and recorded in a spectrofluorimeter (As described in Chapter 4 or Appendix 7).

5.4.1.2 Contamination in conjunction with surfactant

For each treatment 28 two-year-old (1+1) Sitka spruce were used. There were two factors each with two levels; nozzle (AI or FE) and surfactant (with or without), giving a total of four treatments. Within each treatment a latex glove was used to handle four treated saplings (as described above). The saplings had been treated with a solution of 0.25g l^{-1} of sodium fluorescein with each nozzle spraying at 2.5 bar pressure with the nozzles in a Front position. The latex glove was then carefully placed into a resealable plastic bag (22.5cm x 15cm) and the fluorescence of the wash solution measured as described above.

Non-surfactant treatments were carried out first to reduce the risk that the surfactant would not be properly cleared from the pipes and nozzle system. Surfactant (Agral) was added to the dye solution, giving a final concentration of 0.01% (v/v). To remove any possible enhanced washing effects that the surfactant treated trees may have had, surfactant was added at a rate of 0.02ml per bag to 'swamp' the surfactant that was on the tree or glove. This was to ensure that all trees/gloves would be washed equally. The trees and gloves were then washed and the fluorescence of the wash measured as described above.

5.4.1.3 Operator efforts to reduce contamination

One hundred two-year-old (1+1) Sitka spruce were treated with a solution of 0.25g l^{-1} Sodium fluorescein at 2.5 bar pressure with the AI nozzle. There were only two treatments; trees handled carefully and trees handled carelessly. Carefully handled trees were delicately held at the tip and by the roots with two hands, whilst carelessly handled trees were gripped tightly in the treated zone with one hand (as described above). For every four trees one or a pair of latex gloves were used. Only the gloves were washed and analysed for measurement of contamination.

5.4.2 Results and analysis

5.4.2.1 Relationship between number of trees handled and contamination

There was a significant difference in volume deposited on the gloves between the AI and the FE nozzles ($P<0.05$, df=1, F=4.8, n=60), with the FE nozzle causing less contamination on the latex gloves. The relationship between the number of trees handled and the level of contamination showed a cumulative decrease of deposit with increase in number of trees handled ($P<0.001$, $R^2=0.77$, n=60, Figure 5.6).

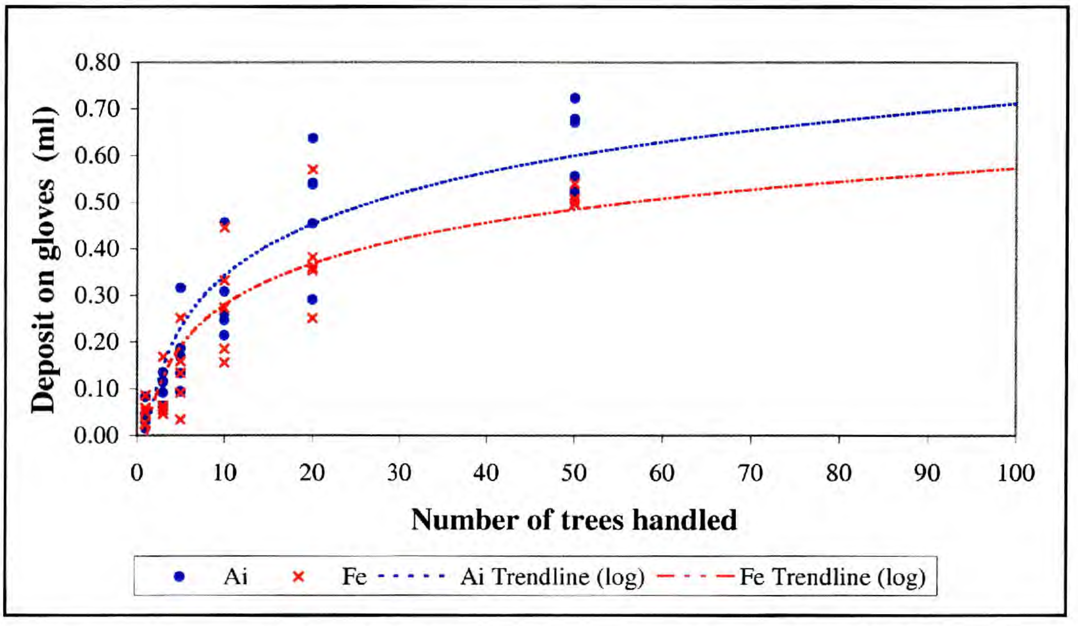


Figure 5.6 **Relationship between the number of treated trees handled and the volume of dye deposit on an operator's gloves. There was an exponential decrease in deposit with an increase in the number of trees handled.**

Figure 5.6 shows an extrapolation of this relationship and shows that increasing the number of trees handled to 100, results in a relatively small increase in deposit on the gloves. When extrapolated to 1000 trees handled, the level of deposit is only 1.08ml for the AI nozzle and 0.85ml for the FE nozzle.

5.4.2.2 Contamination in conjunction with surfactant

There were significantly different volumes of deposit on the saplings between both the AI and the FE nozzles and also between the use of surfactant and without it. This was evident in the highly significant interaction term between the nozzle type and use of surfactant ($P<0.001$, $df=1$, $F=25.2$, $n=104$). This can be clearly seen in Figure 5.7 and is discussed later. In addition, there was significantly more deposit on the tree than the gloves ($P<0.001$, $df=1$, $F=382.3$, $n=104$).

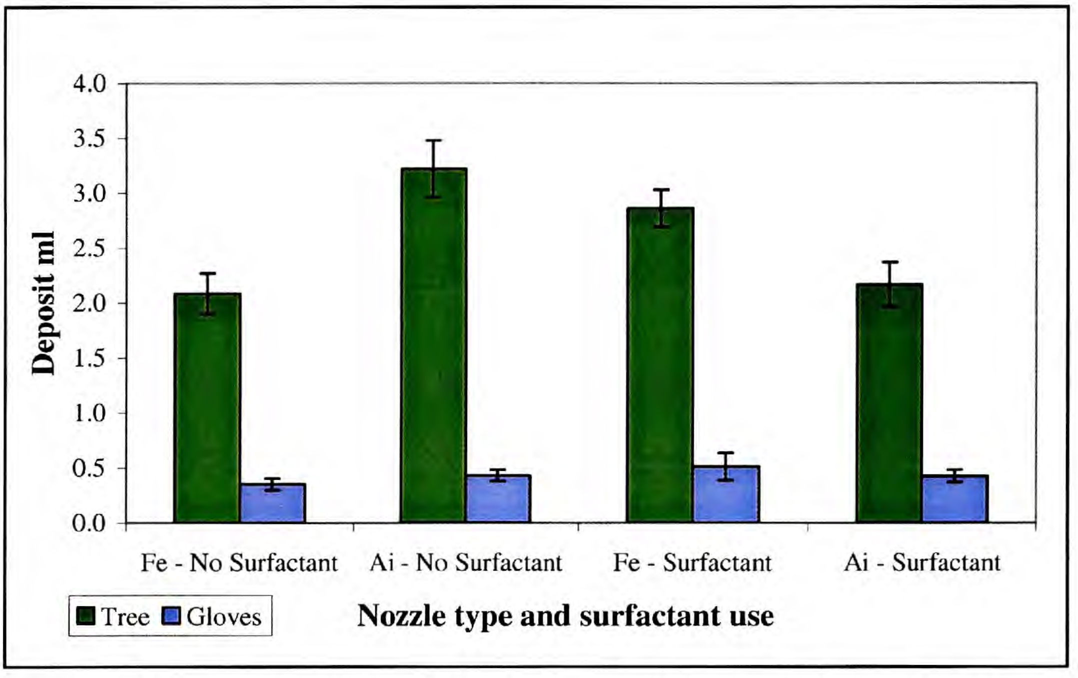


Figure 5.7 **Comparison of the deposit on Sitka spruce saplings and on the operator's gloves after handling four trees. There was a significant interaction between the nozzle type and use of surfactant ($P<0.001$, $df=1$, $F=25.2$, $n=104$). Surfactant reduced the deposit on the tree by the AI nozzle, but increased it for the FE nozzle.**

5.4.2.3 Operator efforts to reduce contamination

Typical deposits of around 0.5ml have been shown to accumulate onto the operator's gloves after handling several trees. However, in both of the previous experiments the operator firmly grasped the treated area of the tree. This was a poor and unsafe method of handling trees and would result in higher than normal contamination. This experiment addressed whether safe and sensible handling of the treated tree made a difference to the level of contamination.

Figure 5.8 shows the volume of deposit by contamination for each of the handling types. There is a clear and significant difference between the two methods (P<0.001, df=1, F=32.8, n=22).

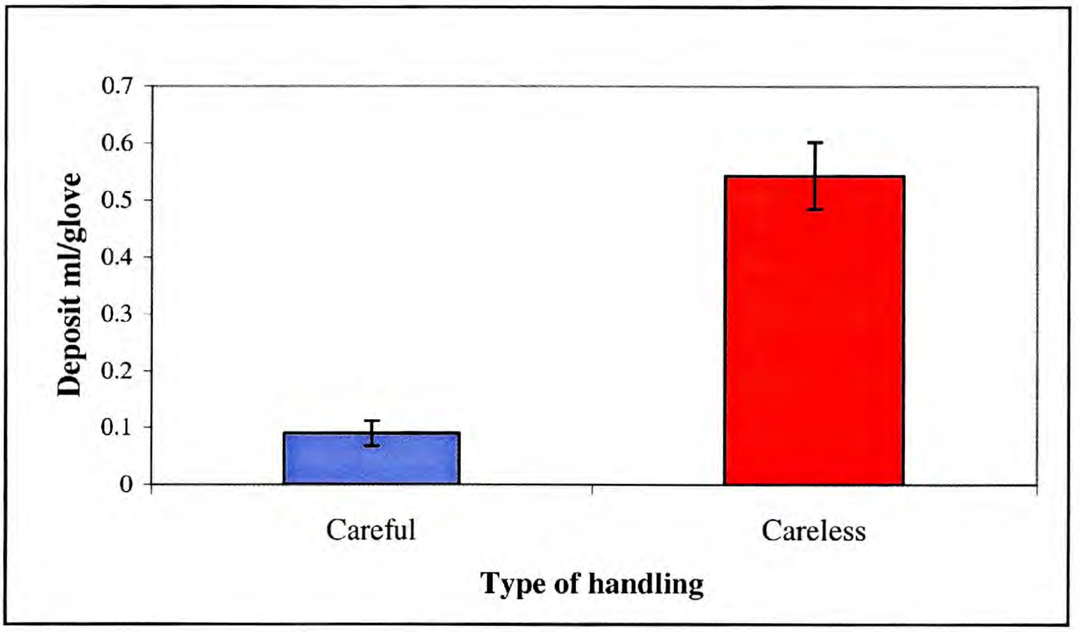


Figure 5.8 **Difference in deposit by contamination between safe and unsafe handling of treated trees. There was a significant difference in the volume of deposit (P<0.001, df=1, F=32.8, n=22).**

5.5 Discussion

The safety issues in this section examine the potential risk of pesticide exposure when an operator uses the DCS system. In addition, the use of an extractor fan system to reduce the risk posed by airborne particles was addressed. The results gave some insight in to the optimal system set-up in terms of safety. Unfortunately, this was not necessarily the best-set up given the results presented in Chapter 4.

When positioned correctly, the extractor fan system was extremely efficient at removing the very small droplets produced by the FE nozzle. Unfortunately the extractor fan significantly reduced the deposit on the tree. In addition, the FE nozzle caused a lower level of contamination on the operator's gloves. In contrast, the AI nozzle was less affected by the extractor system, as the mean droplet diameter is much larger (Appendix 8) with wider spacing between droplets. Therefore, the trajectory of the droplets remains unaltered due to the decreased turbulence created by the proximity of the droplets. The number of droplets escaping through the front entrance was minimal but not reduced by use of the extractor fan. Instead, better and more accurately positioned shielding would reduce contamination further, particularly if combined with a longer conveyor belt which would move the landing area of the sprayed droplets further from the operator.

The AI nozzle caused more glove contamination than the FE nozzle. This would be expected based on the findings by Brouwer *et al.* (1992) who correlated the level of dislodgeable deposit with dermal contamination. However, this glove contamination can again be reduced substantially by careful handling of the trees or by only handling the trees that become stuck in the belts. Many of the trees will drop freely from the conveyor belts and with a small chute below the drop point, the trees will line up in neat bunches.

For the AI nozzle, the volume of escaping droplets from the front of the conveyor belt could be reduced to near zero, whilst a substantial reduction was seen for the FE nozzle. However, the Side position was less effective at equally spraying all trees and was less reliable than the Front position (Chapter 4). When the FE nozzle was set to spray from the side position, droplets were not confined to escaping from the front of the conveyor belt and were seen to drift in both directions. Therefore, they would require a second pair of extractor fan ducts to remove these airborne droplets. This would be considerably more problematic as multiple inlets from an extractor fan would substantially reduce the airflow and result in different air flow speeds across each inlet. It has also been shown how poor positioning of the duct inlets can alter airflow so that more droplets escape! Therefore, extractor fan inlets at each end of the conveyor belt could reduce the effectiveness of the nozzles.

Finally, the effects of a surfactant were examined to see if the larger droplets produced by the AI nozzle could be encouraged to disperse and become less likely to run off or be knocked off. The results strongly showed that tree deposit was enhanced for the FE nozzle but reduced for the AI nozzle when a surfactant was used. There was little difference in deposit on the gloves when using surfactant. It is likely that the surfactant altered the droplet formation resulting in a lower deposit. This correlates with results by Butler Ellis & Tuck (2000), who found that the use of a water soluble surfactant such as Agral significantly increased droplet size for the AI nozzle, whilst reducing droplet size for a flat fan nozzle compared to a water control. For the FE nozzle it may have encouraged better deposition and reduced splash back or bounce off leading to the enhanced deposit witnessed. In contrast, the larger droplets of the AI nozzle, which tend to have more air rather than liquid, may have led to decreased deposition.

In summary, the two areas of possible pesticide exposure through use of the DCS system can be reduced by inclusion of an air extraction system and by reduced handling of treated trees. The use of different nozzles can also affect the risk of exposure. Unfortunately, the two nozzles work antagonistically, with the FE nozzle reducing the risk of dermal exposure whilst the AI nozzle reduces the risk of inhalation. Of the two types of exposure, it would be better to reduce the airborne particles by use of the AI nozzle, rather than use the FE nozzle combined with the extractor fan, as this combination may reduce deposit. Proper use of protective clothing and gloves would then reduce the increased risk of actual dermal exposure caused by the use of the AI nozzle.

The final risk is to that of the planters. In the event that the trees have not dried in the bag in transport from the nursery to the planting site, it is likely that the larger droplets that are prone to contaminating gloves will spread quite rapidly after spraying. Therefore, there is unlikely to be any easily dislodgeable pesticide when the planters handle the trees. The surface of the tree may be damp, but handling the saplings with the proper protective equipment (as is currently used) will eliminate risk without the need for further protective equipment.

Chapter 6
Recognition and avoidance of pesticide treated Scots pine (*Pinus sylvestris*) by *Hylobius abietis* L.

6.1 Introduction

The Forestry Commission typically prophylactically treats all transplant saplings that are to be planted in clear fell sites where *Hylobius abietis* populations are high. In doing this they avoid from 30% to 100% mortality that may otherwise be sustained (Heritage *et al.*, 1989). This treatment has typically been applied either by dipping in a vat of the pesticide permethrin or by the Electrodyn sprayer conveyor system (Heritage, 1996; Stoakley & Heritage, 1990). These treatments occur in the nursery before the saplings are transplanted. The applied insecticide gives one to two seasons protection from damage by *H. abietis* and also by the less damaging *Hylastes sp.* (Heritage, 1997). Prophylactic treatment avoids the necessity of spraying non-target areas and therefore some of the potentially hazardous effects of pesticide use.

Following the results of the trials described in Chapter 3, the DCS has a potential future fulfilling the above role of prophylactic treatment of saplings in forestry. Therefore, it was considered important that all aspects of the system are fully understood. Chapters 4 and 5 examined the effects of various settings to enhance deposit and decrease the risk of exposure. This chapter examines the nature of protection given by the pesticide. It is known that the pyrethroids in question are repellent to many insects (Armstrong & Bonner, 1985; Herve, 1985; Tan, 1981) and that pyrethroids rapidly deter feeding in *H. abietis* (Eidmann *et al.*, 1996). Additionally, pyrethroids have been shown to reduce the attraction of cut pine sticks in olfactometer tests (Watson, 1999). It has also been reported (Heritage S.G., *Pers. Comm.*) that for some insecticides, large numbers of dead *H. abietis* can be found at the base of treated saplings. Therefore, it remains unclear whether the pesticides are repelling or killing the insects.

In this study, there were four key areas of research. Firstly, the preferences of *H. abietis* for treated and untreated food were determined in simple choice and no-choice tests. Secondly, time to death studies were used to determine how long *H. abietis* lived in the presence of treated food and whether the pattern of death differs significantly from starvation. In addition, one experiment examined the recovery of the weevils after exposure to the pesticide. This should provide some indication as to how the pesticide is killing the weevils and on recovery (if it occurs) will damage occur on the saplings? Finally, the data collected were applied to a mock field situation in a glasshouse trial to indicate the potential that *H. abietis* might have to cause damage to planted saplings when only some of the saplings were treated.

6.2 Materials and methods

6.2.1 Collection of weevils

All weevils were provided by the Forestry Commission and were collected from Newton field station (UK NJ 165 635) on the 26/10/2001. The weevils were kept in large rectangular plastic boxes (280mm x 160mm x 70mm) at 5°C for no more than two months prior to use. The weevils were separated according to sex and starved for a period of 48 hours at 15°C before the experiment, to ensure consistency in feeding damage between replicates.

6.2.2 Treatment of Scots pine twigs

A single method of treatment was used to spray pesticide onto the prepared Scots Pine (*Pinus sylvestris*) twigs. Small branches were collected from Crowthorne woods, Bracknell, Berkshire (UK SU 856 654) a few days prior to the experiment. The branches were cut into 100mm lengths and roughly sorted into size classes. Only one size class was used for any one individual experiment to avoid any element of choice by size selection. The overall range was quite small with diameters ranging from 5-10mm. Those twigs outside these boundaries were discarded. *H. abietis* have been shown to feed on twigs up to 20mm in diameter (Orlander *et al.*, 2000).

Twigs were placed on tin foil covered plastic trays (500mm x 300mm) arranged side by side and with a 1.5cm gap between each twig. Twigs were then sprayed using the Mardrive (IPARC, Silwood Park, Imperial College) that simulates tractor or conveyor spraying methods. Approximately 0.018ml of pesticide solution or water was sprayed onto each twig using a TP6501 even spray flat fan nozzle at 2.5 bar pressure. The trays of twigs were then left to dry for 24 hours at room temperature.

6.2.3 Choice trials

No choice trials and choice trials were set up to establish preferences that *H. abietis* may exhibit for untreated food or to identify if *H. abietis* is able to detect pesticide deposit on a food source. All trials were carried out in a controlled temperature room (CT room) at 15 °C, ambient humidity and a 16/8 hour light/dark cycle.

Thirty *H. abietis* were assigned to one of three treatments; double control; double treatment and control/treatment. Each treatment had ten replicates with five males and five females.

Treated twigs were sprayed with either 0.1% ι-cyhalothrin (a pyrethroid) or 0.1% imidicloprid (a nicotinoid). Control twigs were sprayed with distilled water under similar conditions but dried in a separate location to avoid cross contamination.

For each treatment, two 100mm Scots Pine (*Pinus sylvestris*) twigs were placed in resealable plastic containers (100mm x 150mm x 60mm) with a damp piece of tissue paper. An even gap was left all the way around each of the twigs. The experiment was run for seven days. At the end of this period the weevils were assessed for mortality and the twigs were assessed for damage.

6.2.4 No choice trials

The no choice trials followed a format identical to the choice trials. The *H. abietis* were assigned to one of three treatments; control, 0.1% and 0.5% pesticide treatments. Treated twigs consisted of either 0.1% and 0.5% ι-cyhalothrin or 0.1% and 0.5%

imidicloprid. These doses were chosen, as they were similar to the pesticide concentrations used in the field trial (Chapter 3). Control twigs were treated as for the choice trial. For this experiment only one twig was placed in each container and the experiment run as before.

6.2.5 Time to death experiments

Forty-two *H. abietis* were assigned to one of four treatments, control, starved, ι-cyhalothrin or imidicloprid. For the latter two treatments, twigs were treated with pesticide at a concentration of 0.5%. Weevils were placed into resealable plastic containers as previously described above. Each container held one twig, except for the starved group, which did not receive any food. The weevils were checked every 24 hours for mortality. A weevil was considered dead when it was unresponsive to poking or stroking of its antennae. It usually had its legs curled up underneath its body. As *H. abietis* shows exceptional thanatosis, assessment of mortality was always continued for a few days after presumed death. The containers were moistened with distilled water once a week with a hand mister.

6.2.6 Recovery after exposure

H. abietis were assigned to one of six treatments consisting of two different factors. There were two periods of pesticide exposure, one day and seven day durations. There were three concentrations of pesticide including a water control (Control, 0.1%, 1.0%). There were 15 weevils per treatment group. The weevils were starved for 48 hours and placed in resealable plastic containers under the conditions described previously.

During the exposure period the weevils were given one 100mm Scots pine twig that had been treated with ι-cyhalothrin as described. This twig was then removed and the damage assessed. For the next 14 days new untreated 40mm lengths of Scots pine twigs were added. Each day the old twig was removed and the damage measured. After the 14 day period the weevils were monitored daily for mortality for a further two weeks.

6.2.7 Glasshouse experiment

To simulate field conditions but with an enclosed environment, a glasshouse trial was set-up. Three 3-year-old Sitka Spruce (*Picea sitchensis*) were planted in pots, equally spaced. Large 25ι black pots (400mm in diameter) were filled with 5ι of soil. The trees were watered and left for one week to bed in.

The experiment consisted of four treatments; three control trees; two control trees and one treated; one control tree and two treated and three treated trees. Treated trees had been sprayed with a 1% solution of ι-cyhalothrin two days prior to planting. Treatment took place in the Mardrive apparatus under conditions described above. The concentration used was directly comparable to the field rate.

Five *H. abietis* were added to each pot and fine muslin mesh covers were constructed to cover the trees and pot. Weevils had been starved for 48 hours prior to the experiment. These treatments were then left for two weeks in the glasshouse with ambient humidity, 16/8hr light dark regime & minimum daily temperature of 15 °C.

6.2.8 Measurement of feeding damage

An updated version of the method described by Leather *et al.* (1994) was used to assess feeding damage. Damage was traced onto a 100mm x 30mm piece of acetate with a black acetate pen. The damage was then scanned using an Epson Scanner and measured using Area V software (designed by Peter Mueller), which measures the area of selected objects. The mean area of damage per treatment was then calculated and this was analysed using Anova, time to death analysis or time series analysis in S-Plus.

6.2.9 Measurement of fat content

Fat content was measured by the methods described by Strohm (2000). Weevils taken from several experiments were starved for one day before freezing at –17 °C to give the gut contents time to pass. The weevils were dried at 50 °C for seven days and the dry weight measured. Petroleum ether (1-2ml) was added to a glass vial (5mm x 40mm) containing the insect, amply covering the body. The head or head and thorax were separated from the rest of the body to allow the petroleum ether to penetrate the cuticle. The weevils were soaked in the petroleum ether for five days. During this time the petroleum ether was completely exchanged to remove the dissolved fat. At the end of this period the insects were dried for a further four days at 50 °C.

The dry weight was measured again and the difference between the before and after weight was taken as the absolute fat content. The relative fat content was a measure of the before weight and the absolute fat content. Analysis of Covariance (Ancova) was used to compare treatments.

6.3 Results

6.3.1 Choice and no choice

The effects of two chemically different pesticides were studied in these trials; ι-cyhalothrin, a pyrethroid insecticide known for its repellent properties and imidicloprid, a nicotinoid for which its repellency effects are less clear.

In the choice trials there was a clear effect of preference against both pesticides ($P<0.05$, $F=4.05$ (ι-cyhalothrin) $F=13.54$ (imidicloprid), $df=2$, $n=30$) when the combined damage per treatment was examined (Figure 6.1). In addition, comparisons showed a difference between the damage for treated and control twigs in the control/treatment group ($P<0.001$, $F=15.64$ (ι-cyhalothrin) $F=16.89$ (imidicloprid) $df=1$, $n=30$).

A similar pattern of results was found for the no-choice trials. *H. abietis* showed a significant difference in the levels of feeding damage between treatments for both pesticides. Treated twigs were largely untouched whilst untreated ones were heavily damaged. Anova showed that for both pesticides there was a significant difference between treatments ($P<0.001$, $F=46.89$ (ι-cyhalothrin) $F=103.55$ (imidicloprid) $df=2$, n=30) whilst further Tukey analysis showed the differences occurred between the pesticide treated and control groups. There was no difference between the two pesticide concentrations (Figure 6.2). In addition, the pesticide significantly reduced survival (P<0.001, F=11.14, df=2, n=30).

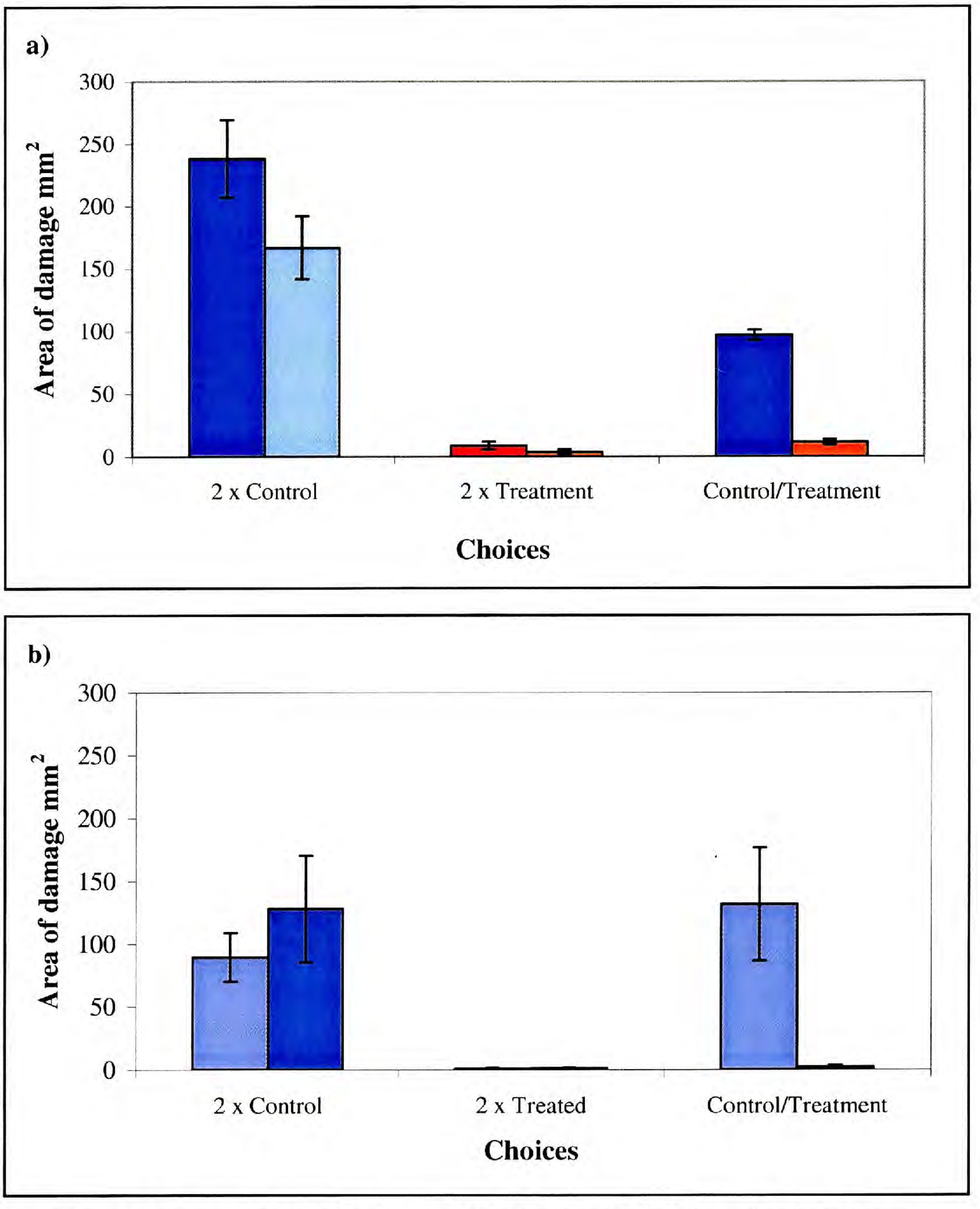
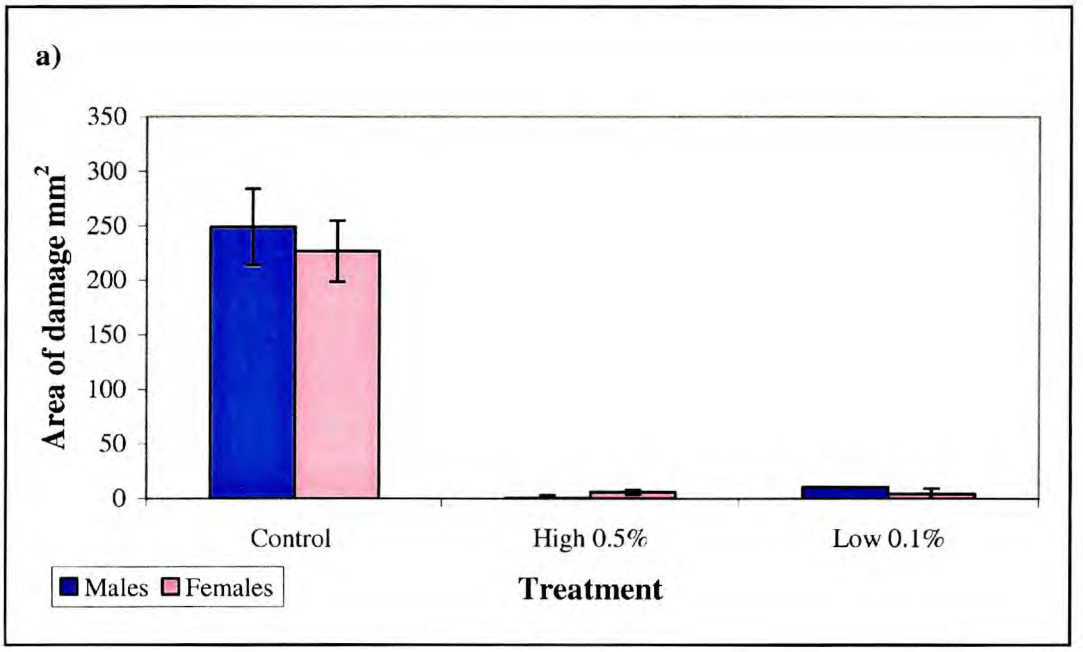


Figure 6.1 **Choice trials for ι-cyhalothrin (a) and imidicloprid (b). *Hylobius abietis* showed a significant preference for untreated twigs (blue bars) and caused very little damage to pesticide treated twigs (red bars). There was no difference between the two sexes. See text for P values.**



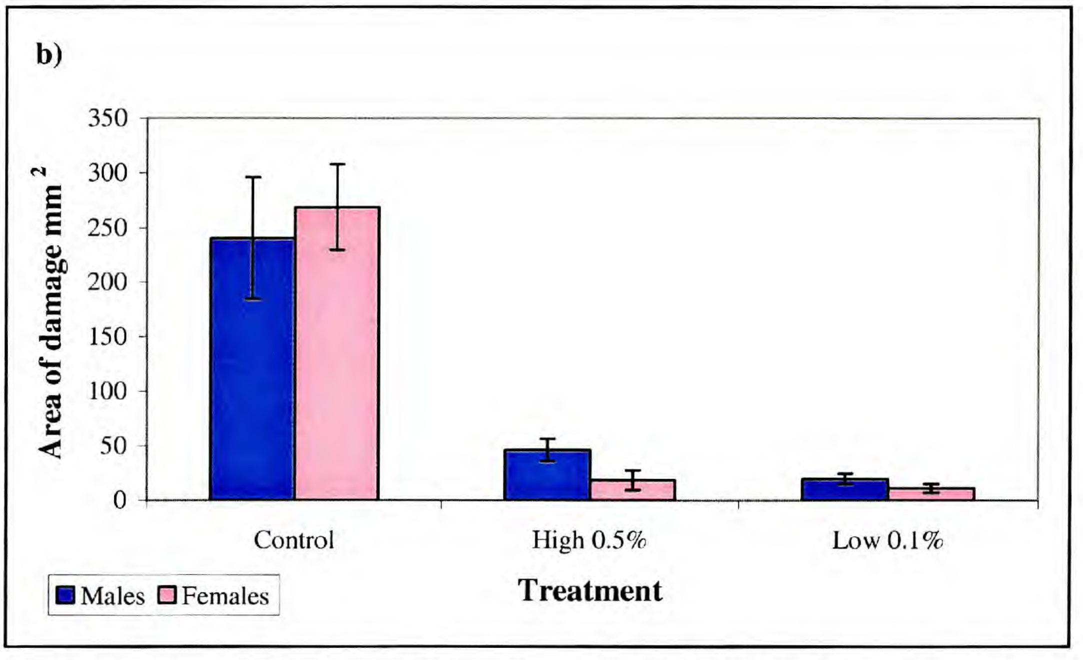


Figure 6.2 **No-choice trials examining the level of feeding damage incurred by Scots pine twigs when treated with two concentrations of either ι-cyhalothrin (a) or imidicloprid (b). There was a significant difference in the mean feeding damage by *Hylobius abietis*. There was no significant effect of sex on feeding damage. See text for P values.**

Throughout the choice and no choice trials there was no significant effect of sex for either pesticide. Therefore in the above analyses males and females were grouped to give more replication.

6.3.2 Time to death

At the end of the choice and no choice trials mortality of the weevils was assessed. As not more than 30% of the ι-cyhalothrin groups and 10% of the imidicloprid groups were dead, a time to death experiment was carried out to examine whether the weevils were starving themselves or if the pesticide was killing them very slowly. It was expected that a group of starved weevils would show the same time to death curve as a group of weevils with treated food if the mode of death was the same.

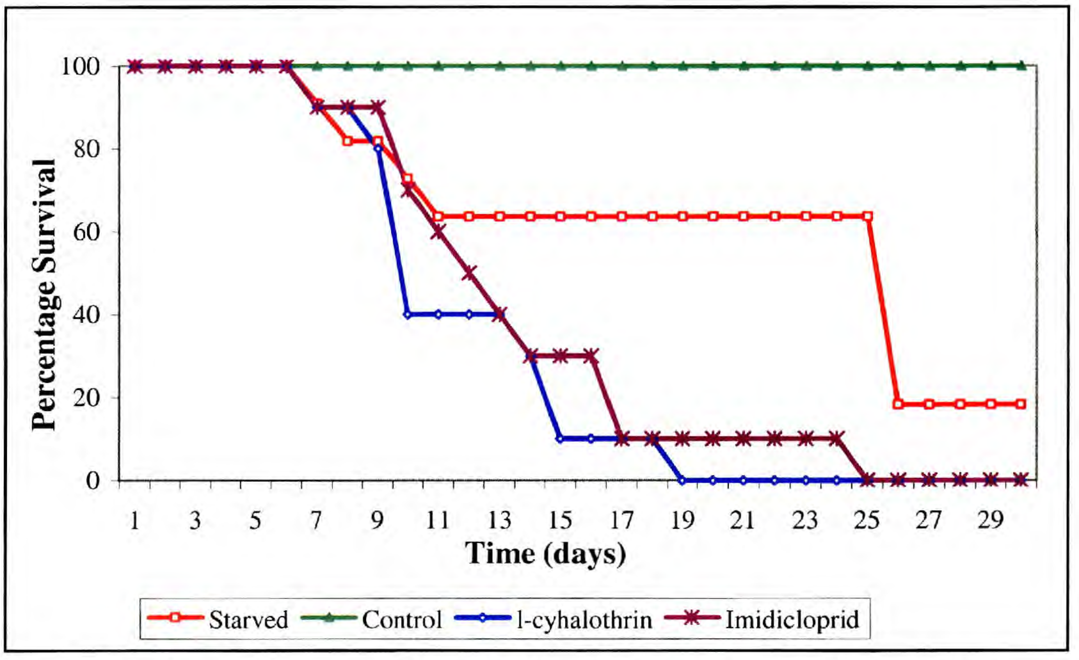


Figure 6.3 **Time to death for *Hylobius abietis* showing control groups, those that were starved and two groups that were fed pesticide treated food. There was a significant difference in the survival of the control groups and the treatment groups ($P<0.01$, $Chi^2= 22.24$, df=2, n=43).**

Clearly, there were apparent differences in the survival of pesticide treated, starved and control groups (Figure 6.3). The two pesticides showed very similar trends, whilst weevils lived much longer before dying in the starved group. Survival analysis showed significant differences in the survival of control groups from the other treatments ($P<0.01$, $Chi^2= 22.24$, df=2, n=43). Further analysis showed there was a significant difference between pesticide treatments and the starved group in both mean time to death ($P<0.01$, $Chi^2= 9.5$, df=1, n=31) and also in the shape of the fitted curves (Survival regression, $P<0.001$, z=34.06, df=1; n=31).

6.3.3 Recovery after exposure

The data were analysed in two ways using S-plus. Firstly, a time series analysis was used to estimate the effects of exposure duration, concentration of pesticide and sex of the weevil. The analysis showed that exposure and pesticide concentration were significant (Exposure: $P<0.001$, df=1, t=4.13, n=1153; Concentration: $P<0.001$, df=1, t=2.59, n=1153). There was no significant effect of sex on the model (P=NS, df=1, t=1.25, n=1153).

Although it was important to confirm the effects of exposure length on the recovery of the weevils, the important part of the analysis concerned the point at which the weevils recovered from the pesticide exposure. This was tested by carrying out a separate Anova for each day, looking for the point where there was no significant difference between treatments.

The data for one day exposure showed convergence on day five after exposure (Figure 6.4). In addition, there was initially no difference between the two pesticide concentrations, but by day three weevils that fed on 0.1% treated twigs were not significantly different from the control. The level of damage appeared to become more uniform from day six and in particular the two pesticides behaved more similarly than the control. However, there were no significant differences in any of these groups. Additionally, the weevils exposed to pesticide caused slightly more damage, but again there were no significant differences.

This was not the case for the seven day exposed weevils, which caused significantly different levels of damage compared to the controls through much of the experiment. Convergence was not as clear in this experiment except on the very first day after exposure (Figure 6.5). Thereafter, the low concentration treated twigs suffered significantly more damage than the controls throughout the experiment. The weevils exposed to the higher concentration took several days before they repeated the pattern and this occurred on day ten. Additionally, they caused significantly less damage than the controls until day four, where convergence occurred.

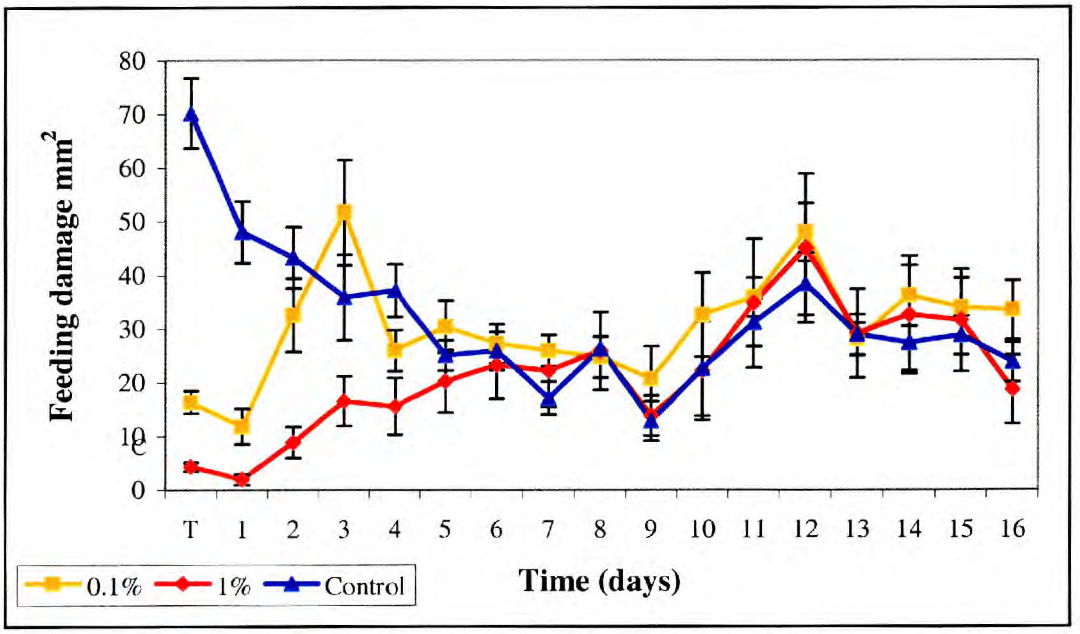


Figure 6.4 **Daily feeding damage by *Hylobius abietis* after exposure to 0.1% and 1% ι-cyhalothrin treated food for <u>one day</u>. There was no significant difference between treatments by day five through to the end of the experiment. See text for full explanation.**

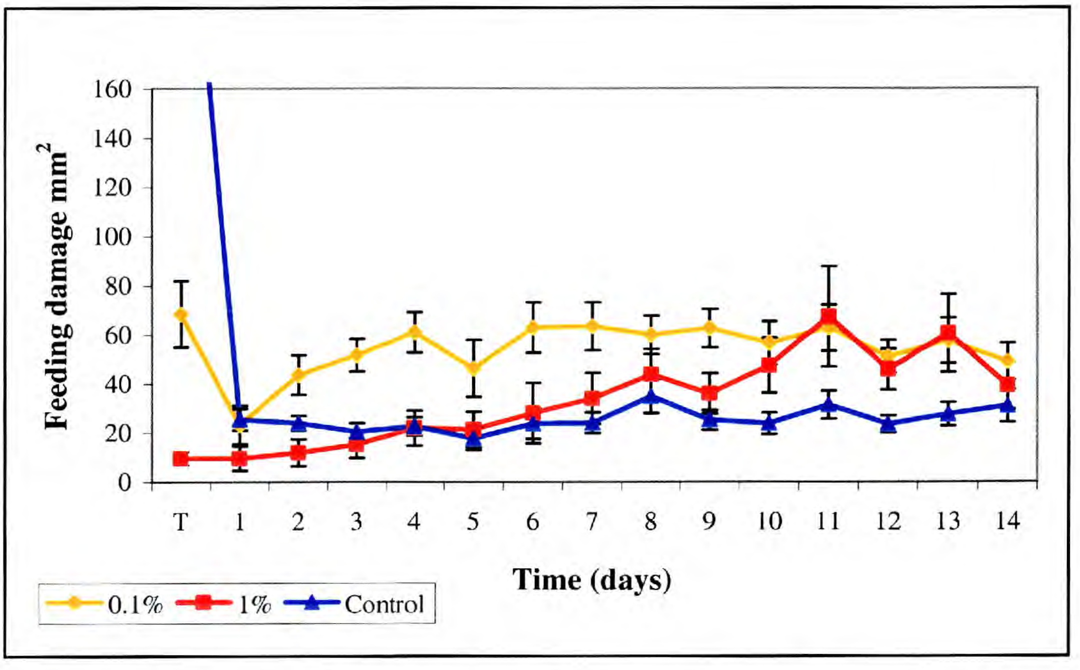


Figure 6.5 **Daily feeding damage by *Hylobius abietis* after exposure to 0.1% and 1% ι-cyhalothrin treated food for <u>seven days</u>. Initially there were no significant differences between the highest concentration and controls in the level of feeding damage. See text for full explanation.**

6.3.4 Glasshouse trial

From this section of the work, two clear patterns emerged. Firstly, untreated trees were heavily damaged when in the presence of treated trees ($P<0.05$, $F=6.27$, df=1, n=40, model fitted allowing for pseudo-replication, Figure 6.6). *H. abietis* were able to detect those trees that were untreated. At the end of the experiment over 70% of the weevils were recovered alive, the remainder were either missing or dead. Missing weevils may have escaped through the drainage holes or dead and therefore too difficult to find in the soil.

Secondly, there was a strong negative trend in the level of measured damage compared to the total number of trees treated ($R^2= 0.66$, $y=1894.22 + x.-0.812$). This indicated that when untreated trees were rarer, the damage they suffered may have been higher.

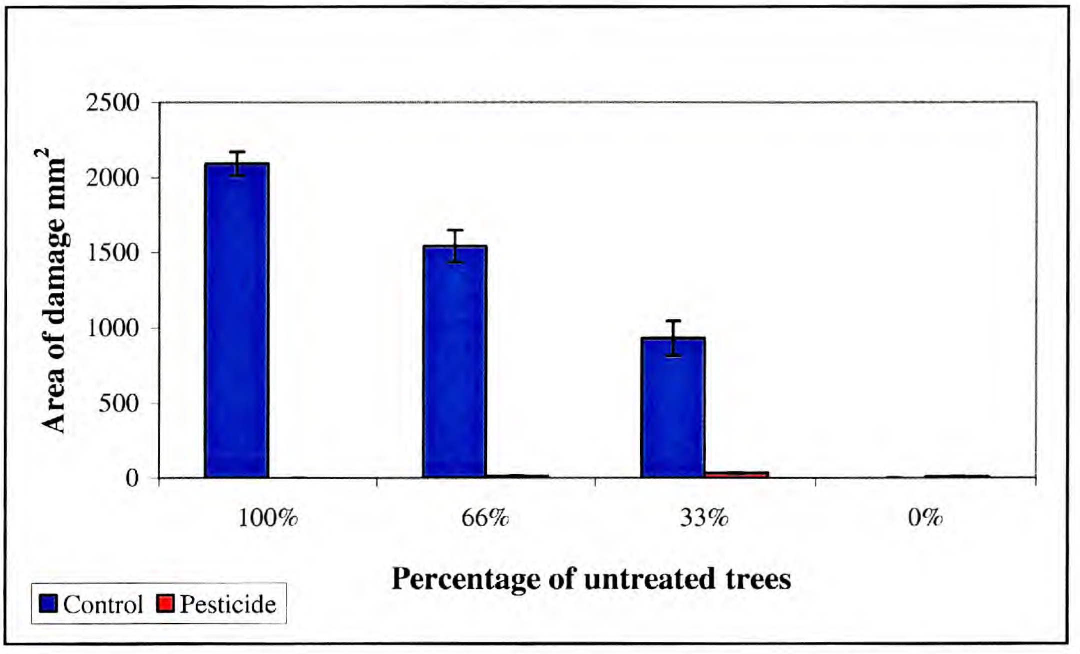


Figure 6.6 **Summary of the feeding damage per tree per pot, incurred by *Hylobius abietis* to groups of Sitka spruce saplings in a glasshouse experiment. There was a significant effect of treatment on the damage done to the saplings ($P<0.05$, df=1, n=40). Untreated trees were much more heavily damaged. In each pot there were 1, 2 or 3 like treated trees.**

6.3.5 Measurement of fat content

Fat content was measured in 148 insects across a range of treatments. The weevils were exposed to two pesticide concentrations that had been applied recently or two months previously for differing lengths of time (one or seven days). In addition, some were given a period of recovery (two weeks) whilst others were starved for this period (Figure 6.7).

There was no significant effect of sex on the fat content of weevils in relation to the pesticide regime that the weevils experienced (P=NS, df=1, F=2.4, n=146). Therefore, sex was combined to give greater replication. Overall, there was a significant effect of treatment (P<0.001, df=8, F=7.93, n=146).

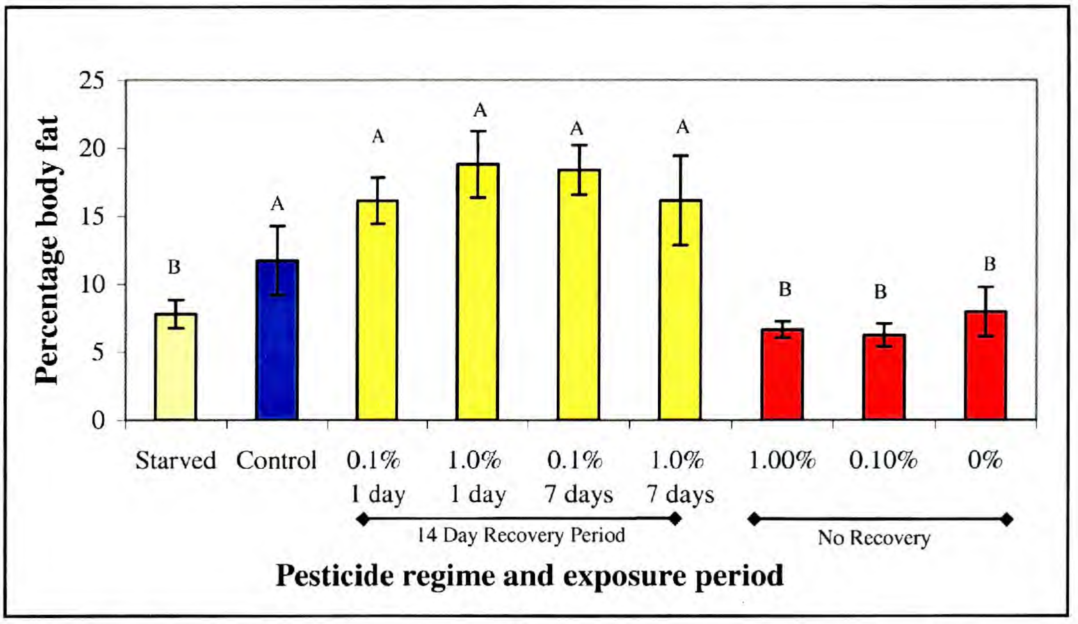


Figure 6.7 **Comparison of the relative fat content of *Hylobius abietis* exposed to a range of pesticide conditions. The starved group were starved for seven days. The control groups were allowed to feed individually for 14 days. The '14 day recovery period' groups were exposed to one of two pesticide concentrations for either one or seven days and then given 14 days to recover. The 'no-recovery' groups were exposed to pesticide for seven days.**

On further analysis, a Tukey post hoc test identified the treatments that were significantly different from each other (labelled A or B) (Figure 6.7). Both the control group (fed during the experiment) and those weevils given a 14-day period of recovery had significantly higher relative fat contents than weevils either recently exposed to pesticide or starved during the experiment.

The initial data set included weevils that had been exposed to pesticide that was either sprayed onto the food source two months previously (Old) or a few days before the experiment (New). Previously, it was shown that weevils exposed to old food sources caused less damage than to fresh food sources, regardless of pesticide treatment (Brun, 2002 Unpublished). There was also a significantly greater difference in the relative fat content of the Old compared to the New treatments ($P<0.05$, df=1, $F=6.54$, n=60). Only the New group are shown in Figure 6.7.

Finally, there was a significant effect of pesticide concentration ($P<0.05$, df=3, $F=2.70$, n=146) and also the presence of a recovery period ($P<0.001$, df=1, $F=52.4$, n=146). The highest pesticide concentrations had marginally lower fat contents, but the effect was primarily due to the differences between the control and treated groups.

6.4 Discussion

The object of these experiments was to identify the means by which pesticide treatment provided protection to saplings from *H. abietis*. *H. abietis* has been shown to feed on a variety of food sources ranging from deciduous trees to mature conifers (Orlander *et al.*, 2000) as well as economically important conifer saplings. It is important to identify whether pesticide is merely deflecting *H. abietis* attack onto these sources or actually suppressing the population.

Evidence from this study has clearly shown that *H. abietis* is able to detect the presence of a pesticide, notably ι-cyhalothrin and imidicloprid. Feeding on food sources treated with these insecticides is significantly depressed and in most cases non-existent. Throughout this work and other studies, this effect is consistent, as is the low mortality *H. abietis* suffers. Therefore, it is important to examine the potential of *H. abietis* to show the same trends in the field. The glasshouse trial, which used live saplings as opposed to cut twigs, again emulated the pattern. This suggested that in the field, *H. abietis* would choose food sources that are pesticide free.

The time to death study showed that *H. abietis* may take up to three weeks to die when fed on pesticide treated food. This mortality may be due to the action of the pesticide or it may be a combination of starvation and pesticide action. If this an effect of the pesticide, then *H. abietis* will only have a limited amount of time to find other food sources and damage them before it dies. However, it may be that the weevil feels 'nauseous' after consuming the pesticide and slowly returns to normal health if given untreated food. Alternatively such nausea may prevent the weevil from feeding for several days. In a field situation there is potentially a substantial period for *H. abietis* to find untreated food sources and cause significant damage. The evidence from this trial indicated that there may have been a different underlying cause of death between starved and pesticide treated groups, as shown by the different mean time to death and the shape of curve in the time to death experiment.

The long-term effect of the pesticide was investigated in the recovery study. This study suggested that even after short pesticide exposure the weevil damage may be initially suppressed. When the weevils were exposed for one week the feeding suppression still occurred, but there was a subsequent period of increased feeding damage. An increased feeding rate after pesticide exposure has been shown by Pinch (2002, Unpublished) with ground beetles and the insecticide pirimicarb. There was also a difference between high and low pesticide concentrations, with the weevils exposed to the highest concentrations taking longer to recover.

Assessment of body fat content revealed that the weevils given a recovery period restored their body fat whilst those recently exposed to pesticide for one week had reduced body fat levels similar to that of a starved group. The similarity in the relative body fat levels between the starved and recently exposed groups strongly suggests that death by starvation may have played an important role in the cause of mortality.

This combination of evidence strongly suggests that *H. abietis* is likely to avoid treated trees in the field and seek out those that have failed to be treated in the nursery. Additionally, the glasshouse trial also showed that there was a increase in damage done to untreated trees as they became rarer. Therefore, it is unlikely that a few untreated trees in a field may have increased protection from a nearest neighbour. In addition, weevils that encounter pesticide treated food sources and also feed on it may suffer temporarily reduced feeding. It is unclear if the pesticide prevents fat deposition by interference of the esterases responsible, as occurs with pirimicarb and *Pterostichus cupreus* (Wallin *et al.*, 1992). Alternatively, it is likely to be due to enforced starvation when only a treated food source is available.

Field populations of *H. abietis* have been reported to be as high as 150,000 (Heritage, 1996) and 220,000 adults per hectare (Leather *et al.*, 1995). This is significantly more than used in these experiments and at extreme densities the relationships presented here may no longer apply. However, the complete lack of feeding damage to many of the treated trees and twigs even after several weeks without suitable food indicated a very strong dislike by *H. abietis* of pesticide treated food. Therefore, it is unlikely that even in the field at high population densities the preferences of *H. abietis* would change.

In summary, *H. abietis* showed a strong preference for untreated food sources and are capable of making this choice in both laboratory and glasshouse conditions. It is probable that *H. abietis* would ignore treated saplings in the field and instead search for those that have been untreated or feed on the abundant alternative food sources.

Chapter 7
Effects of patchy pesticide coverage on the feeding behaviour of *Hylobius abietis*

7.1 Introduction

Previous experiments have shown that *Hylobius abietis* shows a strong ability to select for food sources that are not treated with pesticides (Chapter 6). In these experiments, however, the presented food source was either completely treated or not. Typically in a field situation treated saplings are unlikely to be completely covered in pesticide, as side branches will impede deposition of pesticide on the main stem. In these situations it is important to know whether *H. abietis* is able to detect the treated and untreated patches, or if a small treated zone is enough to deter the insect from feeding and stimulate it to find another food source.

It has been reported for other beetles, notably *Rhyzopertha dominica* (F.), that uneven pesticide coverage does not reduce the overall effectiveness (Muda & Cribb, 1999). However, this was for a stored grain pest and at least 50% coverage was still required.

Sub-lethal effects of pesticides on insects are common ranging from reduced foraging activities due to increased grooming, hyperactivity with no repellency, decreased fecundity and delays in development or emergence from larval stages (Alzogaray & Zerba, 2001; Epstein *et al.*, 2001; Liess & Schulz, 1996; Wiles & Jepson, 1994). Anti-feedant effects may be due to a stomach action of the pesticide, through a repellant action or by interruption of other behaviours leading to the prevention of feeding (Armstrong & Bonner, 1985; Gerard *et al.*, 1999).

Anti-feedant effects of pesticides have been reported for a variety of insects; *Spodoptera litura* (Fabricius) with azadirachtin (neem) (Mukherjee & Sharma, 1996); *Drosophila melanogaster* with permethrin (Armstrong & Bonner, 1985); cypermethrin against *Pieris brassicae* larvae (Tan, 1981); pyrethroids against the Mexican bean beetle (*Epilachna sp.*) (Dobrin & Hammond, 1985) and lastly against *H. abietis* (Eidmann *et al.*, 1996). These effects also occur with some other chemicals such as limonene against *H. abietis* and also spider mites, fleas, some Diptera and

Coleoptera (Ibrahim *et al.*, 2001). Therefore, identification of some pesticides as potent repellents rather than knock-down pesticides may have important implications for IPM programs in forestry.

A series of experiments were set up to examine the scale at which *H. abietis* is able to detect pesticide coverage. These experiments used both Scots pine twigs (*Pinus sylvestris*) and Sitka spruce saplings (*Picea sitchensis*). The first experiment examined the effects of different types of coverage, for example, banded, patchy, half radius, i.e. those patterns that may typically occur in spraying. The second experiment examined the scale at which *H. abietis* is able to detect pesticide.

7.2 Materials and methods

7.2.1 Patchiness on whole Sitka spruce saplings

Two experimental factors were investigated, concentration of pesticide with three levels (0.1%; 1% and 2%) and pattern of pesticide coverage of which there were four levels. These were; full coverage (Full); coverage over half of the circumference (Half radius); two bands of 3.5cm and 4cm at the top and bottom of the 15cm zone respectively (Bands) and a control (water spray). All of these patterns were applied only to the lower 15cm of stem of two-year-old Sitka spruce, the usual area for treatment. The two other coverage patterns (Half radius and Bands) covered the same total area of the tree (Figure 7.1).

The insecticide ι-cyhalothrin was applied at each of the three concentrations with a small paintbrush. Unfortunately, the volume applied was not quantifiable but this was considered to be less important than precise coverage. Only one coat was applied, the paintbrush was reloaded each time and any drips were quickly wiped off.

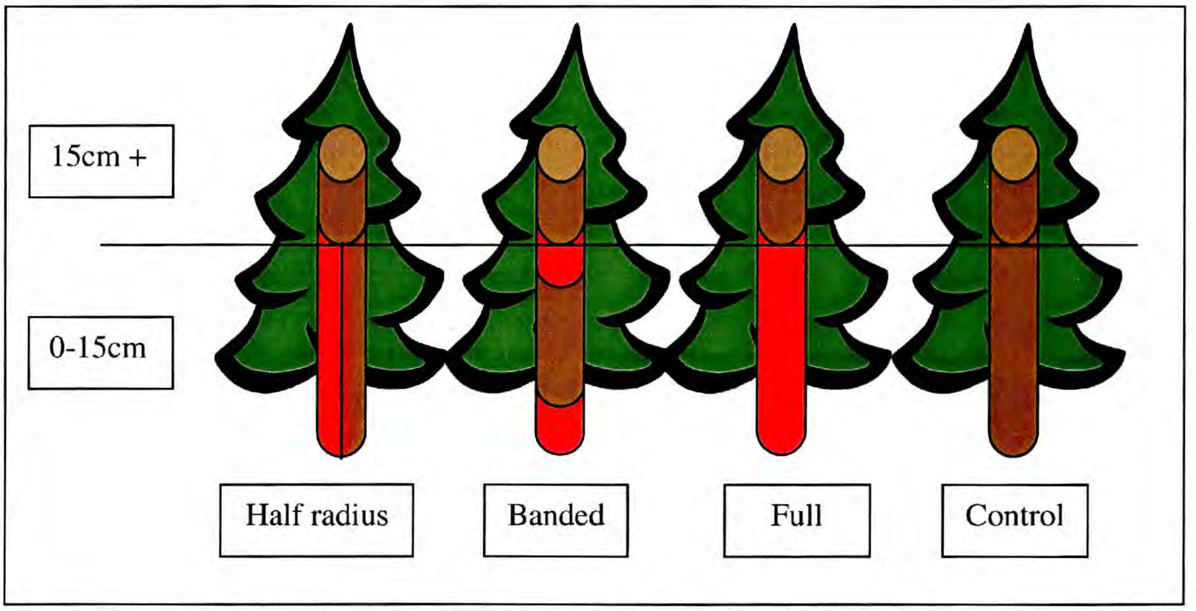


Figure 7.1 **Pesticide coverage patterns painted onto two year old Sitka spruce saplings.**

The Sitka spruce were planted in 1l pots with John Innes No.2 compost a few weeks prior to the experiment. A 5-10mm layer of plaster of Paris (approximately 50ml) was added to the top of the soil and left to set. This prevented *H. abietis* from burrowing in the soil and forced it to participate in the experiment. Randomly selected groups of four pots were then placed into washing-up bowls containing a shallow layer of water. One insect was added to each pot (four males and four females per treatment) and the pot placed in a fine muslin tube. This was tied up at the top with an elastic band and naturally formed a tight seal around the bottom of the pot. The muslin prevented an excess build up of humid conditions, whilst containing the weevil.

The experiment was monitored for one month. Insect mortality was recorded after one week and again at the end of the experiment. Bark damage was measured using the Acetate/Scanner method described in Section 6.2.9. Insect weight was recorded at the start and finish of the experiment. For this and all following experiments the data were log transformed and analysed using Anova.

7.2.2 Scale of detection on Scots pine twigs

Eighty *H. abietis* were assigned to one of eight treatments (four spray patterns at two concentrations). Scots pine (*Pinus sylvestris*) twigs were collected from Crowthorne

Woods, Bracknell, Berkshire (UK SU 856 654). These were cut in to 100mm lengths and assigned to one of four factor levels; Control (water); Full cover; four sections (Quarter) and eight sections (Eighths). The last three factor levels were each divided into 0.1% and 1% ι-cyhalothrin pesticide concentrations.

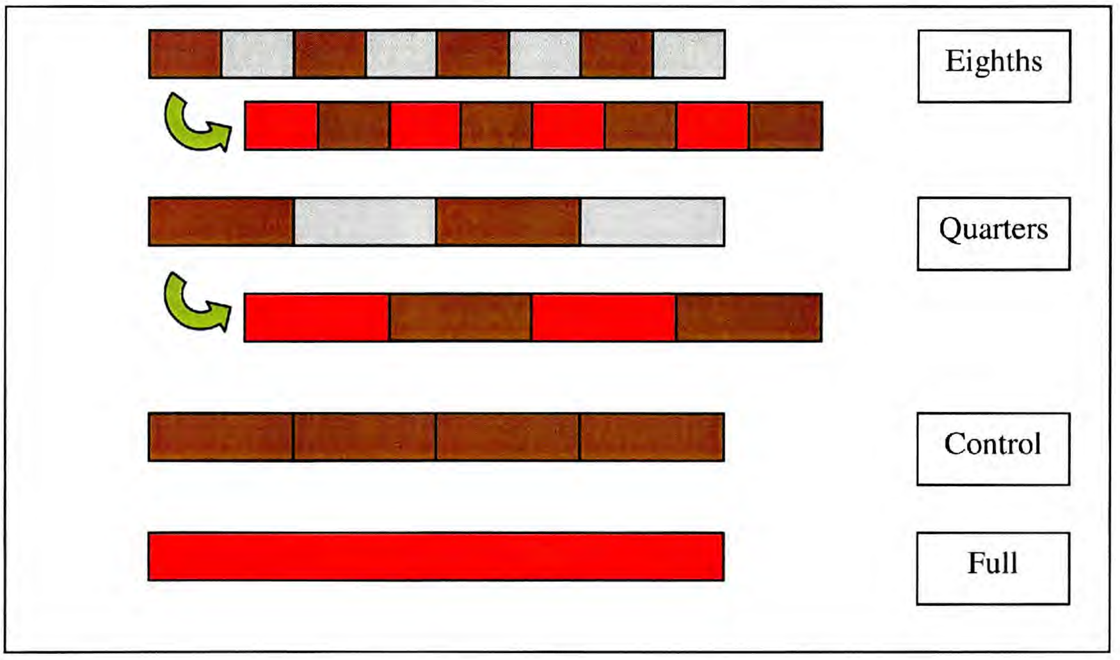


Figure 7.2 **Method of treatment of twigs. Tin foil wrappers were wrapped around portions of the twigs. The twigs were then sprayed and the tin foil wrappers removed. This left bands of treated and untreated zones as indicated by the red sections. Water controls were sprayed with water in the same manner.**

Tin foil wrappers were taped on to the twigs in the patterns shown in Figure 7.2. Permanent marker pen was used to indicate the edge of the zones. The twigs were then sprayed in the Mardrive (IPARC, Silwood Park) with the appropriate concentration of ι-cyhalothrin at 2.5 bar pressure, forward speed 24m min^{-1} with a TP6501E (FE) nozzle. The twigs were left to dry in a fume cupboard for two days to allow any excess volatile to evaporate. The tin foil wrappers were removed leaving treated and untreated bands.

One twig was placed into a 160mm x 110mm x 60mm resealable plastic container with a piece of damp tissue paper and one weevil. Equal numbers of each sex were used in each treatment. The weevils were left for one week and then the damage from each zone was recorded. The data were log transformed and analysed using the average damage per twig.

7.2.3 Olfactory detection and pesticide movement

This experiment was carried out as described in section 7.2.2. However, only half of each twig was treated with pesticide. The second half was wrapped in tin foil to protect it from pesticide spray. Once dry, the tin foil was removed and new tin foil was wrapped around the treated half of the twig. The weevil had no access to any of the treated parts and therefore any reduction in feeding damage would not be due to detection of the pesticide by taste. Two likely explanations are detection by olfactory methods or that the pesticide spread along the bark as oil would on water.

There were three experimental treatments, control, 0.1% and 1% ι-cyhalothrin, which were applied to the twigs in the Mardrive (IPARC, Imperial College, Ascot) using an FE nozzle at 2.5 bar pressure.

7.2.4 Detection of different droplet sizes

Scots pine stems were collected from Crowthorne Woods, Bracknell, Berks, and cut in to 10cm long pieces, then divided in to three treatments. Two groups were treated with 0.5% ι-cyhalothrin using either an AI nozzle or FE nozzle (flat fan even spray) in the Mardrive system (IPARC, Silwood Park) (Figure 7.3). The third group was sprayed with water using the FE nozzle. All three treated groups were left to dry for two days to allow any excess pesticide to evaporate.

Three groups of *H. abietis*, also collected from Crowthorne Woods, were starved for 24 hours prior to the experiment and placed in resealable plastic containers (100mm x 150mm x 60mm). A damp piece of tissue paper was added to maintain humidity and the experiment was conducted at 15 °C with 16/8-hour light/dark cycle.

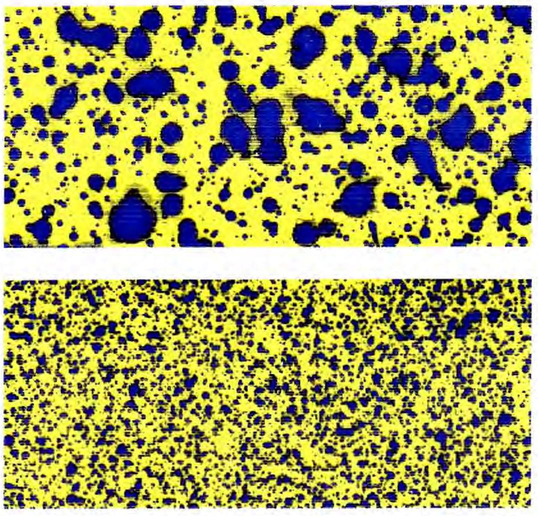


Figure 7.3 Nozzle Patterns

Comparison of the spray deposition of the FE (flat fan even spray) and the AI (Air induction) droplet structure. The AI nozzle (top) has a much coarser spray with larger gaps between droplets. The FE nozzle (bottom) has finer droplets with narrow droplet spacing. Plates actual size.

The twigs were added to the containers and the experiment monitored for one week. At the end of this time the damage was assessed as described previously. A total of 60 insects were divided between the three treatments. Larger replication was used to reduce the variance so that differences between the two nozzles could be highlighted.

7.3 Results and analysis

7.3.1 Patchiness on whole saplings

There was very little damage done to treated trees (Figure 7.4). However, it is not overly clear from these data that the levels of damage on treated trees were actually due to a few individuals rather than consistent low-level damage. Therefore, there is large variation in the results and the analysis was mostly non-significant. The whole data set showed no significant differences in either concentration or in the type of coverage. However, contrasting the two control groups with the three treated groups showed highly significant differences ($P<0.001$, $n=86$, $F=136.6$, $df=1$ and $P<0.001$, $n=86$, $F=53.9$, $df=1$ for above and below 15cm, respectively). There were no significant differences within the treated groups. In addition there were no significant differences between the two control treatments.

However, when comparing damage done above the 15cm zone to that done in the treated zone, there was significantly less damage in the treated zone ($P<0.001$, df=1, F=9.7, n=86, Figure 7.5).

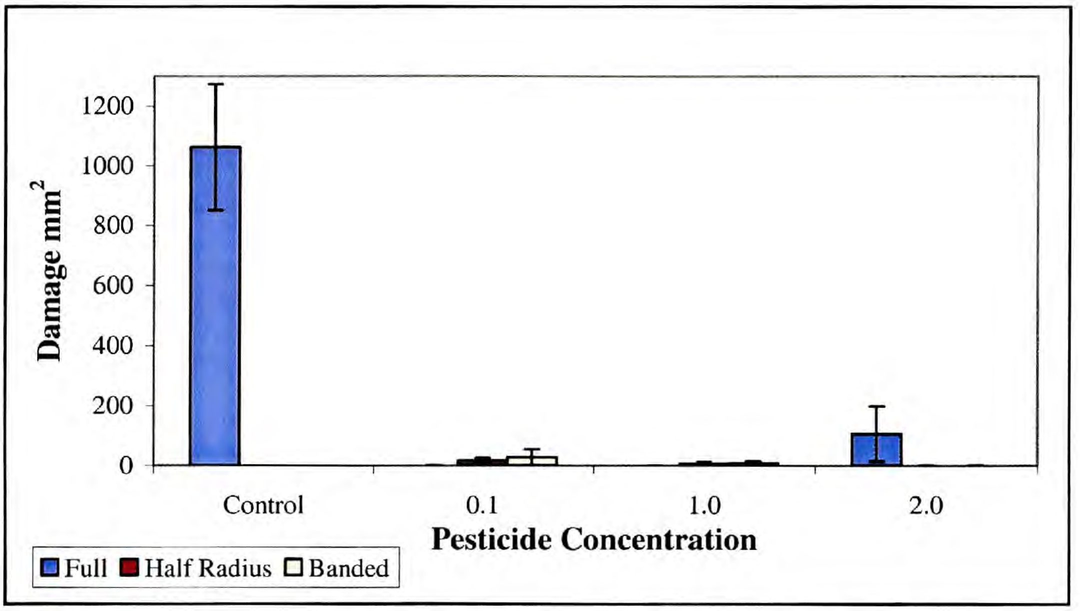


Figure 7.4 **Comparison of the damage caused by *Hylobius. abietis* using different pesticide coverage patterns at three concentrations within the treated zone. Controls have been grouped since there was no significant difference between them. Treated groups suffered very little damage. There was a significant difference between the control and treated groups (see text).**

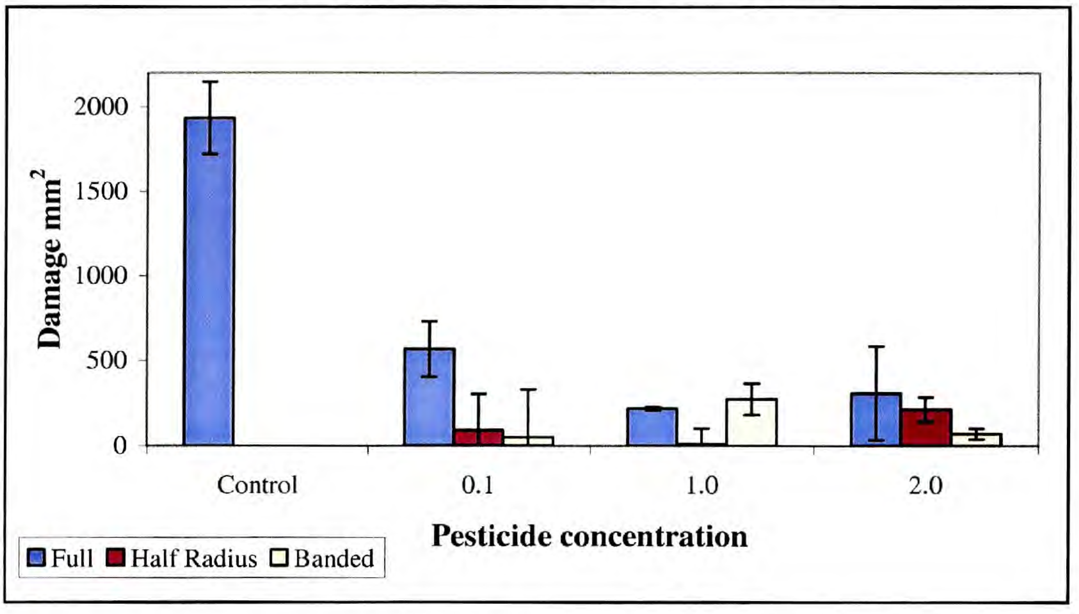


Figure 7.5 **Comparison of the damage caused by *Hylobius. abietis* using different pesticide coverage patterns at three concentrations above the treated zone. Controls have been grouped since there was no significant difference between them. Treated groups suffered significantly more damage than the treated zone ($P<0.001$, n=86, df=1). There was a significant difference between the control and treated groups ($P<0.001$, n=86, df=1).**

Weevil weights were also recorded before and after the experiment. Analysis showed that there was a significant decrease in mean weevil weight at the end of the experiment for the treated groups but not for the control groups ($P<0.001$, $n=83$, $F=5.4$, $df=1$).

7.3.2 Scale of detection on Scots pine twigs

An overall analysis was carried out, taking averages for the measurements from each twig and the data were log transformed. There was a significant difference between controls and treated twigs ($P<0.001$, $df=1$, $F=40.6$, $n=71$). In addition, controls were removed from the analysis and the differences between the three treatments examined. This also showed a strong effect of 'band width' on feeding damage ($P <0.044$, $df=2$, $F=29.4$, $n=53$).

Analysis of the controls revealed no significant difference in the level of damage to treated or untreated zones. This was to be expected since the whole twig had the same treatment but was marked off into four sections. There was however, a significant effect of sex on feeding damage ($P<0.001$, $df=1$, $F=14.09$, $n=20$) with females causing more damage than males, particularly in the control groups.

There was an indication of greater damage on the untreated parts (Figure 7.6 and Figure 7.7). In addition, the effect was more pronounced in Quarters than Eighths for males but not for females. This pattern was also evident in the previous experiment (Section 7.2.1) where greater damage occurred above the treated zone.

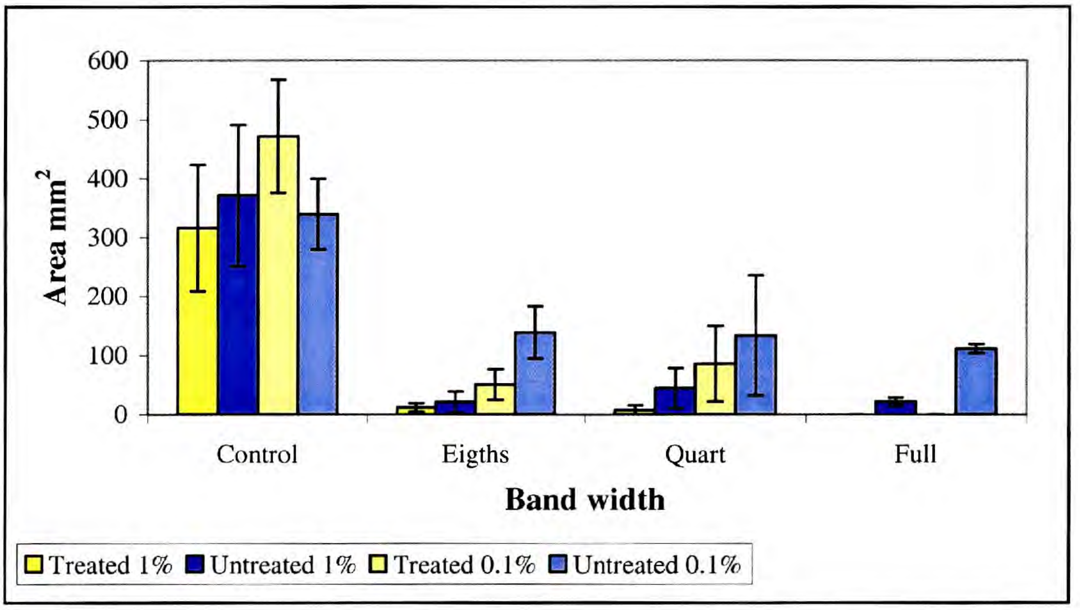


Figure 7.6 **The mean feeding damage caused by *Hylobius abietis* to treated and untreated parts of a Scots pine twig by <u>female</u> *H. abietis*. The lower concentrations are illustrated by the lighter shade bars. Twigs were treated with ι-cyhalothrin. All controls were sprayed with water only, but damage was recorded on "treated" and "untreated" sections to look for any natural partitioning of feeding.**

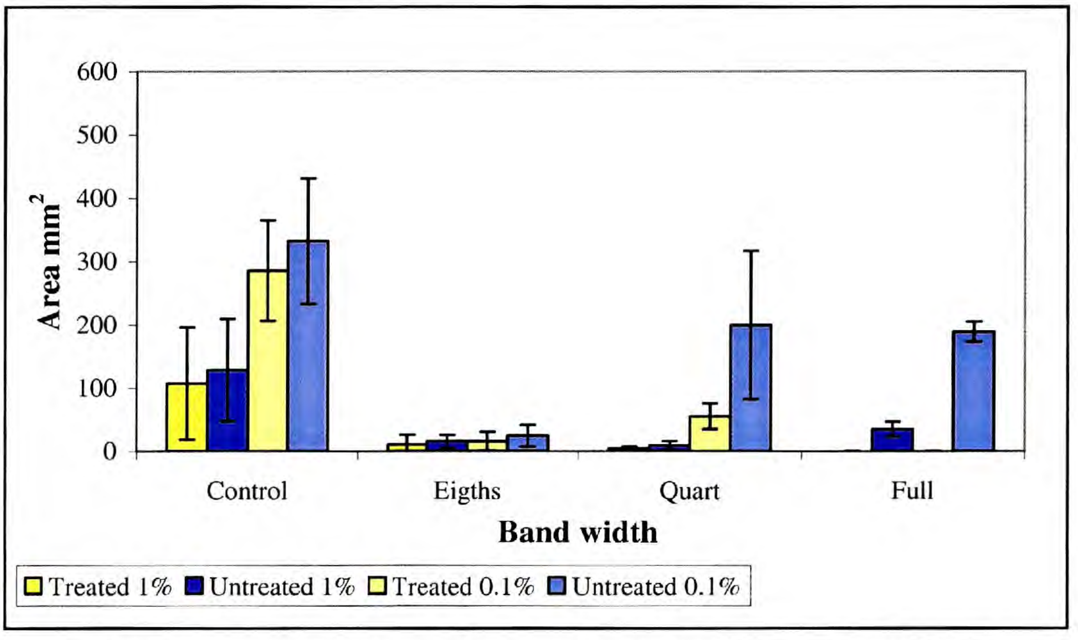


Figure 7.7 **The mean feeding damage caused by *Hylobius abietis* to treated and untreated parts of a Scots pine twig by <u>male</u> *H. abietis*. The lower concentrations are illustrated by the lighter shade bars. Twigs were treated with ι-cyhalothrin.All controls were sprayed with water only, but damage was recorded on "treated" and "untreated" sections to look for any natural partitioning of feeding.**

7.3.3 Olfactory detection and pesticide movement

There were no significant differences between any of the three treatments (P=NS, df=2, F=2.2, n=30). Analysis of the two pesticide treatments also did not reveal any differences. Feeding damage was observed very close to the tin foil wrapper indicating that there was none of the usual observed feeding reduction when pesticide was present.

7.3.4 Detection of different droplet sizes

There was a significant effect of treatment (Control > AI > FE) on the level of feeding damage (P<0.001, df=2, F= 87.9, n=56). There was no effect of initial weight or sex (Figure 7.8).

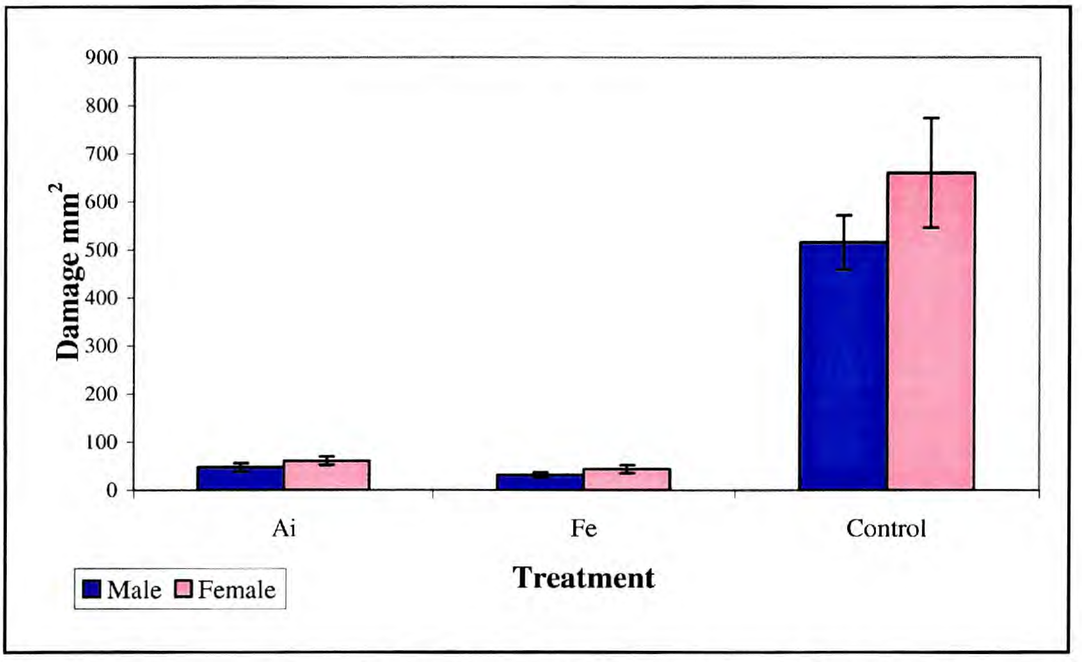


Figure 7.8 **Effects of nozzle type on the feeding damage of *Hylobius abietis*. There was a significant difference between the three treatments (P<0.001, df=2, F=87.9, n=56), in particular there was a significant difference between the two nozzles (P<0.001, df=1, F=92.88, n=38). There was no effect of sex.**

However, further analysis revealed an interesting difference between the two nozzles. Trees treated using the AI nozzle suffered significantly more damage than those treated using the FE nozzle (P<0.001, df=1, F=92.88, n=38).

7.4 Discussion

This section of work aimed to identify whether adult *H. abietis* are able to detect pesticide treated food and if so, at what scale of coverage this was possible. Simple choice experiments and no choice experiments (Chapter 6) confirmed that *H. abietis* were indeed able to detect pesticide treated food.

Chapter 6 examined the effects of pesticide treated food sources on the feeding behaviour of *H. abietis*. This section confirmed that *H. abietis* were able to discriminate between treated and untreated food sources. Furthermore, extrapolation to a field type scenario (in Chapter 6) showed that the weevils were capable of seeking out those untreated trees amongst a group of treated trees. This work raised the question of the scale at which this detection occurs, from the largest scale, of a tree that is treated or not, to a much finer scale of spacing between droplets of pesticide. This knowledge would have major implications for the future design of the DCS system, for example, the mistreatment of trees. During the field trial it was noted that some trees would not 'trip' the infra-red detector and therefore not be sprayed. The operators noticed many of these spray failures and the tree would be treated a second time. However, it is likely that some trees escaped detection. Therefore, they would be at risk of herbivory in the field.

The work in this section has further examined the aspects of pesticide detection. It was shown that *H. abietis* were able to differentiate between treated and untreated zones on both whole trees and twigs. Secondly, the level of damage was significantly greater when the AI nozzle was used than when the FE nozzle was used, which has a much finer spray structure with smaller gaps between droplets.

Each of these experiments examined a different type of coverage, from spray patterns likely to be found in the field (Section 7.3.1) through to more uniform patterns on twigs (Section 7.3.2). In the whole tree experiment it was not noted whether the damage came from treated or untreated zones, only whether the damage was above or below the 15cm high treated band. However, there was a clear reduction in the level of damage within the treated zone (at the base of the tree). This is contrary to the literature evidence (Hannerz *et al.*, 2002; Heritage, 1996) and also preliminary experiments that showed that *H. abietis* fed more heavily at the base of the tree (Appendix 2). Therefore, this shift in feeding pattern could be attributed to the effects of the pesticide.

In addition to the distribution of damage across the two zones, the different types of damage also had a significant effect on the level of damage. Trees with a full coverage of pesticide suffered much lower damage than either of the other two treatments, which effectively covered half the area of the full coverage. Therefore, the difference in levels of damage between these treatments could be attributed to the ability of *H. abietis* to detect pesticides on a food source. There was no difference between a pattern that covered half of the radius (like an open drain pipe) to one that consisted of a pair of bands at the top and bottom of the 15cm zone. It was thought at the outset of the experiment that if *H. abietis* followed its usual feeding pattern of moving from the bottom of the tree to the top, that the small band of pesticide at the base may be enough to prevent feeding or even kill the weevil. However, no evidence of this was observed.

One likely consequence of these different treatment patterns is that the half radius coverage should effectively prevent *H. abietis* from girdling the tree (feeding all the way around the stem), which is responsible for the death of the tree. Therefore, if a banding pattern were to be used, a half radius pattern would be better. However, the results here indicate that full coverage is necessary to prevent damage.

The second part of the experiment attempted to identify more precisely the level of feeding that occurs on treated and untreated zones of a twig. Twigs were used instead of whole saplings because they could be easily manipulated. The results in this section followed the same pattern to that of the previous experiment. There was a significant difference between different treatments, with greater damage occurring on twigs that were only partially treated. Observations of the results revealed that there was greater damage on untreated parts of the twig than on treated parts of the twig.

The final aspect of this section examined the level of detection at a very fine scale, examining feeding differences between two nozzles. It was hypothesized that there may be greater damage between the AI nozzle which has a very coarse spray with large gaps between droplets, compared to the FE nozzle that has a much finer spray with very small gaps between droplets. The results showed a significant difference in the level of damage between the controls and the treated twigs, reconfirming earlier work. More interestingly, the results showed that there was a significant difference between the AI and FE nozzles. More damage occurred on the twigs treated with the AI nozzle, as hypothesized. This could be attributed to either the larger gaps that occurred between droplets or the random feeding pattern of *H. abietis,* which results in damage occurring on the untreated gaps before the discovery of a pesticide patch which then inhibits further feeding.

This work has clearly shown that *H. abietis* is able to detect untreated and treated parts of saplings and twigs in the laboratory. In these experiments only individual insects were used, in contrast to a field situation where many more insects are likely to attack a tree. It would be expected that this problem would be intensified under these conditions, particularly as most of the trees and twigs in the previous two chapters showed at least a very minor level of damage. It is probable that *H. abietis* detected the pesticide by taste. Therefore under high population pressure, even treated bark may suffer substantial damage.

It is clear from these results that saplings to be transplanted into a field site need to be carefully treated. Any mistreated areas should that occur should not be vital to the tree's survival or result in a reduction in growth if the sapling is to establish and flourish. However, it is less clear exactly how *H. abietis* reacts to untreated and treated sections. In the last experiment in this section (effects of nozzle type) a thorough examination was made of every twig for very small damage marks, particularly bite marks, which are not usually measured. Bite marks usually show up as small exudations of sap or pin head size holes and have not been measured in previous experiments because they are easily drawn larger than their actual size in the measurement process. It was found that every treated twig had at least a small level of damage. Coupled with the effects of pesticides on the daily feeding damage shown in the recovery experiment in Chapter 6 (Section 6.3.3), it is very likely that *H. abietis* does need to taste the pesticide to detect it.

Pyrethroid insecticides work by disrupting the flow of ions through the sodium channels (Soderlund *et al.*, 2002; Vais *et al.*, 2001). The pyrethroid binds to the sodium channel causing a slow down of the action potential decay, which results in repetitive discharges in motor and sensory axons. The site of binding is different from that of other channel modulators such as anaesthetics and scorpion toxins (Vais *et al.*, 2001).

However, the behavioural effect and the knock-down effect may be a result of two distinctly different phases of pesticide poisoning (Gerard *et al.*, 1999). Initially, a very high dose of insecticide may be responsible for severe local effects. This is followed by more widespread effects as the pesticide penetrates to all parts of the insect. At this stage the pesticide will have reached the insect's central nervous system resulting in abnormal behaviour (Gerard *et al.*, 1999). However, these physiological processes do not necessarily explain the repellent nature of the pyrethroids.

Pyrethroids affect the nerve membranes and induce physiological effects, whereas repellents and anti-feedants affect the sensory organs (Saito *et al.*, 2000). In addition, it has been reported for permethrin, deltamethrin and DDT that the repellent component may be different from the knock-down component. Both mosquitoes and horn flies were repelled from these pesticides even when they were not able to touch the treated surface (Chareonviriyaphap *et al.*, 1997; Zyzak *et al.*, 1996).

For other chemicals the anti-feedant properties have been characterised. One of the most reported anti-feedant chemicals can be extracted from the Neem tree, azadirachtin (Mordue *et al.*, 1998). In *Spodoptera sp.* azadirachtin stimulates the deterrent neurones in taste sensilla (Mordue *et al.*, 1998). In other studies, host plant polyhydroxy alkaloids (PHAs) stimulated a dose dependent neural response from neurones in the styloconic taste sensilla on the mouthparts of different caterpillar species. These neurones produce action potentials in different ratios and it is these ratios that indicate host and non-host plants (Simmonds, 1998). Finally, in studies on non-alkaloid ryanoids, which are antagonists of the sodium channels, similar patterns of taste neurone interference were found (Gonzalez-Coloma *et al.*, 1999). Therefore, it is possible that the pyrethroids act on taste sensilla in similar ways.

The formulation of ι-cyhalothrin used in these experiments consisted of 10% active ingredient and 90% inert components consisting of an emulsifier and solvent. There are also other minor components that are not listed that are commercially confidential. It is possible that these non-active components, rather than the active ingredient are responsible for the repellent effects. Unfortunately, it was not possible to obtain samples of either the technical product or just the inert component. However, other studies have shown that both components may be important in repellency of permethrin (Lin *et al.*, 1993). In addition, the repellent effects of pyrethroids have been reported for a wide range of formulations and synthetic analogues, therefore it is likely that the active ingredient is the more important component (Herve, 1985).

The experiment described in section 7.2.3 ruled out other potential methods of detection such as smell and movement of the pesticide along the bark surface. Half of a twig was either treated or not, but the treated half was physically protected from damage. Therefore, the only method of detection was through 'smell' or movement of the pesticide along the bark surface. There was no significant difference in the levels of damage on any of the twigs. However, this is in contrast to experiments by Watson (1999), who found that weevils were significantly more attracted to untreated Scots pine twigs than permethrin treated twigs in olfactometer experiments. The difference in experiments may have been due to the different types of pyrethroid used, or the volatile components of the pesticide were contained by the tin foil wrapper. Also, the experiments by Watson (1999), used dipped trees that probably retained a greater volume of pesticide than the sprayed twigs.

It is likely that *H. abietis* shows no more awareness of the treated and untreated zones of the twig than simply that a particular patch is distasteful. Therefore, it is not able to minimise feeding on treated zones. However, there are generally only very low levels of damage on treated twigs indicating that *H. abietis* may move away from a treated food source, as reported in several experiments in Chapter 6. Increased rates of movement after pesticide exposure have been reported by a number of authors (Alzogaray & Zerba, 2001), as has the tendency of insects to move away from the treated zones (Chareonviriyaphap *et al.*, 1997).

In the small-scale detection experiment (Section 7.3.2) there were continually low levels of damage to the treated zones, indicating that *H. abietis* does not remember the treated areas. In this experiment, hunger and chance sampling of untreated areas probably led to the continued feeding on the untreated parts of the twig. This would explain the differences seen between the twigs treated with different nozzles. Twigs treated by the AI nozzle were patchier and would allow more feeding to occur before the weevil detected pesticide and stopped feeding. The trend in this experiment emulated the difference seen between high and low concentrations, so it may be that the weevil 'perceived' an effectively lower concentration of pesticide for the AI nozzle.

The low level of damage observed on treated twigs may in fact, be of no significance at all. The recovery experiment in Chapter 6 showed an increase in the feeding of *H. abietis* after pesticide exposure, with this same effect being recorded for other insects (Pinch, 2002, Unpublished). Therefore, the pesticide may stimulate the weevil to feed on the bark and it is in fact not sensing the pesticide by taste.

In summary, this section of work has highlighted the ability of *H. abietis* to detect the untreated and treated parts of a food source. However, previous choice experiments showed that *H. abietis* exhibit severe dislike for treated food if untreated food is available. Therefore in a field situation, except when under very high population pressure, incomplete treatment of a tree should be sufficient. Even when there is high population pressure and weevils are forced to feed on the untreated parts of a tree, their ability to detect the treated parts should prevent the weevil from girdling the tree and causing its death. This is important when the nozzle selection is considered, as Chapters 4 and 5 highlighted the beneficial effects of the AI nozzle. Although the biological efficacy of the AI nozzle may be slightly lower, the efficacy in the field should remain unaffected because of the repellent effect of the pesticide.

Chapter 8
A place for the DCS in European Forestry?

8.1 Summary of thesis

The work in this thesis has fallen into two distinct but related areas. The first has been the design and development of the Dandrive Conveyor Sprayer system (DCS). This system was shown to successfully treat transplanted saplings at a high daily rate and provide high levels of protection throughout the growing season.

The second aspect of this thesis has examined the ecology of planting treated saplings in a clear fell. It was predicted that the pesticides used may not kill the *Hylobius abietis* in the field, but merely prevent feeding damage occurring on the tree. A series of experiments were carried out to investigate this possible relationship and it was shown that the pesticides acted as an anti-feedant or repellent. Further extrapolation of this work also showed that when exposed to a pesticide treated food source the weevil lost a significant proportion of its body fat, but was able to recover if allowed to feed on untreated food. Finally, it was shown that *H. abietis* was able to detect pesticide at a very fine scale, targeting its feeding on untreated areas of both twigs and saplings. It was concluded that in a clear fell site, *H. abietis* is unlikely to feed on pesticide treated material and would exploit inadequately treated saplings. The implications for the DCS system are fairly obvious and it is important to ensure all trees are correctly treated and thus prevent feeding damage between pesticide deposits. However, it is girdling that kills the trees and partial treatment should prevent weevils feeding all the way around the stem.

8.2 The future of pesticides in European forestry

Currently the future of pesticides in forestry is very unclear and differs from country to country. There is strong public pressure to reduce the use of pesticides in both forestry and agriculture. This is in part fostered by environmental and media pressure groups that present all pesticides as being highly hazardous and poisonous (Hessayon, 1983). In addition, many scientists have political agendas in reducing the use of pesticides so that their research into other areas may continue to be funded (von Hofsten H., *Pers. Comm.*). The use of pesticides in forestry is far less than that of agriculture, most foresters only treat the saplings two to three times and perhaps occasionally in mature forests against other pests. The main treatment of the saplings occurs in a nursery building, reducing the risk of environmental exposure. In contrast, agricultural treatments are often more numerous and typically use higher doses (Dehne & Schonbeck, 1994). Unfortunately arguments of relative pesticide use do not serve to reduce public and government pressure.

In Sweden, the use of permethrin will be prohibited after 2003 and the final use of pesticide treated trees will be the spring planting in 2004 (Orlander G., *Pers. Comm.*). Therefore there is substantial pressure on researchers to find alternatives before this deadline. The Swedish forestry groups are focusing research onto silvicultural methods to reduce damage in possible conjunction with physical barriers. There is currently and deliberately no research into the use of biological control agents due to perceived difficulties in achieving widespread implementation.

In contrast, Denmark has received registration for several new pesticides and appears set to allow pesticide use into the foreseeable future (Christiansen P., *Pers. Comm.*). These pesticides include α-cypermethrin and es-fenvalerate. However, the public owns a third of the forests in Denmark in which pesticides may not be used after 2003. In these forests alternatives are being sought. Additionally overall pesticide use targets are being reduced to two-thirds of the 1995 level. Currently protection of these saplings costs around 713,000 Euros or £452,000 per annum (Viiri, 2001).

The use of pesticides in Denmark may have implications for the neighbouring Scandinavian countries (Finland, Sweden and Norway). It was suggested that as long as pesticides are registered in one of these countries, the neighbouring countries would not withdraw pesticide use (Christiansen P., *Pers. Comm.*). This, however, is a matter of serious debate and as mentioned, political motives are certainly involved. To date, the deadline in Sweden has been continuously postponed since the late eighties.

The remaining Scandinavian countries have adopted different stances. Whilst Finland is following in the footsteps of Sweden moving towards a pesticide ban in forestry, Norway appears set to continue with the use of pesticides. In Finland, the ban is not imminent, but steps are being taken towards replacing pesticide use with fungal and natural enemies to control *H. abietis*. Many saplings are imported from Sweden, up to 10 million annually and many of these are untreated. Additionally, there have been silvicultural changes such as use of containerised stock and soil preparation. However, this has devalued previous research and new research needs to be carried out to understand *H. abietis* under different conditions (Voolma K, *Pers. Comm.*).

Current research in Norway is less focused than in Sweden where there are large collaborations investigating *H. abietis* control. The research in Norway is investigating the use of nematodes and the electrophysiology of chemoreception (Salinas S; Wibe A., *Pers. Comm.*). Typical damage levels in Norway are only 30% for untreated saplings and as many as 80% are treated annually with permethrin (GORI 920LX) or bensultap (Bancol) (Viiri, 2001).

Finally in Estonia the use of pesticides in forestry is very low, only 30kg of active ingredient was used in forestry enterprises in 2000. This consisted of primarily alpha-cypermethrin and deltamethrin. In addition, a few other pesticides have been registered, diflubenzuron (Dimilin) and cypermethrin. There is also a Bt product (DiPel) registered. The application of pesticides is very restricted and requires a forest protection expert to examine the forest first, except in the case of *H. abietis* (Voolma K, *Pers. Comm.*).

In the United Kingdom (UK) and Republic of Ireland (ROI) pesticides are likely to play an important if not solitary role in forestry for some time. Research programs into the use of nematode species as described previously are well underway and providing promising results (Heritage S.G., *Pers. Comm.*). With the more relaxed registration procedures for releasing endemic species than for other species or pesticides, nematodes could become an effective population control method. Unfortunately, the density of weevils in the field is often far in excess of the number of weevils required to kill a sapling. Therefore, it is unlikely that the population could be reduced sufficiently to prevent the use of pesticides, at least until the technology has been substantially developed.

Development of low volume application systems in the UK such as the Electrodyn and the DCS will hopefully lead to a reduced environmental impact compared to the dipping systems. However, in the ROI the use of pesticides has not had the developments seen in the UK. Due to planters' concerns for their safety in handling treated trees, dipping of trees has been stopped and 90% of trees are treated with as much as 20ml of formulated permethrin (1-2%) *in situ*. The pesticide is applied with a knapsack sprayer and is applied until it runs into the soil ('run off'). The soil is deliberately treated so that 'nesting' *H. abietis* are treated. Although permethrin binds strongly to soil and may not move very far (Torstensson *et al.*, 1999), this is very bad practice. There is generally high precipitation and many interconnecting waterways in Ireland and this could lead to leaching of the pesticide as well as detrimental effects on beneficial Coleoptera due to the low specificity of permethrin.

The necessity to apply pesticides twice a year in these forests, demonstrates the possible ineffectiveness of this system. It may be that the pesticide is washed from the saplings by the rain before it dries and becomes rain fast. Some Irish foresters freely admit that they apply pesticides at times when they should not (Ward D, *Pers. Comm.*). Appreciably the weather in Ireland can hamper efforts to treat trees, but if there is rarely the correct weather to treat the trees *in situ*, they should be treated indoors. In contrast the DCS system applies 4ml of formulation (1-2%) per tree with similar nozzles and achieves effective yearlong protection even under high population pressure.

The remaining 10% of trees in ROI are treated in a mechanised dipping system. This is very similar to the high throughput dipping system in use by the UK Forestry Commission. In this mechanized system fewer human operators may be required and they are generally further removed from the pesticide bath. In their place is a clamp that tightens around a tree bundle as they are dragged through a vat of pesticide. Although there is less risk of potential operator exposure, there is no reduction in the risk to planters and other handlers. These trees are too damp to handle and must consequently be stored in a humid store for several days before they are dispatched to the clear fell. Shortly after the installation of this system there were several setbacks. Many of the treated saplings were returned to the nursery because they were too wet to handle.

In summary, the use of pesticides across Europe is highly variable. There is a definite need for improvement in some countries such as the Republic of Ireland. Therefore, there is potential for the use of the DCS system outside of the UK in the rest of Europe.

8.3 Use of the DCS system in Europe

In light of the current forestry situation across Europe, the possibility to export the DCS to other countries could be promising. Many of these countries use dipping techniques and until recently Irish foresters carried this out in the field. In addition, there are no application systems in use in Denmark or Estonia for the treatment of saplings (Viiri, 2001). This was stopped due to concerns that a spillage would cause severe public upset. The automated dipping system described earlier in use by Irish foresters, was imported from Norway and it is likely to be in used in Norwegian nurseries.

The dipping of saplings to protect them from *H. abietis* may not only be potentially unsafe, but also suffers from low efficacy. EC formulations of permethrin tend to come out of solution and float on the surface, as they are not fully water-soluble. This is often referred to as 'creaming'. This usually occurs when the solution is not agitated and left for a period of time, for example, over the weekend (Matthews, 2000). Spot checks within the Forestry Commission have shown that the concentration on the surface may be over thirty times the desired amount. The first few trees to be dipped are likely to remove most of the pesticide, whilst those dipped later are only treated with a very dilute solution (Heritage S.G., *Pers. Comm.*).

The DCS system, therefore, would be of great value in a treatment centre both because of the decreased risk of exposure and the consistency of the treatment. With the suggested modifications, a second important feature of the DCS system will be its resistance to tampering. Although the current prototype has infinitely adjustable controls, a manufactured version would have an enclosed control panel that would not be available for the nursery staff to adjust. The indication of preset loading areas along the conveyor belt should discourage the workers from overloading the system in order to increase daily output. However, providing that there is at least a 0.5 second gap between trees (the time taken to spray) the infrared system should ensure that every tree receives the required dose. This is not the case for the Electrodyn and reports have been made of operators putting in up to 16,000 trees per hour when the system only delivers enough spray to treat 4000 trees per hour (Heritage S.G., *Pers. Comm.*).

The Electrodyn system effectively treats trees with ultra low volumes of pesticide, although it is unclear how effective it would be for very bushy trees. In trials it achieved levels of protection only slightly lower than the DCS system. Also, the Electrodyn system is already installed in many establishments in the UK. However, it also requires a carefully formulated spray solution that needs extensive costly registration with the Pesticide Safety Directorate (PSD). Additionally, this has to be repeated each time the pesticide is changed, such as with the replacement of permethrin. In contrast, the DCS system will be able to use a range of 'off the shelf' products with no reformulation necessary and therefore minimal registration time and costs will be incurred. This would make it ideal for use in other European countries, which may have different availability of pesticides.

In summary, there is significant potential for use of the DCS system in Europe. It is both safer and more reliable than the dipping system whilst providing equal levels of protection. Secondly, it is quicker and cheaper to install and set up than the Electrodyn and also substantially more robust in the ability to treat a range of saplings and more flexible in the use of pesticides. The work in this thesis has demonstrated that the system has significant potential to provide prophylactic protection to transplanted saplings. In addition, this work has investigated a range of parameters and presented the optimal settings to give maximum deposition on the saplings.

8.4 The role of repellents and anti-feedants in forestry

Despite the substantial research into the use of repellents and anti-feedants in forestry (Schlyter *et al.*, 1987; Zhang *et al.*, 2000), the potential for their future use is not especially promising. There are a variety of reasons for this. Firstly, although they are often derived from plants, the ultimate product would almost certainly be manufactured synthetically. Therefore, these products are required to conform to many of the same costly registration procedures that pesticides suffer (Heritage S.G., *Pers. Comm.*). Secondly, many of these molecules are extremely complex and cannot be easily manufactured and often do not live up to farmer's expectations (Plimmer, 1996; Xie *et al.*, 1995). Finally, despite the natural source of the chemical, many of these can be as toxic, if not more toxic than pesticides to humans, for example, juglone, an extract from the walnut plant (Schlyter F., *Pers. Comm.*) (Girzu *et al.*, 1998; Willis, 2000).

The main advantage of these chemicals is that they generally do not harm other insects or wildlife, as they are specific to the pest species (Dent, 1993). For many insects, the deprivation of a host plant because of the applied anti-feedant would result in a decrease in numbers due to the lack of abundance of suitable alternatives. However, this is not the case for *H. abietis*, which will readily feed on a variety of sources in a field site (Orlander *et al.*, 2000). In addition, the specificity of pesticide application, particularly by the DCS system, ensures the pesticide is very selective. This is because only those insects feeding on the sapling will come in to contact with the pesticide. Therefore a wide range of pests can be prevented from damaging the sapling. In contrast, many anti-feedants may be specific to particular species (Dent, 1993). In forestry, *Hylastes sp.* could become a significant secondary pest if anti-feedants were used instead of pesticides.

The research in this thesis, however, shows that there is potential for anti-feedants to be used. The property of the pesticides to repel rather than to kill the weevils clearly demonstrates this. However, many anti-feedants are not as stable in the field as pesticides and their effects may be required for longer. Therefore, effective formulations are required, particularly for *H. abietis*, which is present all year round and typically there are two periods when peak levels of damage occurs.

8.4 Future work

Although the work in this thesis has usually answered and explained all of the results found, there are some areas for which further work may be beneficial. One such aspect is the disparity between actual field observations and the laboratory work. It was hypothesised that weevils in the field would avoid treated trees and find alternative food sources. However, field trials rarely achieve 100% or even 90% protection except at the lowest population levels. Therefore, it is likely that many of the relationships demonstrated in the laboratory break down under high population pressure. This could be for a variety of reasons. Firstly it may be that each weevil causes a small amount of feeding damage before moving onto other sources. When there are up to one hundred weevils per tree at high population levels, the damage from each weevil could additively cause the death of the sapling.

An alternative explanation is that many trees are inadequately treated and the weevils are able to exploit this. There have been reported problems for all of the application systems. Even the dipping system, which is expected to totally immerse the saplings may fail if operators do not thoroughly ensure that the trees are dipped deep enough so that the root collar is treated. The trees are dipped top first and mistreatment could quite easily occur. Studies of the Electrodyn experimental procedure revealed that only those trees with exposed lower stems are treated. This indicates that there may be a known deficiency in the ability of the Electrodyn to treat bushy plants. Observation of the spraying of trees would suggest that much of the spray is attracted to the needles rather than the stem. Finally, even the DCS system may have not adequately treated all of the trees in the field trials. The system was still an early prototype and the targeting of trees with the spray was not 100% accurate at this stage.

In summary, further experimentation examining the type of damage that occurs and attempting to gain an understanding as to why this damage occurs on treated trees would be advisable. Additional experiments investigating the effects of different DCS set-ups could be carried out, although it is likely that the most effective settings have already been tried.

8.5 Conclusion

This work has clearly demonstrated that the safety and efficacy of the current treatment systems in forestry can be improved, in particular by the use of the DCS system. This work has also demonstrated that the effects of pesticides have previously been poorly understood and that a greater understanding of the effects in the field could lead to increased protection of transplanted saplings against the large pine weevil. The future of forestry is unclear, the demands of the public are ever changing and the regulations imposed by authorities swiftly alter, but hopefully the work shown here can be another stepping stone towards our goal of maximum protection of forests with zero environmental impact.

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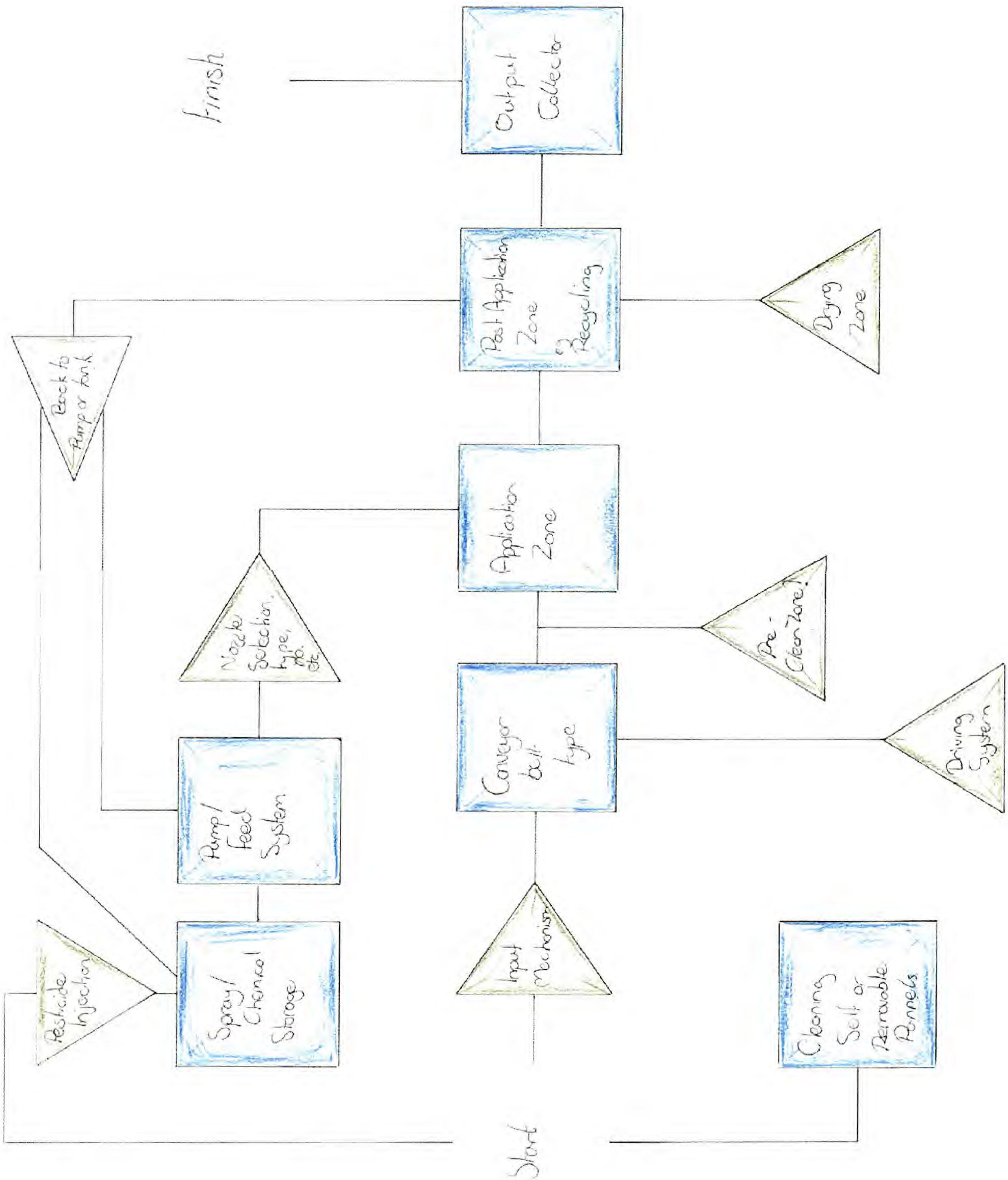
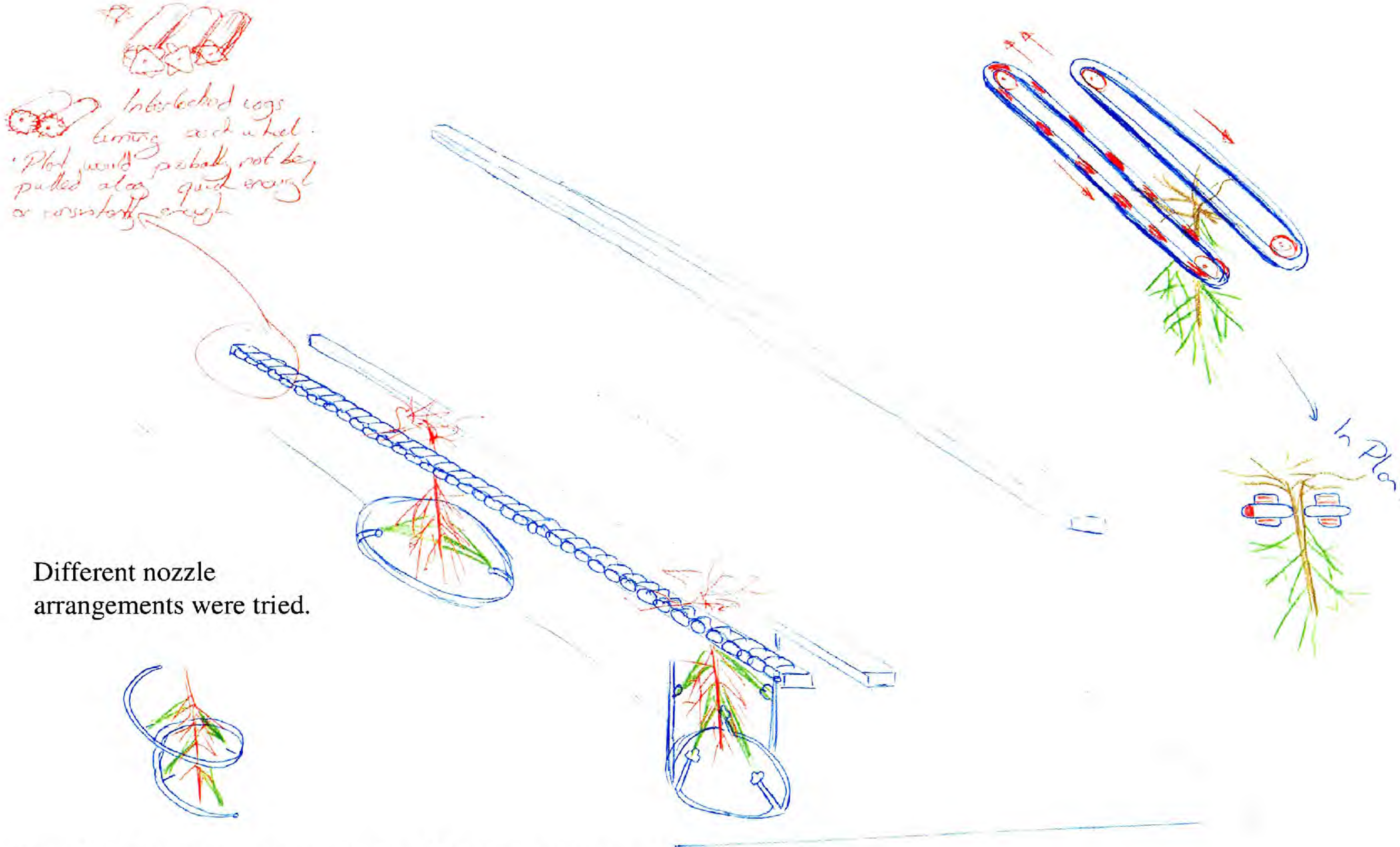


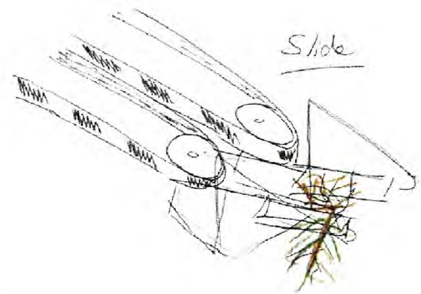
Finish
Output Collector
Post Application Zone & Recycling
Drying Zone
Back to Pump or tank
Application Zone
Nozzle Selection, type, no. etc
Pre-Clean Zone?
Conveyor belt type
Driving System
Pump/Feed System
Spray/Chemical Storage
Pesticide Injection
Input Mechanism
Cleaning Self or Removable Pannels
Start



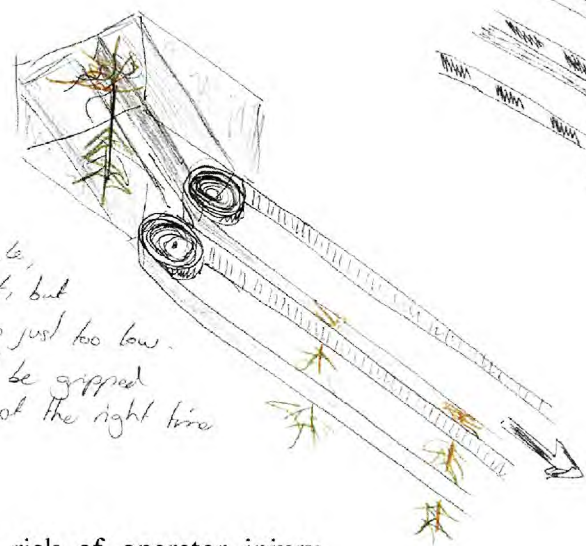
Different nozzle arrangements were tried.

Several ideas were generated for different conveyor belt types that would present the trees to the nozzles in a vertical position. This idea used a series of rollers over which the roots may hang. In reality the trees were unlikely to be so obliging.

Loading Mechanism



Slide



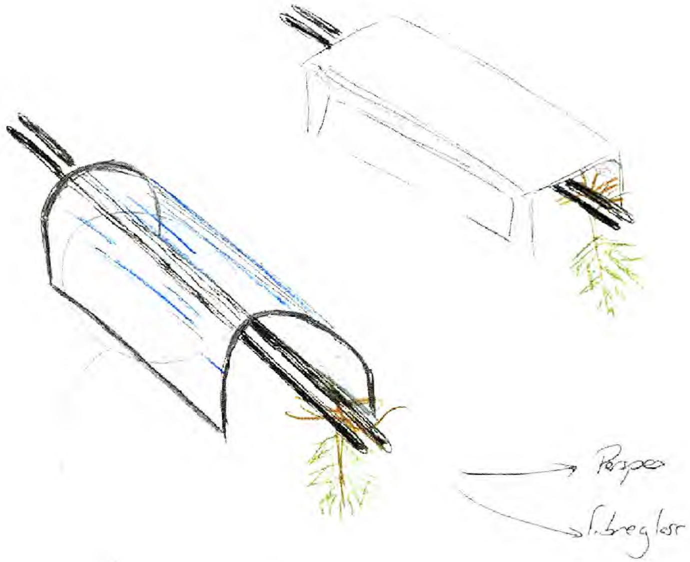
Slide

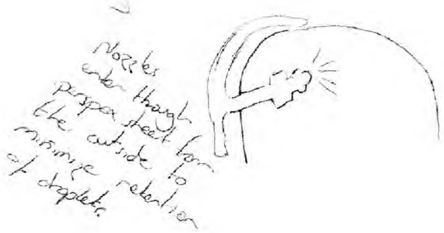
Slides are simple, slides are great, but the accuracy is just too low. The plants won't be gripped properly & taken at the right time

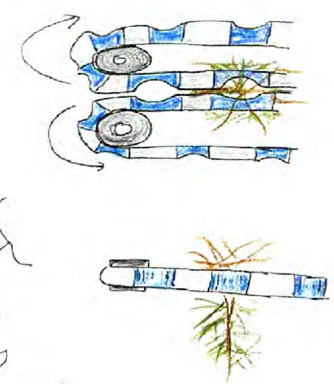
To reduce the risk of operator injury during tree loading (as there is potential for a limb to become caught between the belts), several ideas were generated to automate the process. These ideas were later scrapped as it would require very complicated hardware to separate bundles of trees. Therefore the tree loader itself would need to be loaded!

Carousel.

More complicated device. Shield prevents loaded trees from falling out. These are then taken around to be fed into the belt. However tree must be held in place until shield captures it. This could lead to trapped fingers. One section of conveyor belt drives wheel around. I don't think the gain in safety is that great but the increase in complexity is a bit much





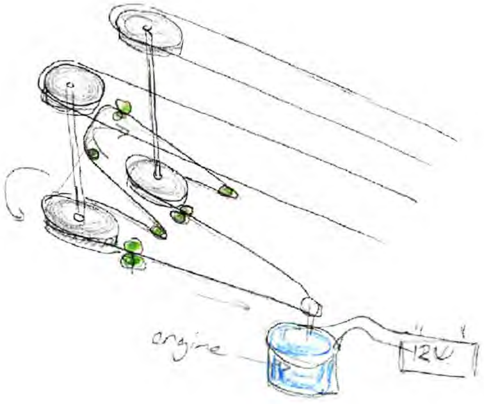


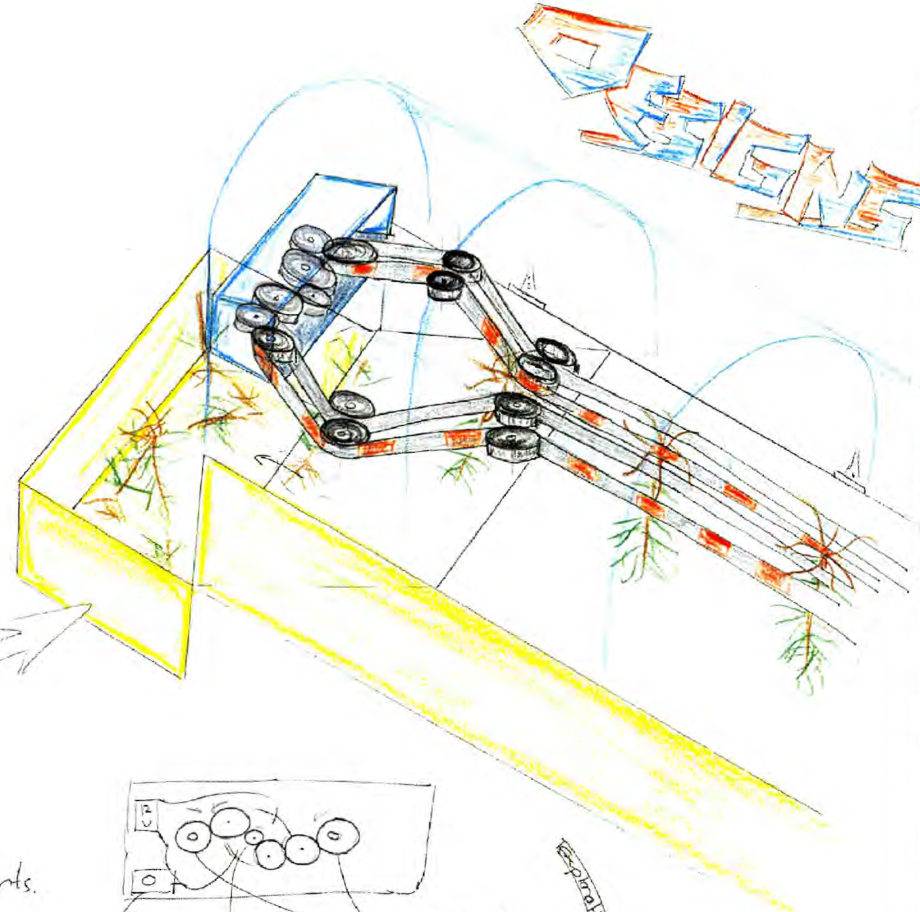
Key Points

1) Sapling must sit low in track, possibly clamped on roots to ensure spray on stem.
2) Only groove one track so that slippage will not cause a mis-alignment problem.
3) Guarded gears/cam wheels from plant roots.

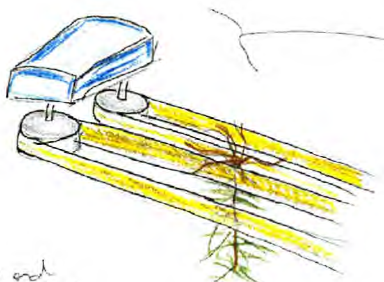
Further design work improved on the vertical tree idea and investigated the use of grooves on the belts to trap the trees.

Configuration of Cams & cam belts. Probably not the best possible design, risk of slippage, breaking and seizing it up.





Would prefer a compact engine design Possibly sitting on top, food mixer style



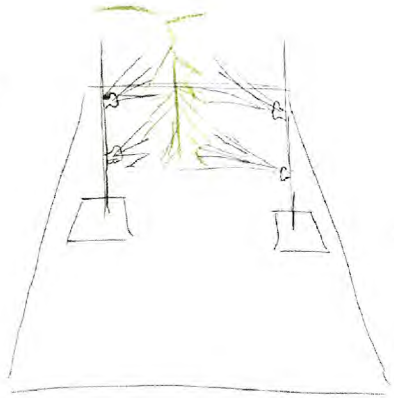
Problem

Drive wheels at the end need to be protected but it is also the point where trees are released.
- See development

The DCS system starts to take shape. Mechanisms to link the drive with the conveyor belts were investigated to remove the potential for tree roots becoming trapped in the gears.

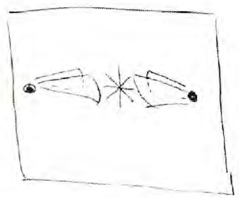
With this design the arms open out & drop the trees into a catchment box. The trees have dropped before they get near the engine & ∴ no jamming. The engine section will be closed but the required arrangement of gears are shown. The entire shield (if light) could be lifted up like a trunk lid.

7



The positioning of the nozzles would be critical to optimising the efficacy of the system.

Speed of Pass should average 5seconds per pass or $\frac{1}{5}$ = 0.2m/s. This equates to 12metres a minute. At this rate 72 plants are treated per minute. 4320 per hour & 34,560 per 8hr day. Later experiments may feature a second higher speed eg 50,000 plants would be at a speed of 20 m/min.

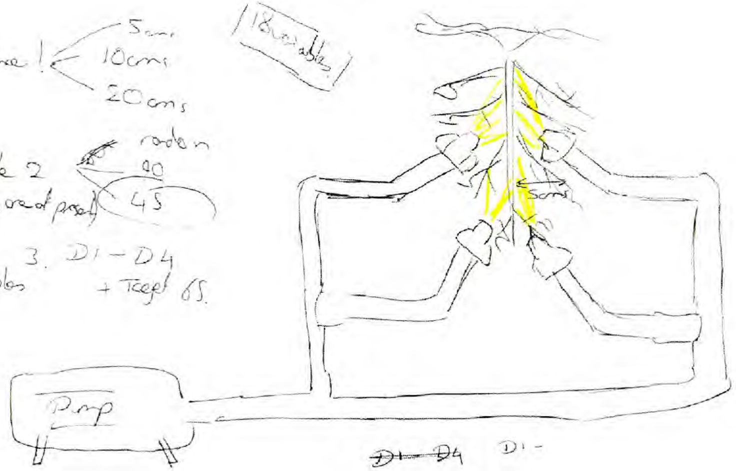


18variables

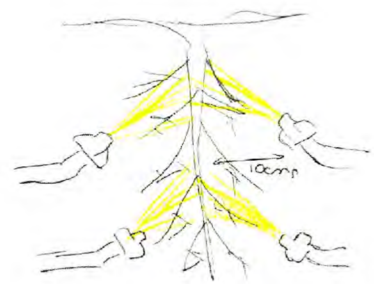
Distance ! 5cms
10cms
20cms

Angle 2 (only one of prob) random
90
45

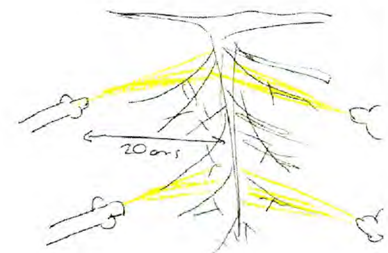
3. D1 – D4
Nozzles + Teejet 65.

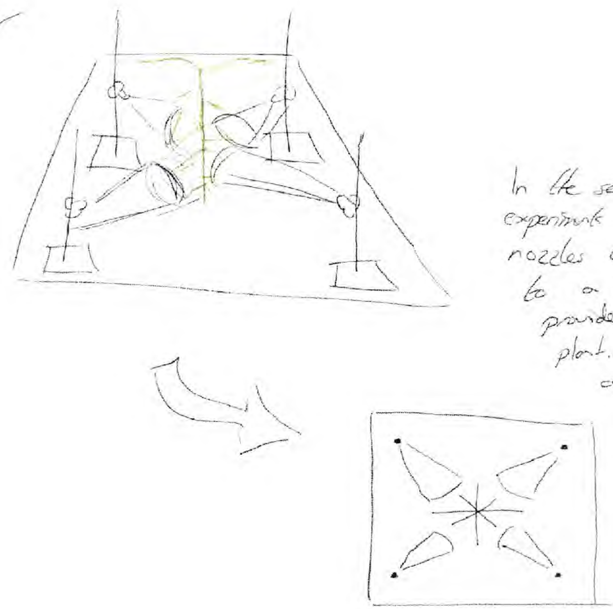


~~D1 – D4~~ D1 -

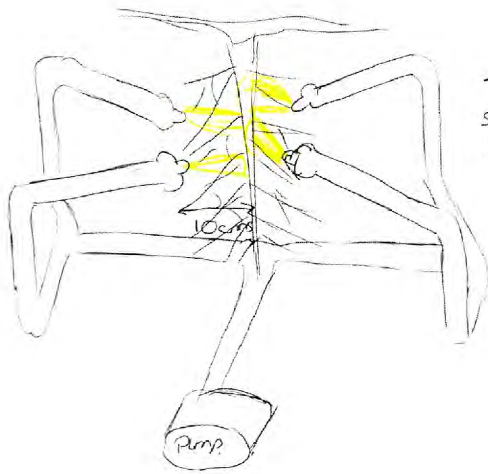


Use all nozzles on all setups unless some obviously are not getting complete coverage then remove end ones

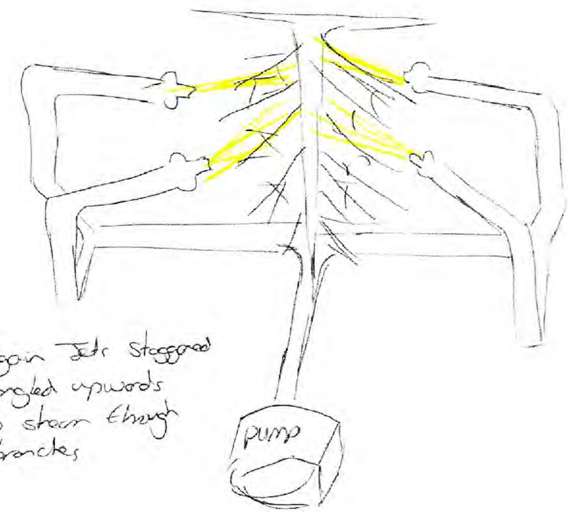




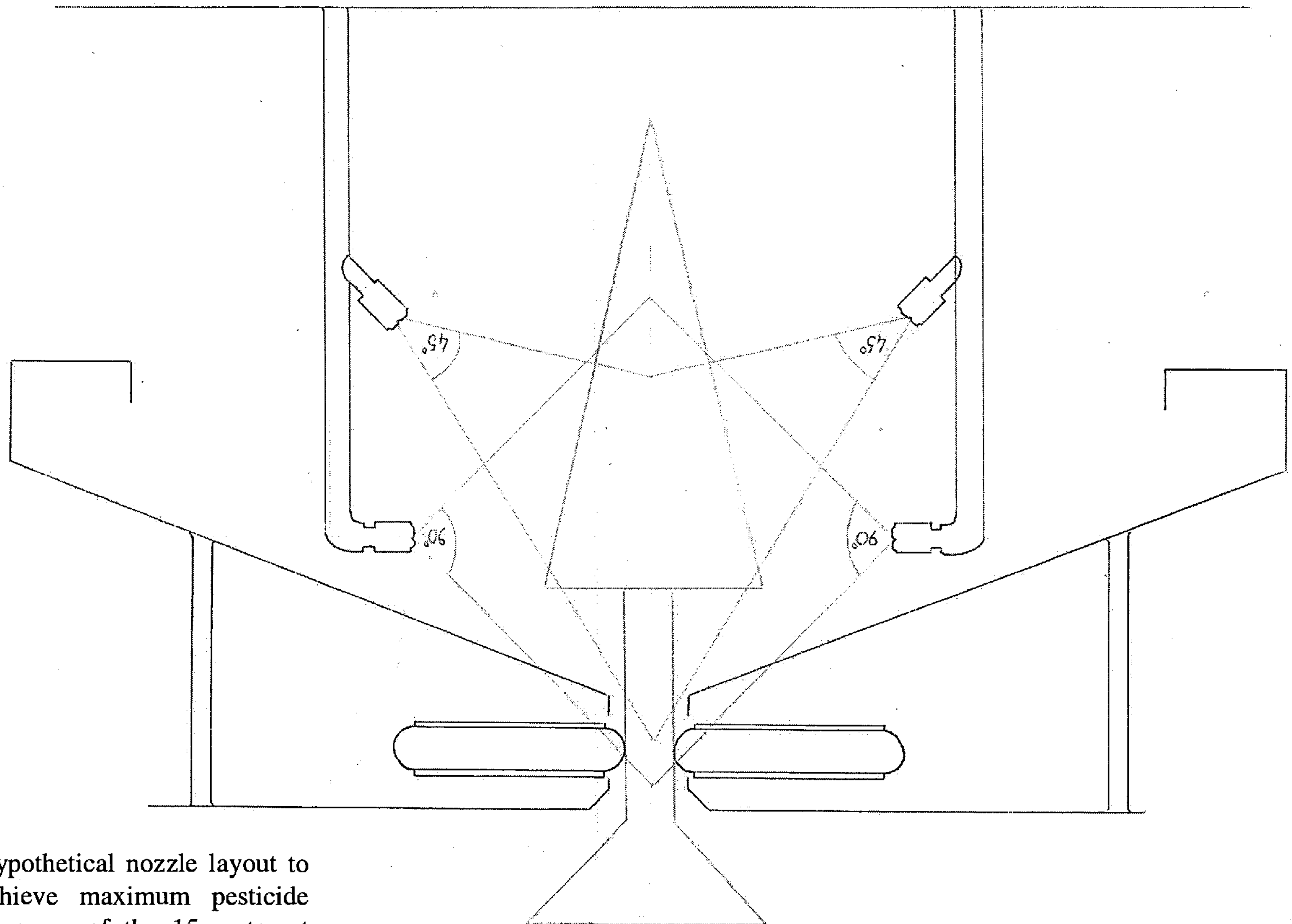
In the second part of the deposition experiment an array of 4 separate nozzles will be used, as opposed to a 2×2 array. This may provide different coverage of the plant. However I fear that as the plant is occupying a space in a 3D area, it may be harder to get the timing right. I will let the experiment decide!



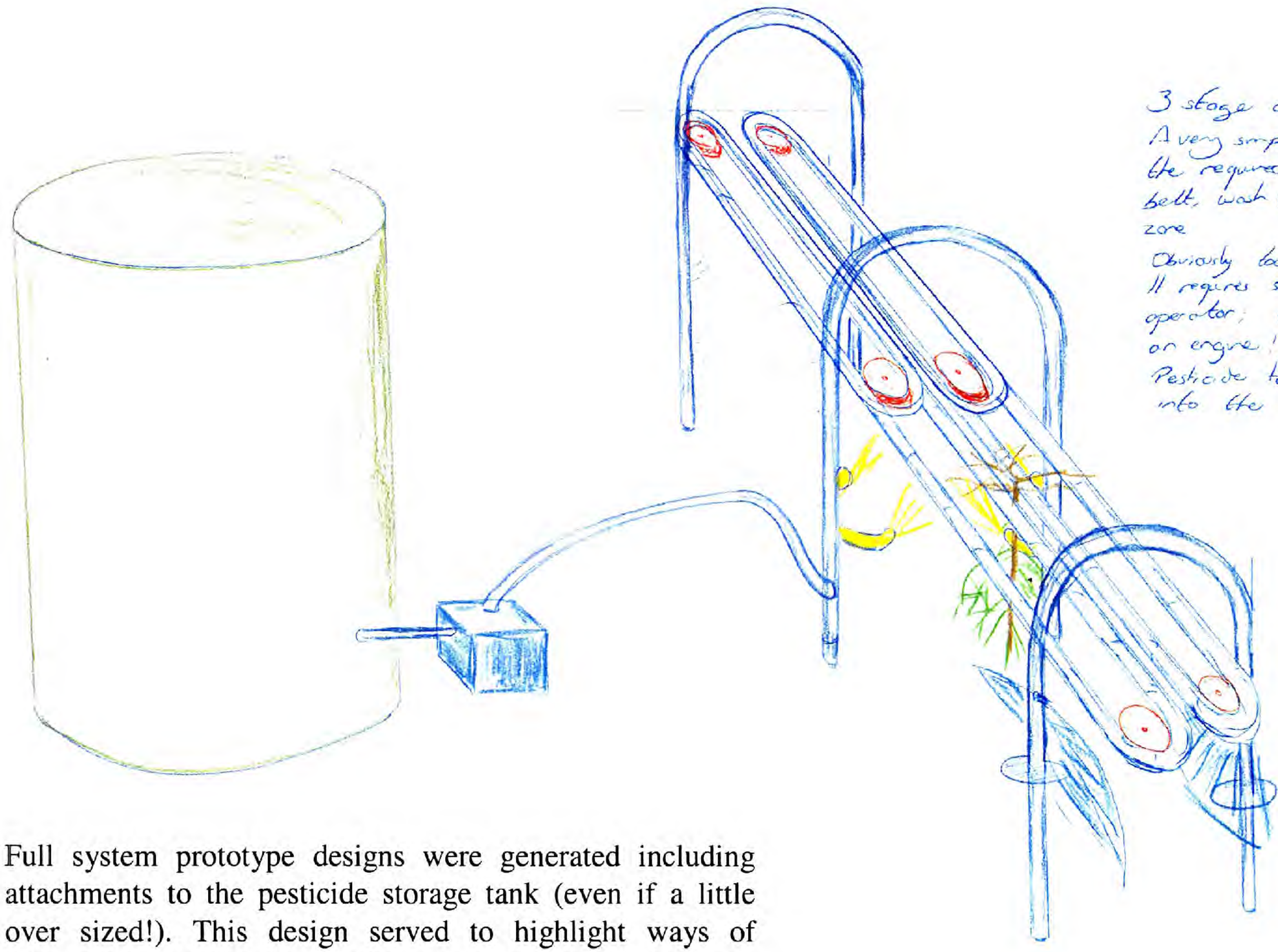
Jets are Staggered, so they all aim at Slightly different Spaces



Again Jets Staggered but angled upwards to stop stream through the branches

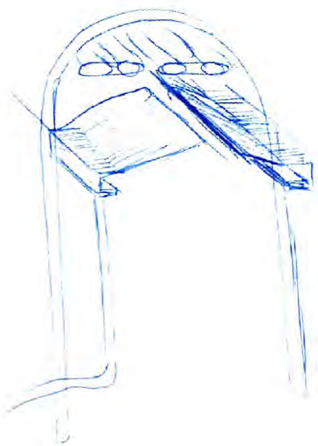


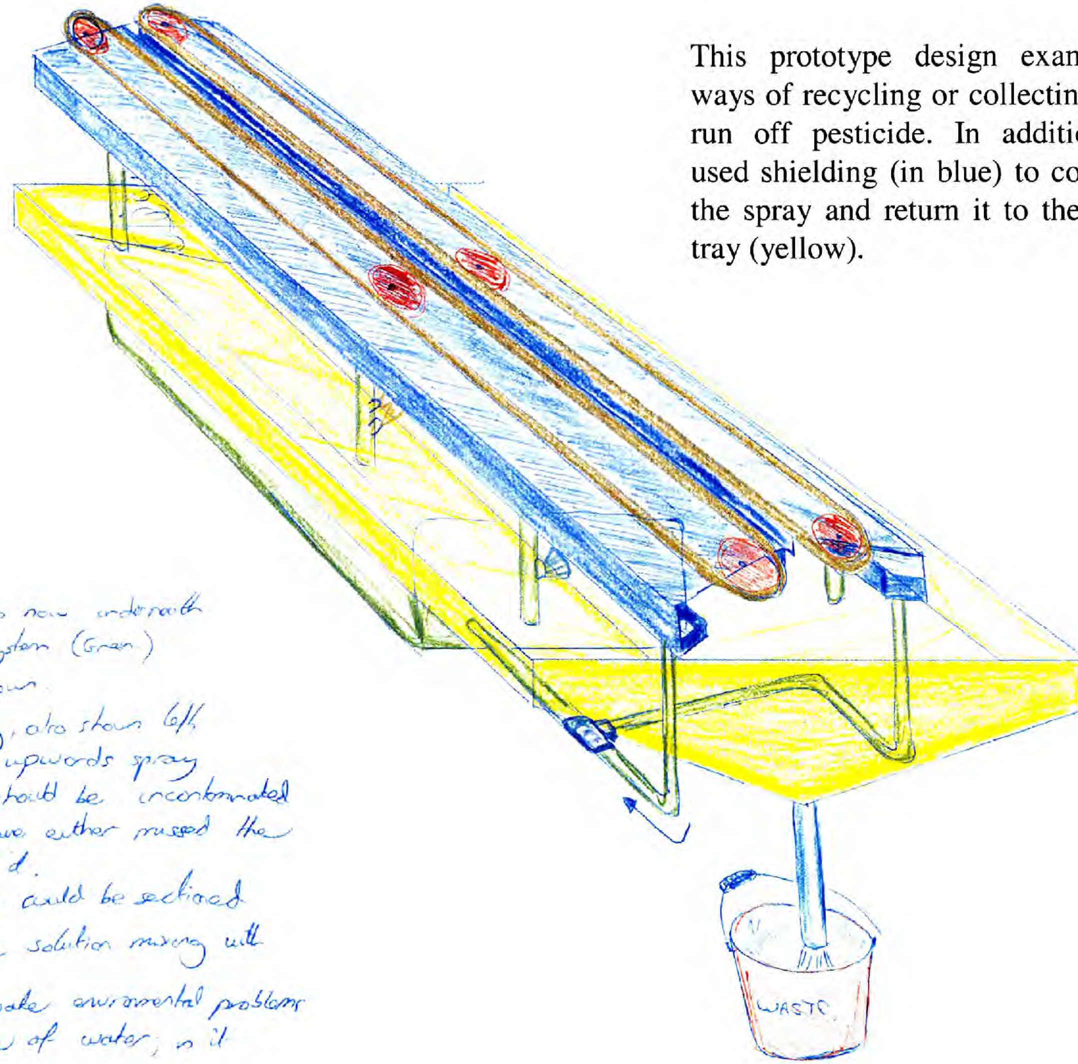
Hypothetical nozzle layout to achieve maximum pesticide coverage of the 15cm target zone.



3 stage conveyor belt
A very simple design incorporating the required components; conveyor belt, wash zone, spray zone, drying zone
Obviously too basic at the moment. It requires shielding to protect the operator; a plant collection zone, an engine!, etc.
Pesticide tank should be incorporated into the unit itself.

Full system prototype designs were generated including attachments to the pesticide storage tank (even if a little over sized!). This design served to highlight ways of integrating nozzle driers, cleaners and sprayers into the bodywork and structure of the system.

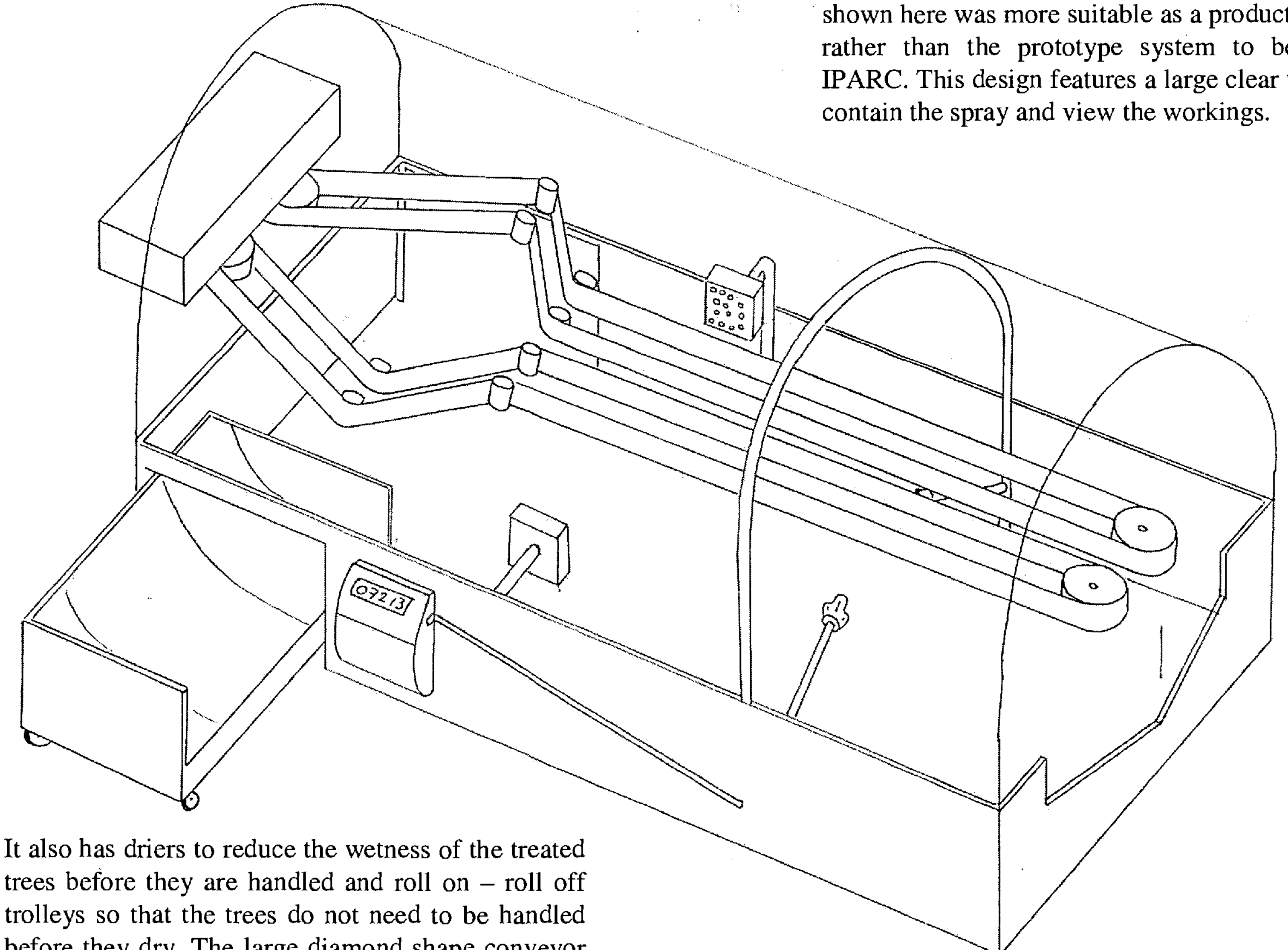




This prototype design examined ways of recycling or collecting the run off pesticide. In addition it used shielding (in blue) to contain the spray and return it to the drip tray (yellow).

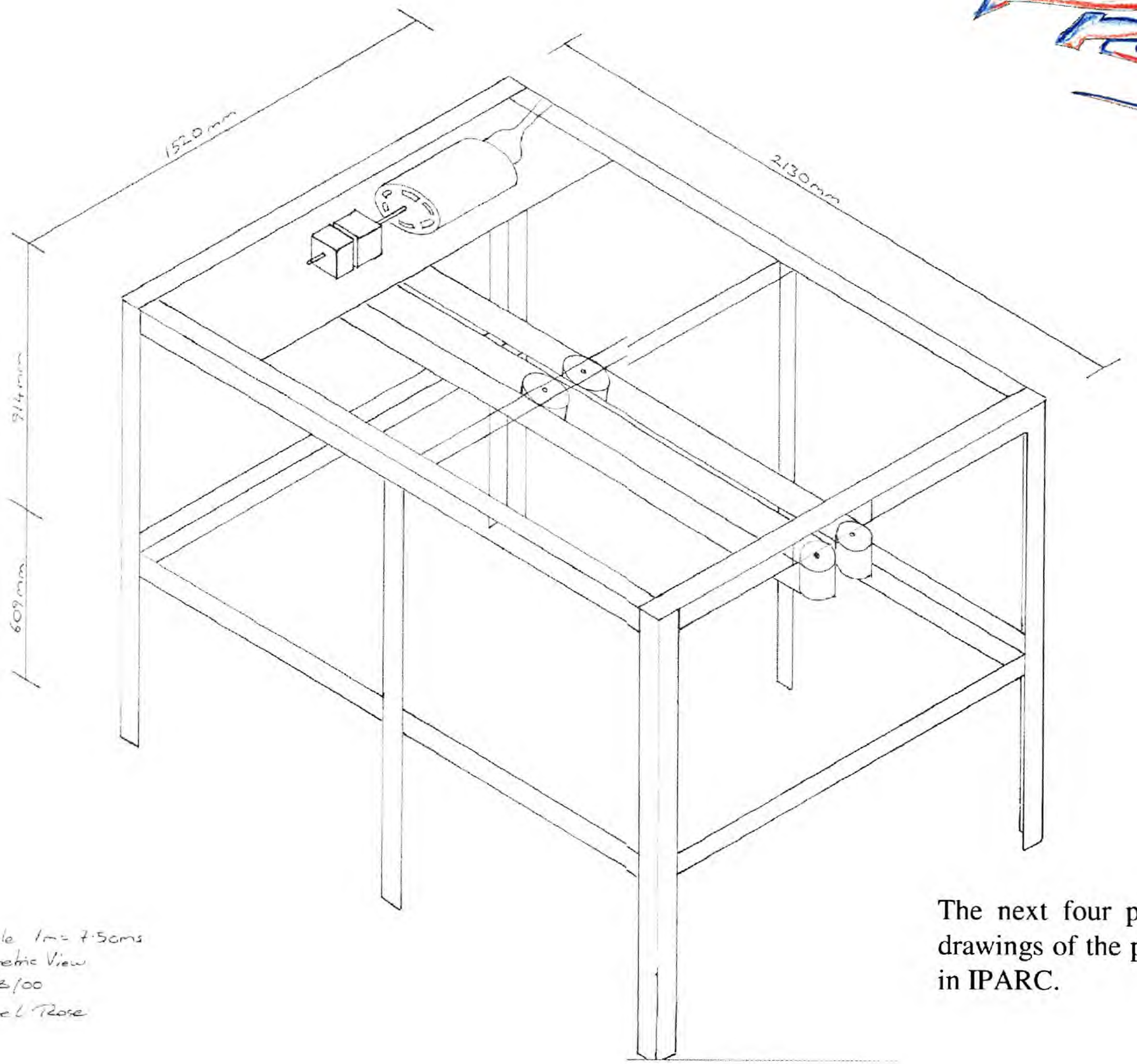
The storage tank is now underneath
the conveyor belt system (Green)
Pumps & Engines not shown.
The above shielding, also shown b/k
(Blue) will collect upwards spray
+ collect it. This should be uncontaminated
because it will have either missed the
tree or bounced off of it.
Large trough (yellow) could be sectioned
to prevent pesticide solution mixing with
pure water.
Wash section may create environmental problems
high waste volume of water, is it
contaminated?

The prototype DCS was finally designed. The model shown here was more suitable as a production model rather than the prototype system to be built in IPARC. This design features a large clear window to contain the spray and view the workings.



It also has driers to reduce the wetness of the treated trees before they are handled and roll on – roll off trolleys so that the trees do not need to be handled before they dry. The large diamond shape conveyor belts were intended to drop the trees before they reached the drive system.

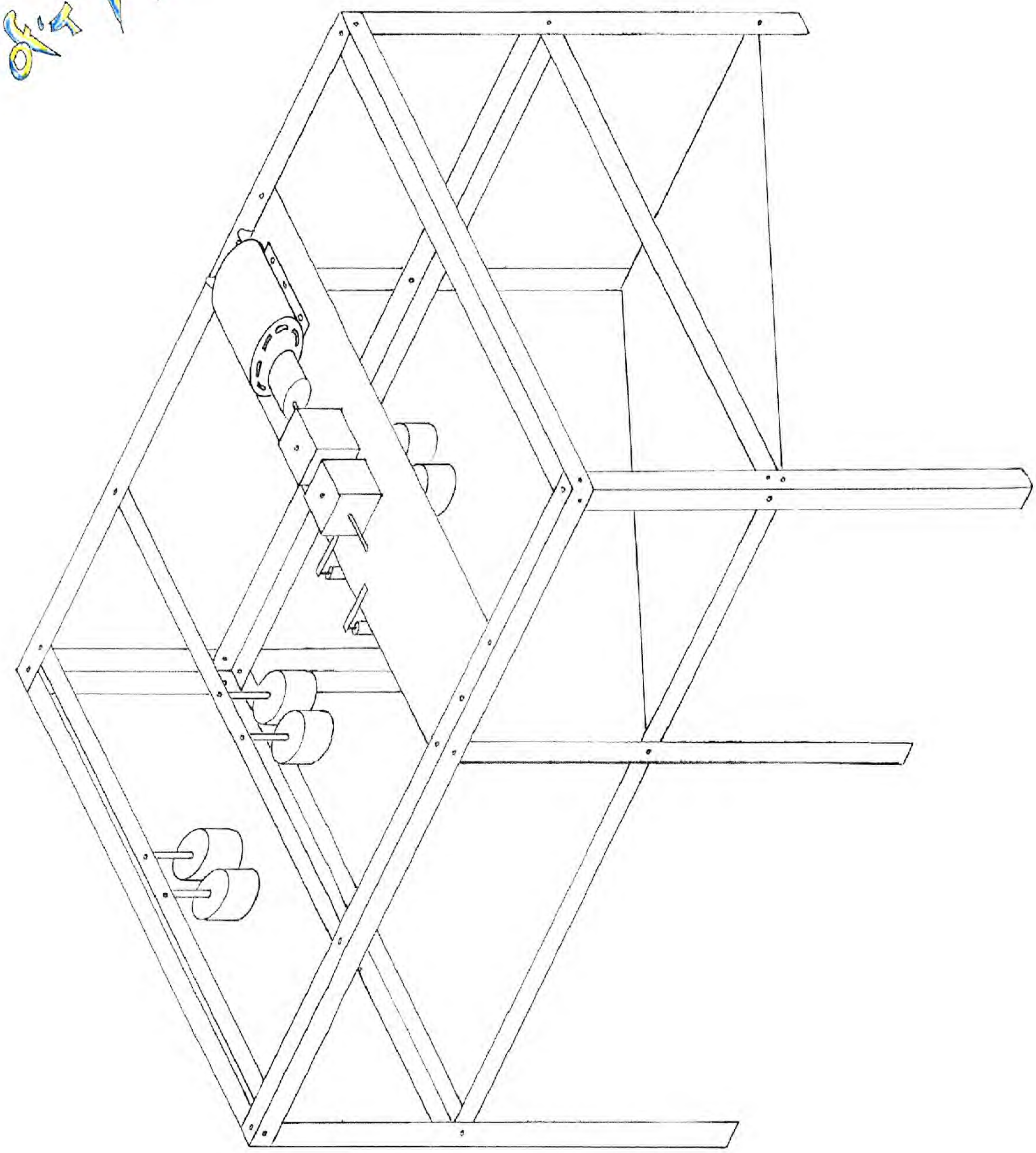
Prototype

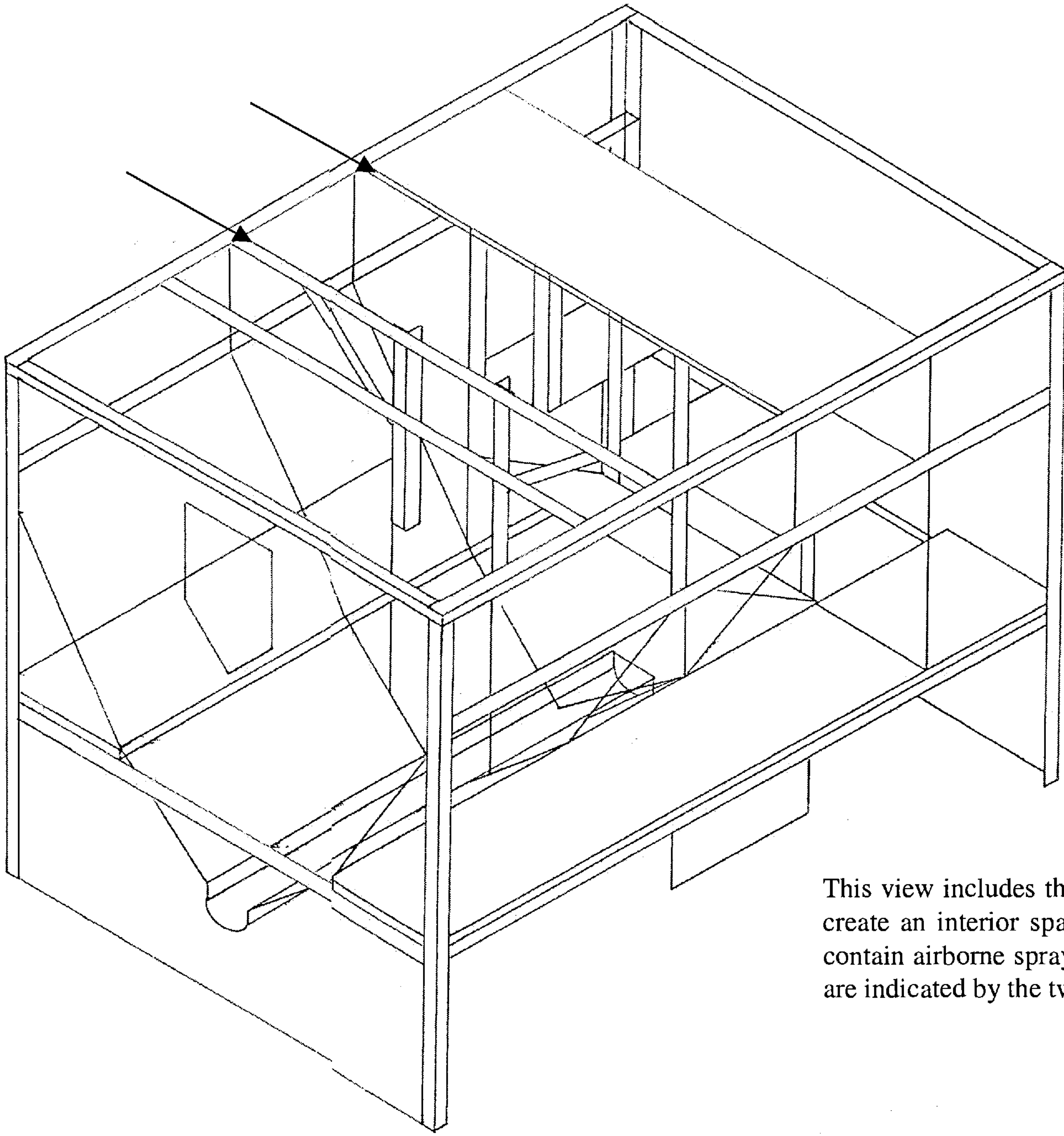


Scale 1m= 7.5cms
Isometric View
7/08/00
Daniel Rose

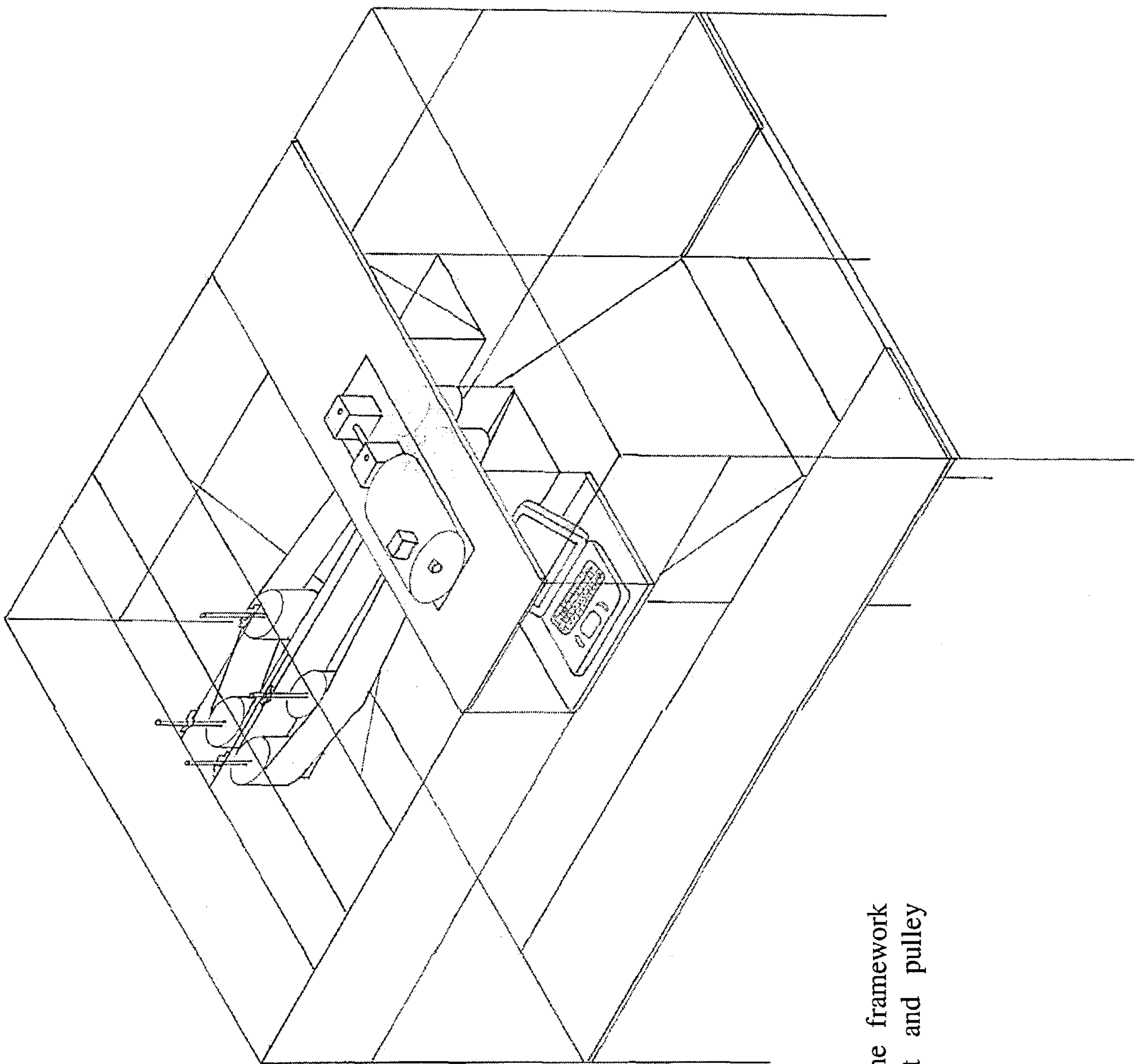
The next four pages feature isometric drawings of the prototype that was built in IPARC.

Rear View of Prototype

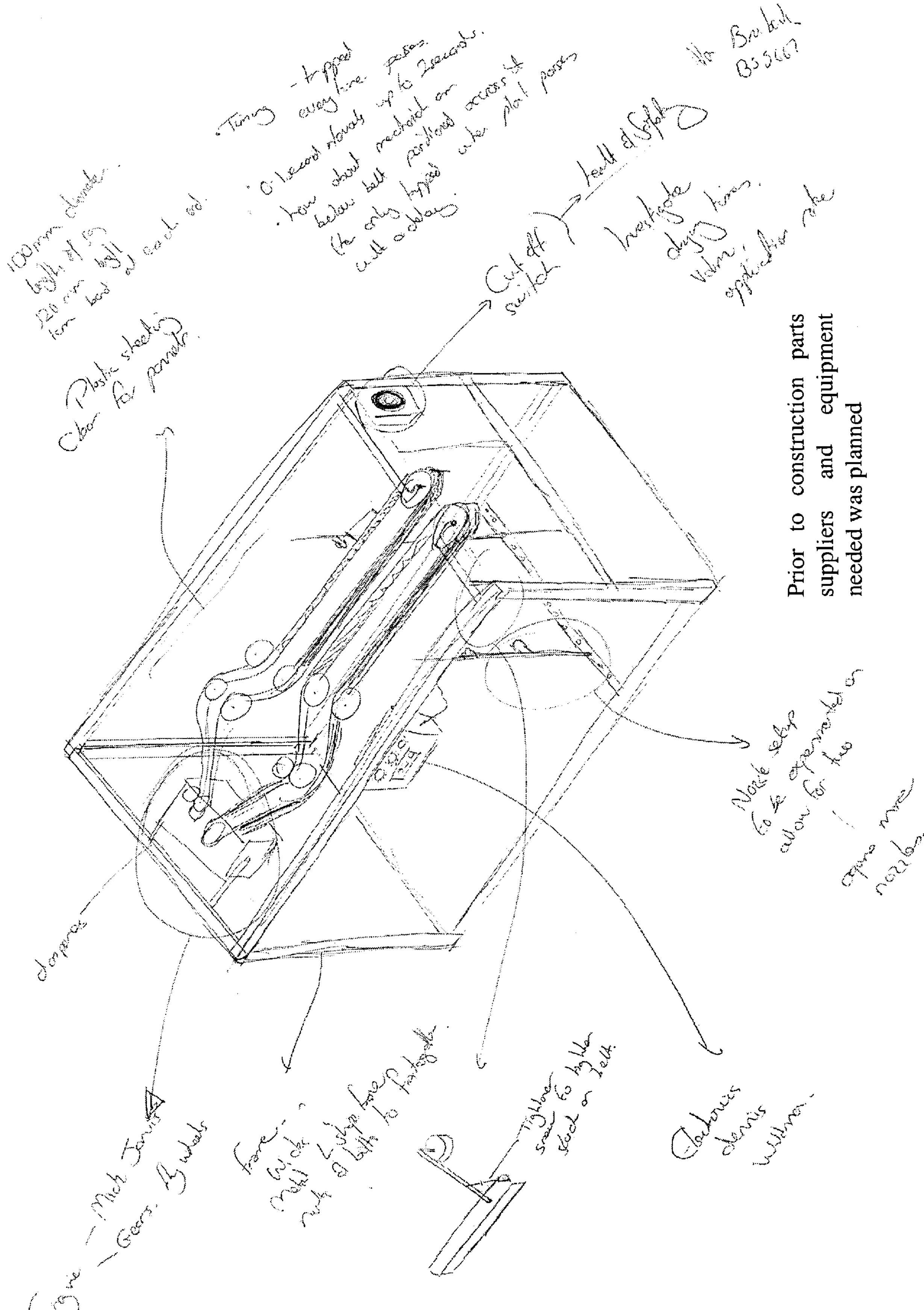




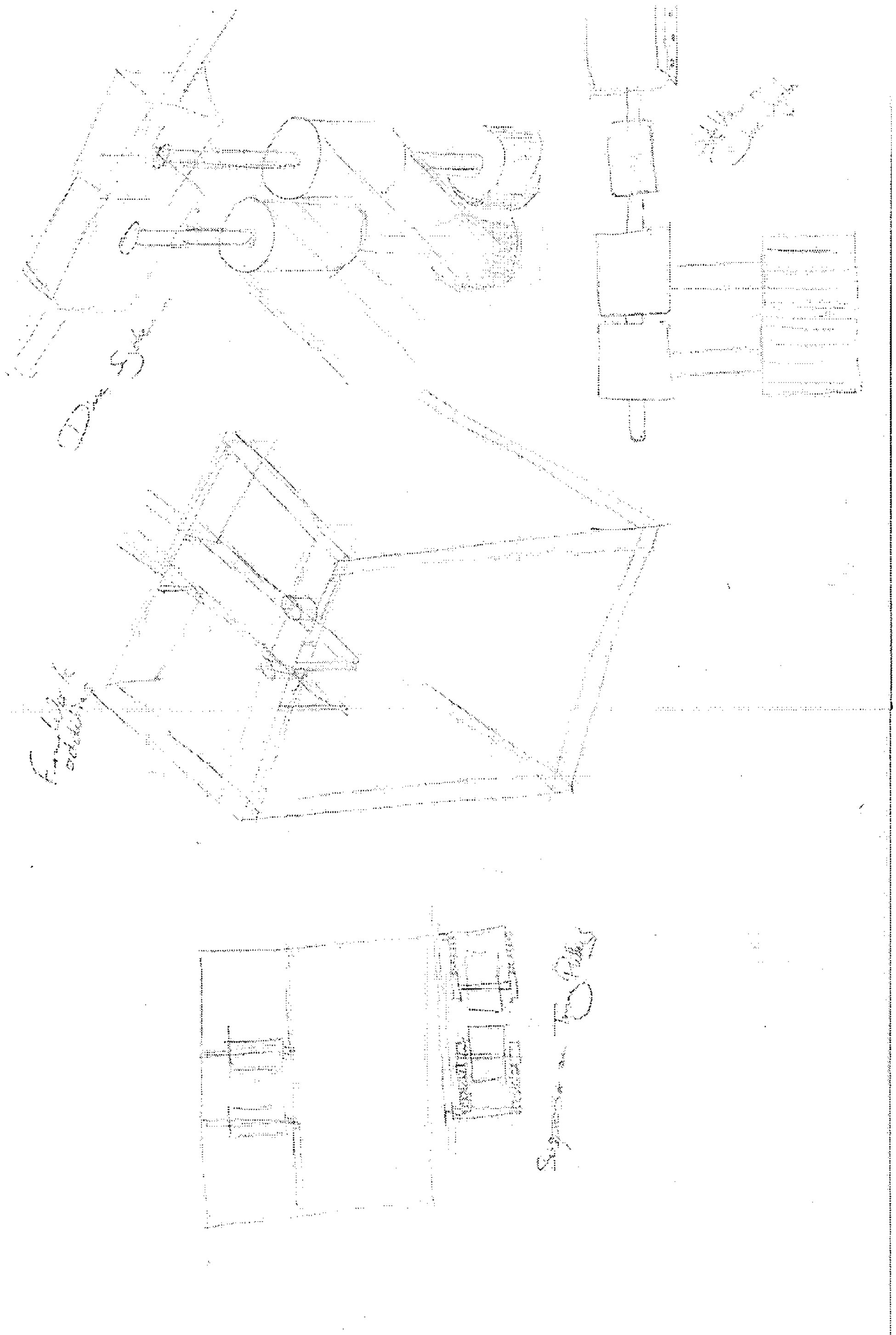
This view includes the panelling that was used to create an interior spray chamber in an attempt to contain airborne spray. The spray chamber panels are indicated by the two arrows.

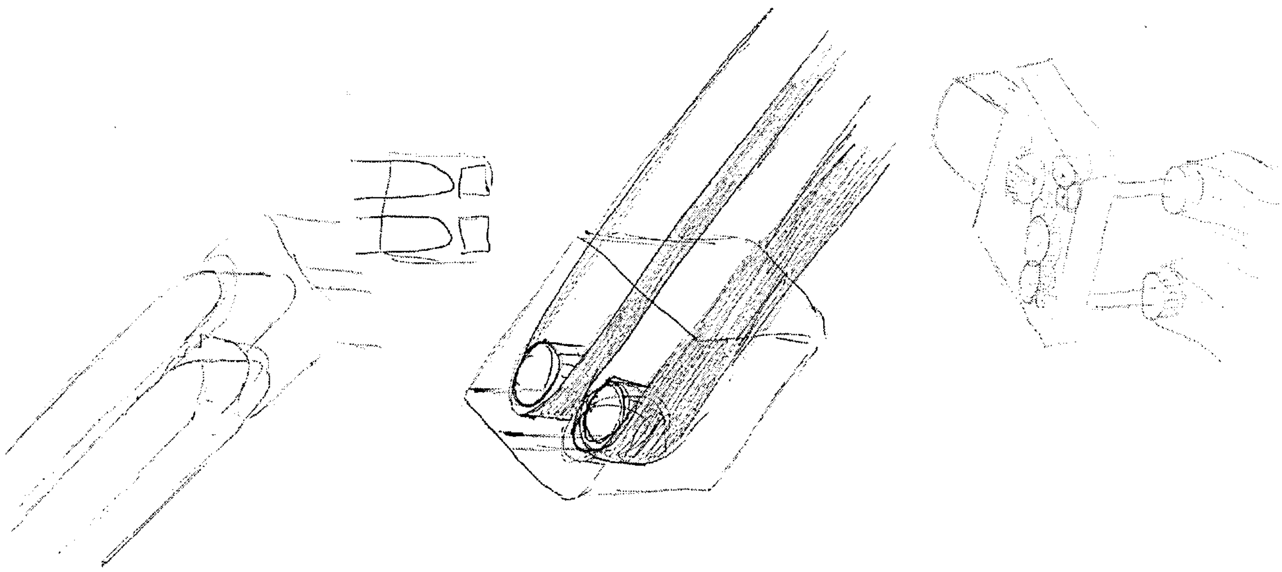


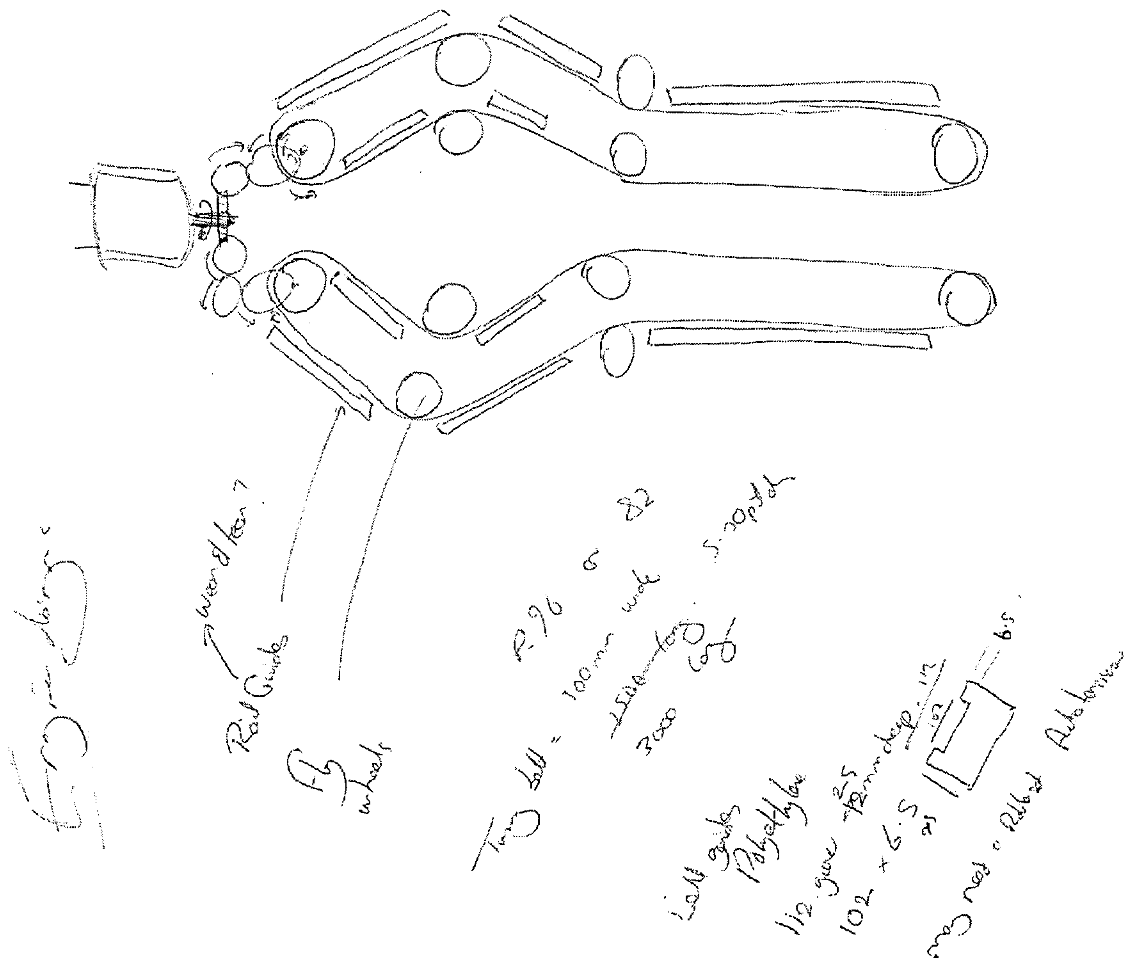
This drawing details the framework and the conveyor belt and pulley positions.

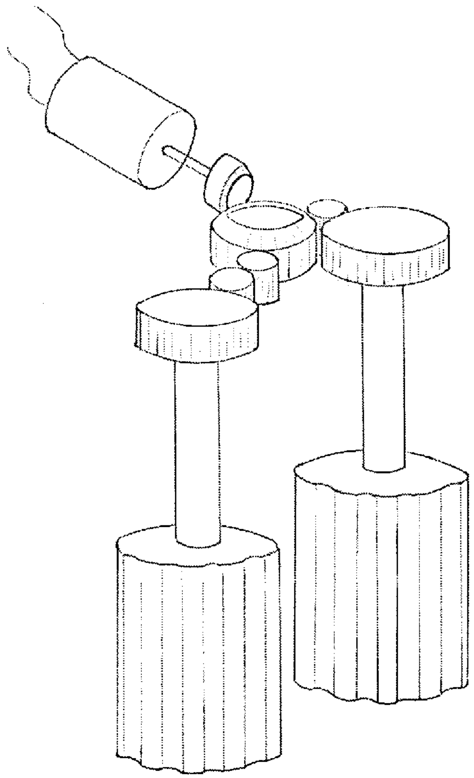


Prior to construction parts suppliers and equipment needed was planned

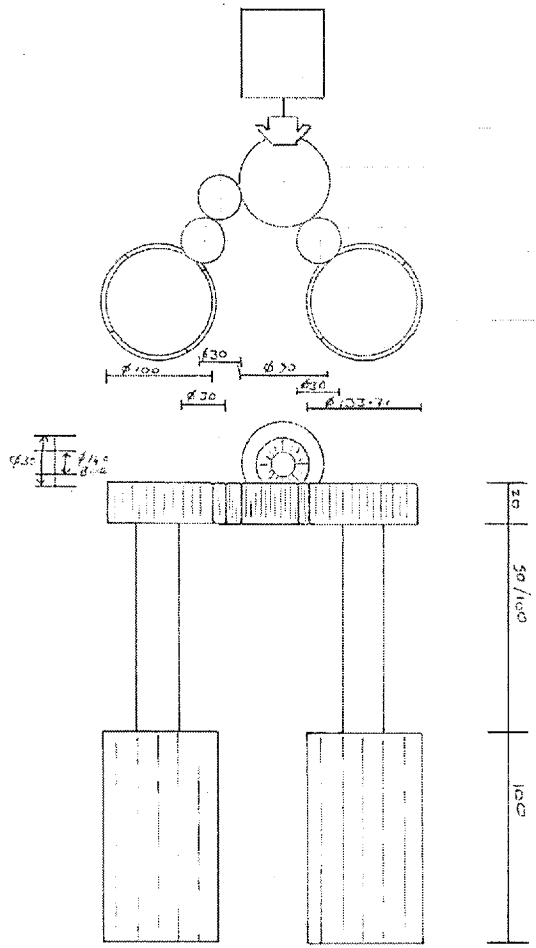








In the early stages of the work, a gearbox was designed which would run the pair of conveyor belts in opposite directions at the required speed. This was later replaced by an off the shelf product.



0·5 hp Engine Approx Rpm = 1500
(not to scale)

Bevel gear ø = 33.79 / ø = 30·00

2nd Bevel gear ø = 31·26 / 30·00 (45 teeth)
dropped sides.

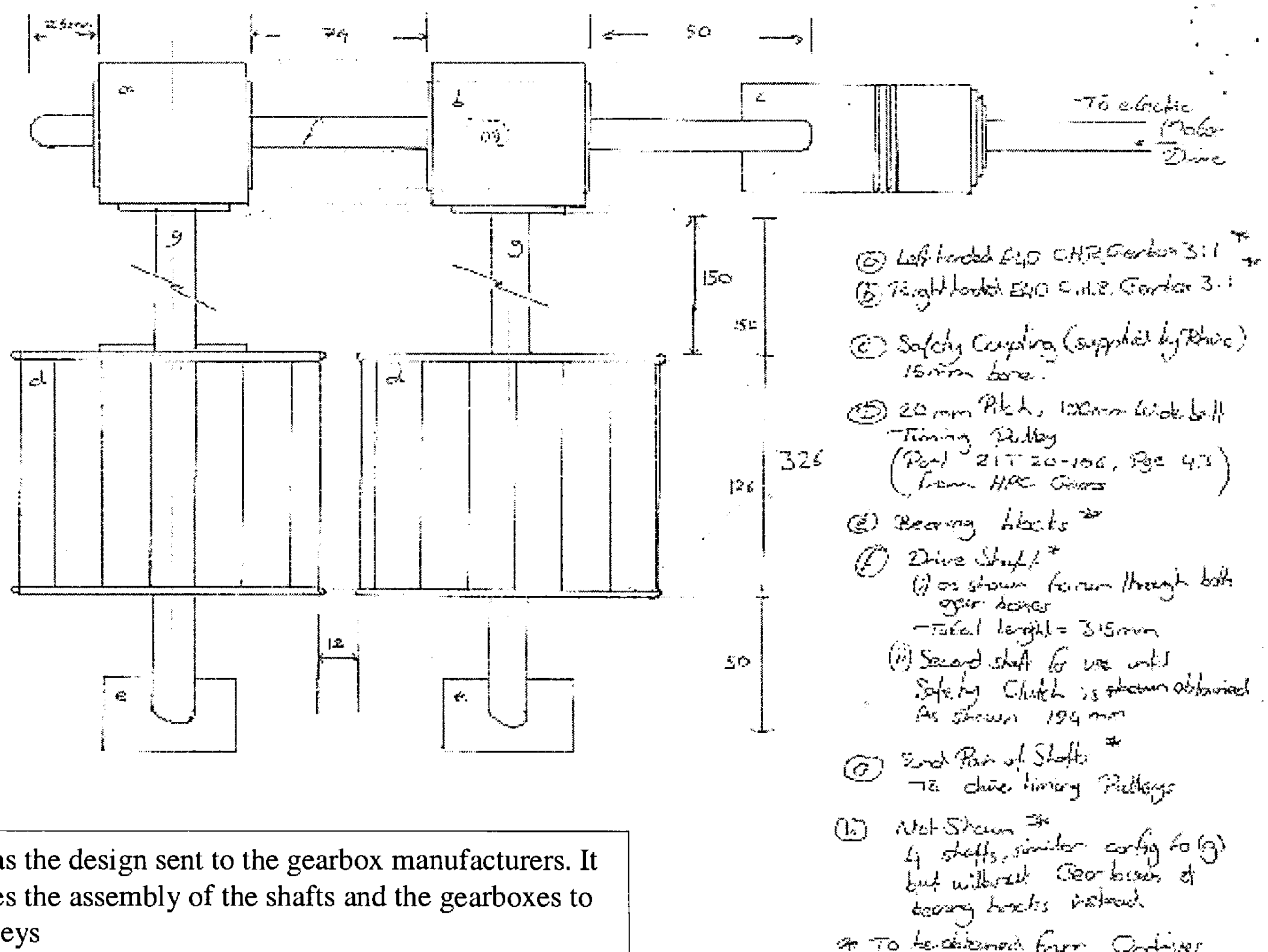
Small gears to create correct
turning direction x3 (15 teeth)
ø 34 mm / ø (PCD) 30 mm

Large final gears Dual ended shaft
Upper end → Spur 20 mod ø 104 ø PCD 99
50 teeth.

Lower end Timing Pulley øPCD 133·71mm
øD 130·85

Belt length approx 2·5ms
Pitch 10/20
Width 20mm

Note
All diameters are PCDs
(distance for use when calculating
how far apart to set gears)
horizontal scale approx 4:1
Vertical Scale approx 2:1
3D view not to scale.



This was the design sent to the gearbox manufacturers. It indicates the assembly of the shafts and the gearboxes to the pulleys

E40 size 15mm bores in/out 2.3Nm-10.0Nm ratios

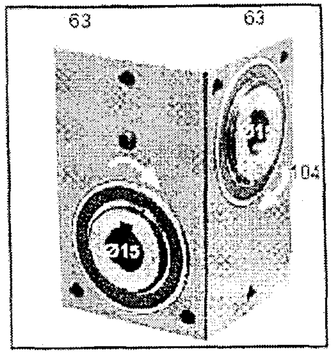
Material
Case:
Alum. HE30TF
Gears:
Alloy Steel 655M13 (EN36) Hardened

Performance
Weight:
1.67kg
Backlash:
1°
Hours Life:
12000 hr
Max Input Speed:
4000 Rpm
Greased for life:
(L2M Hellenic)

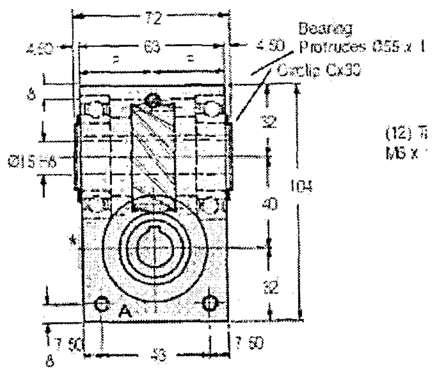
Extras
Output Shafts:
E40-X
E40-DX
Output Key:
KK5-35

R/H Helicals are Standard.
L/H available at **E40-SP** price

E40 Type



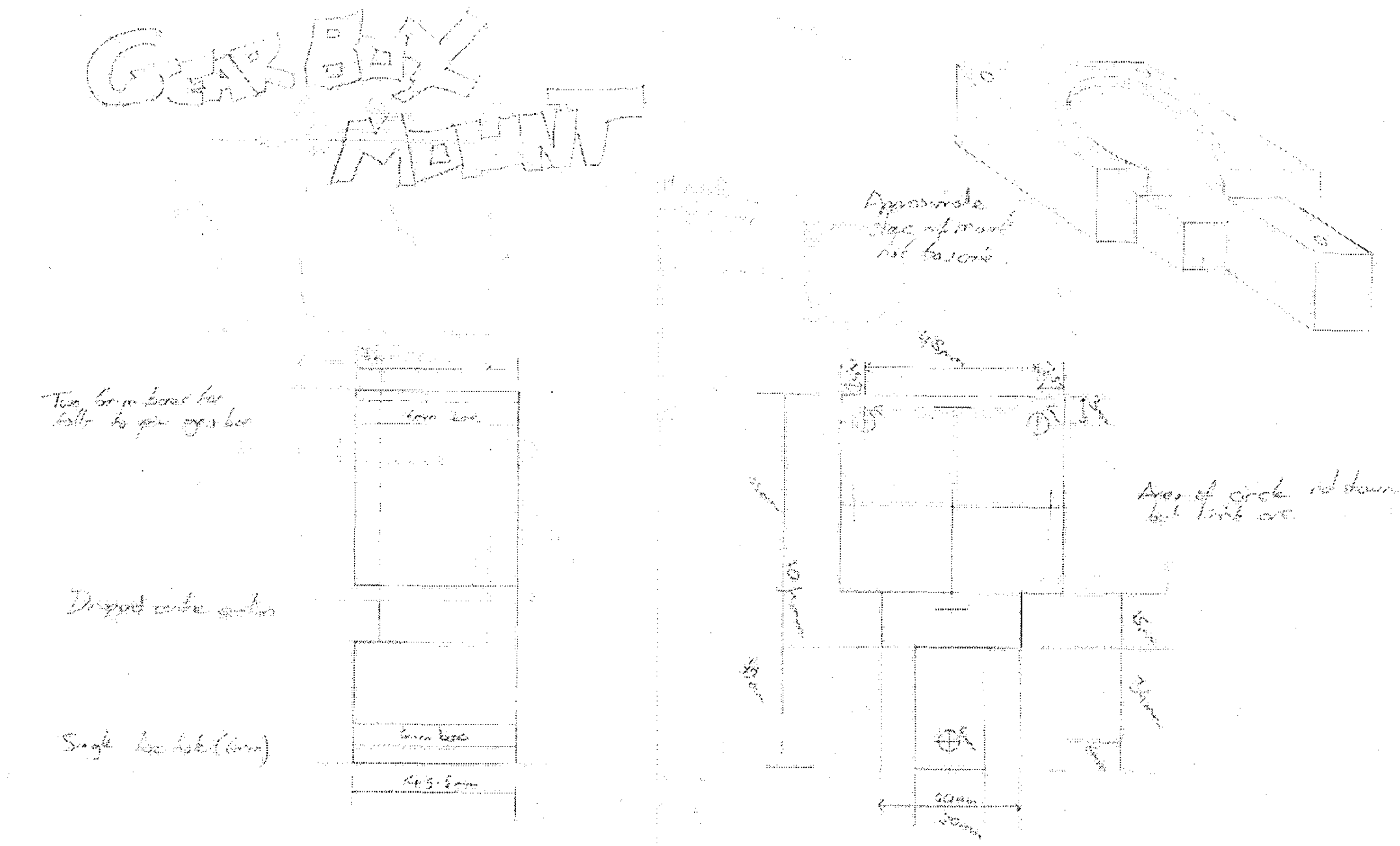
* 1/8" E
fitted o

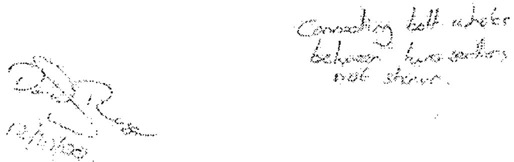


INPUT & OUTPUT FIXINGS ARE BY 5mm KEYW

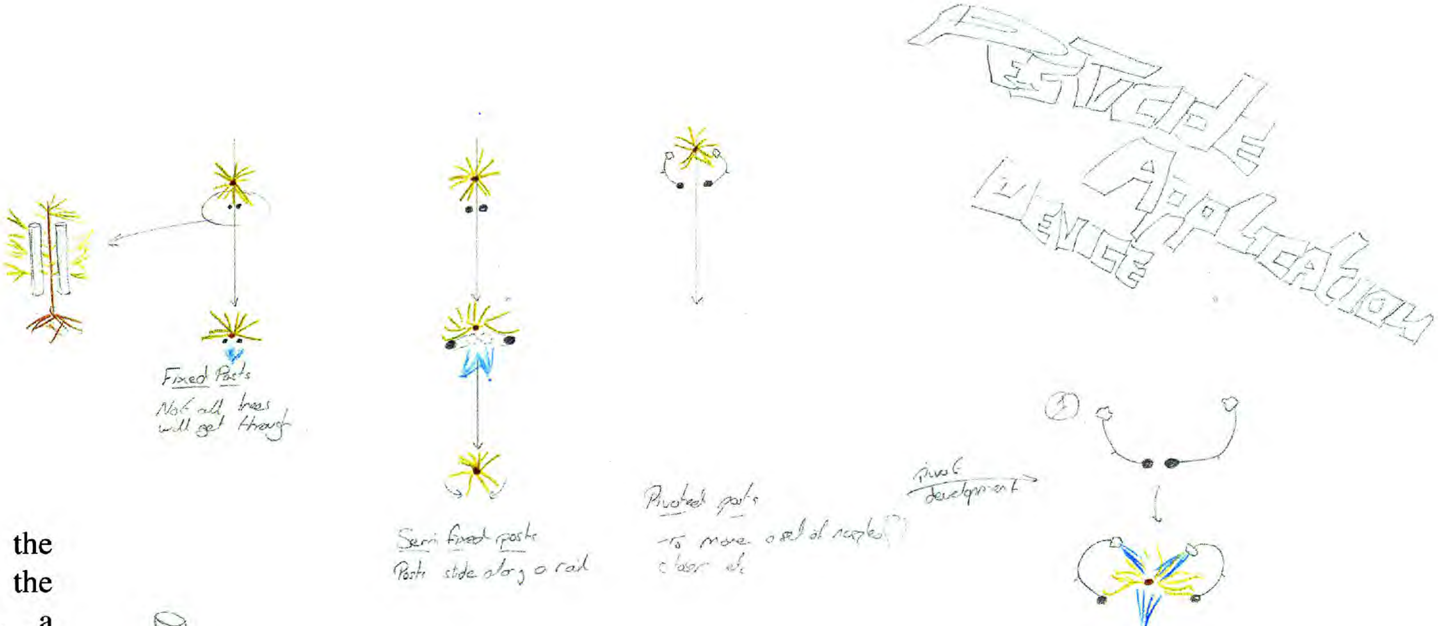
	Rpm Input	Ratio 1:1
N.m. OUTPUT TORQUE	3000	2.30
	2000	2.80
	1000	3.80
	500	5.00
	100	8.00
	50	9.00
	10	10.00

Instead of a home made gearbox, an off the shelf product was purchased. This was a crossed helical axis gearbox. This means that two round discs sat on top of each other have teeth at 45° to each other so that they can interlock. This is a method of converting the drive to a different angle when no other options are possible. Two gearboxes were purchased, a left and right hand model. When joined by a single shaft both gearboxes would join the attached pulleys in different directions (see previous page).

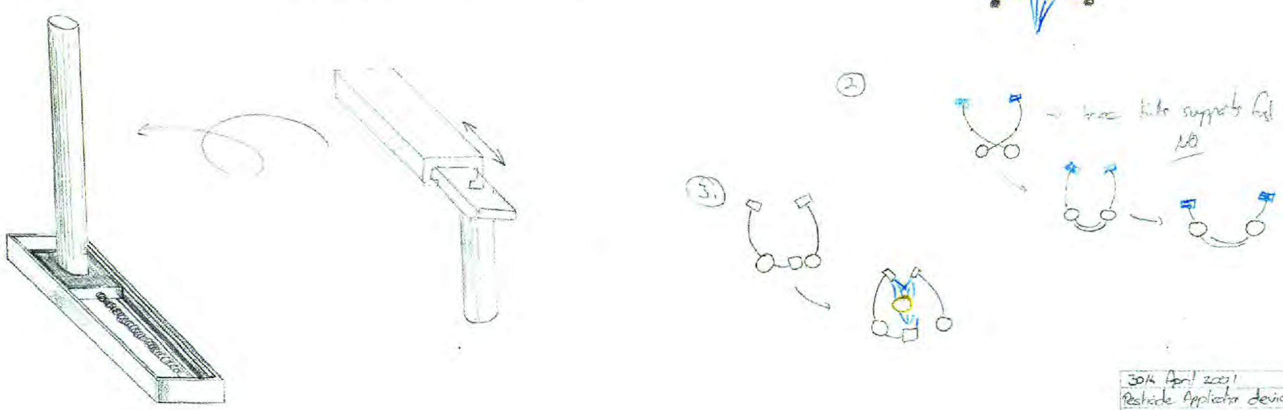




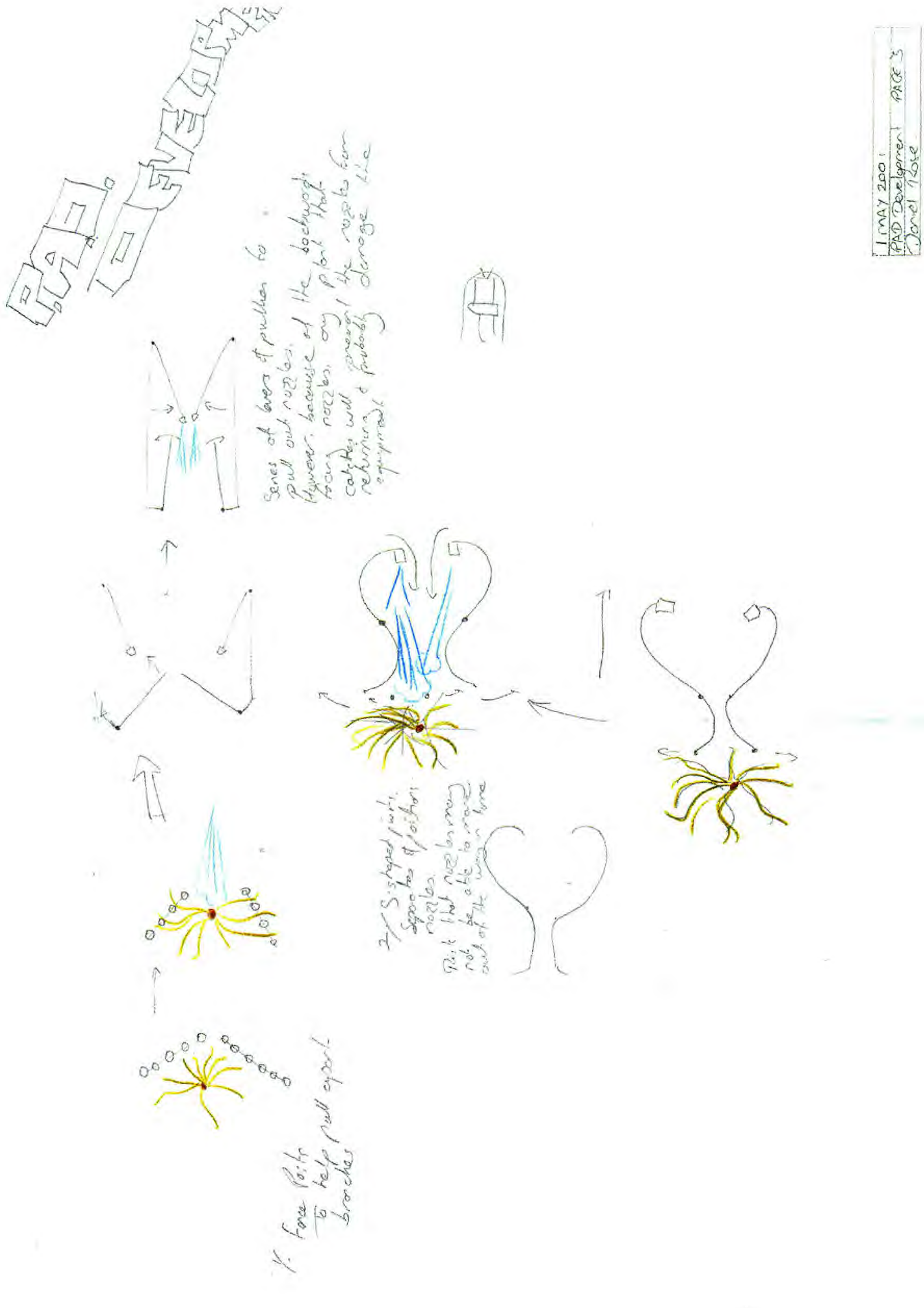
When installing the gearbox and electric motor, it was necessary to very accurately mount the gearboxes so that the shaft opening would directly line up with the motor. This page features the design of the gearbox mounts. It was later decided that a universal coupling should be installed to absorb any misalignment between the gearbox and drive shaft.



To increase the efficacy of the DCS system, a series of designs were generated that could enhance the deposit on the tree. Most of these designs work by opening up the foliage to expose the stem

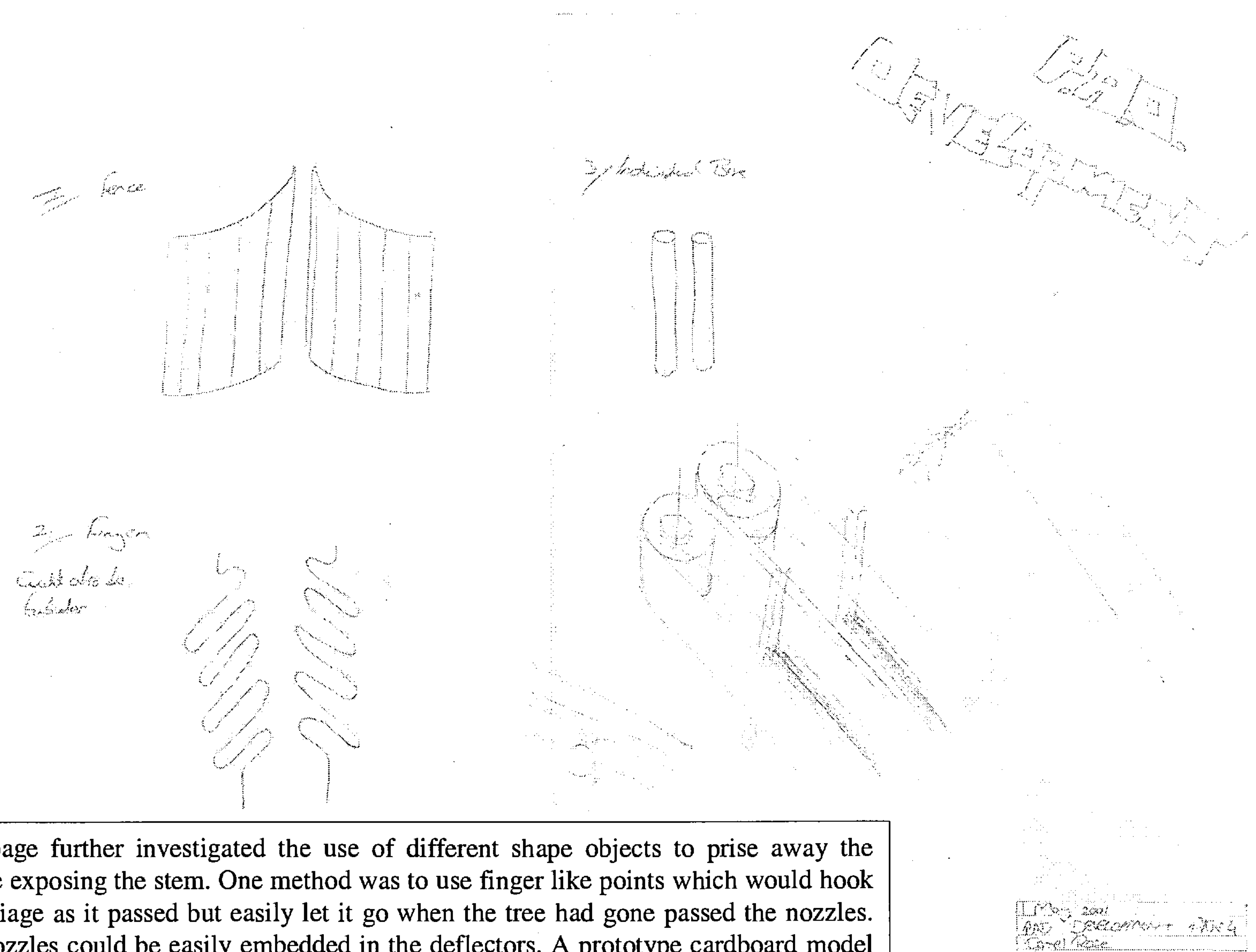


30th April 2001
Pesticide Application device
Daniel Rose



PAD DEVELOPMENT
1 MAY 2001
PAD Development
PAGE 3
Daniel Rose

Daniel Rose
Appendix 1

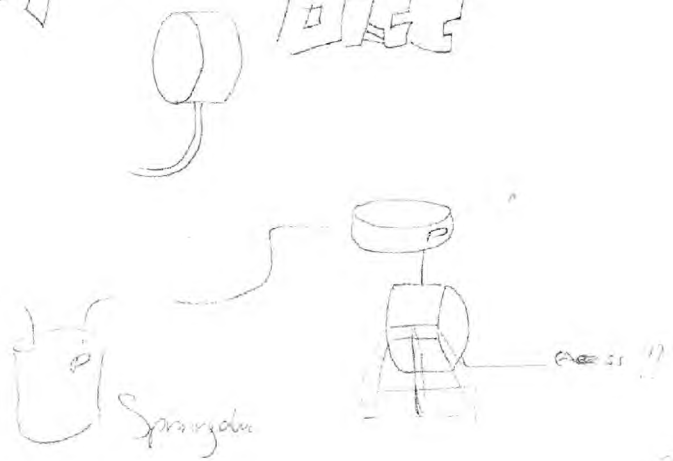


This page further investigated the use of different shape objects to prise away the foliage exposing the stem. One method was to use finger like points which would hook the foliage as it passed but easily let it go when the tree had gone passed the nozzles. The nozzles could be easily embedded in the deflectors. A prototype cardboard model was made of this design. However, it was never included in the final DCS system as it unnecessarily increased the complexity of the system.

Design Portfolio

Pesticide Application Device

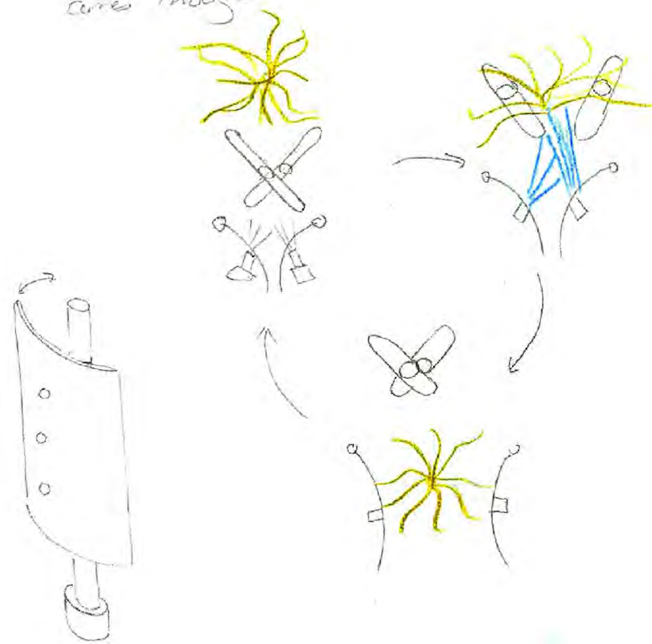
Spinning Disc



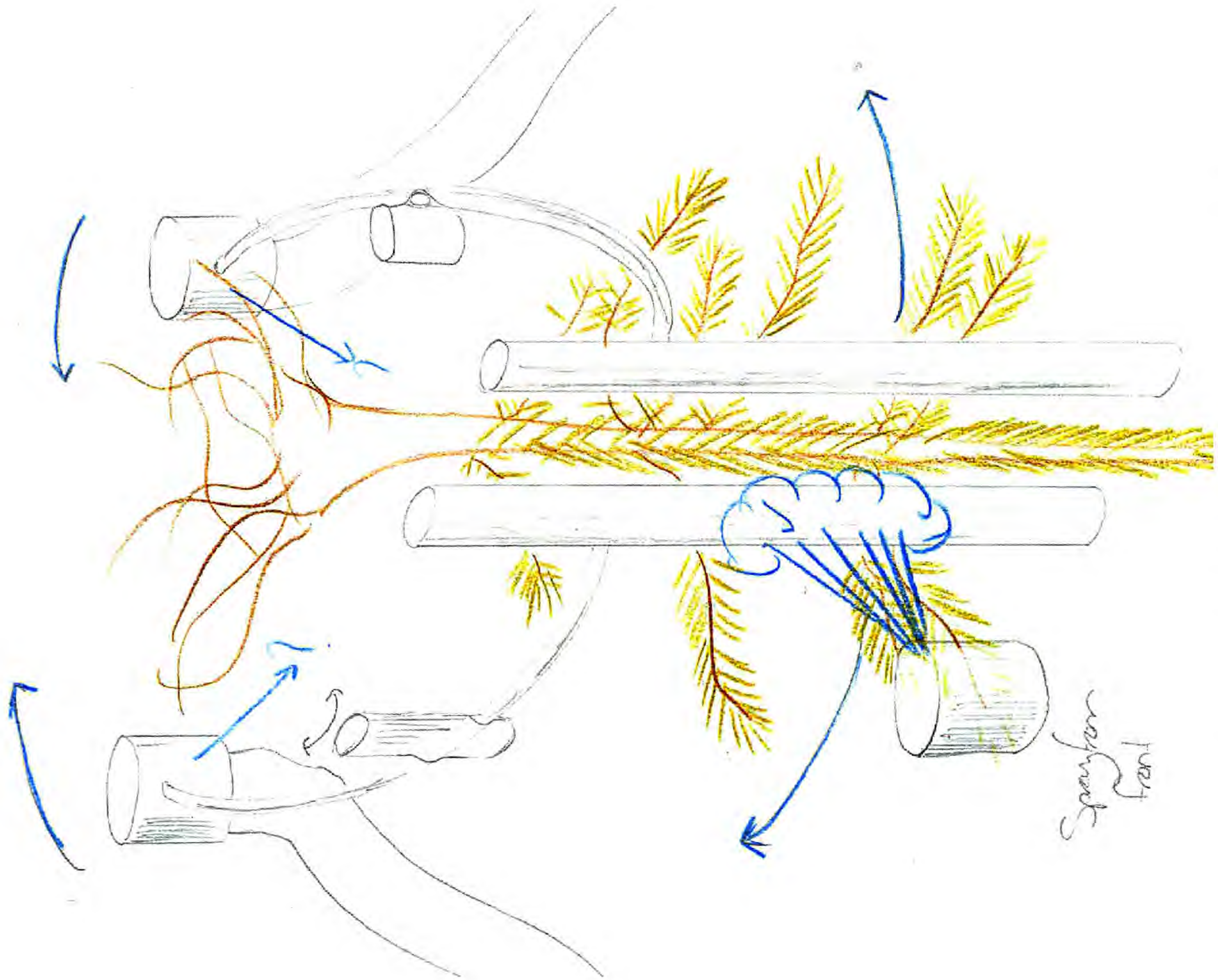
Fingers

- Angle must be acute enough so that they will be pushed in the correct direction
- Must not get stem caught on the wrong side as tree could get stuck
- If they are over lapped then more likely to trap branches
- Nozzles could sit behind fingers & be hinged to be pushed out of the way as tree comes though

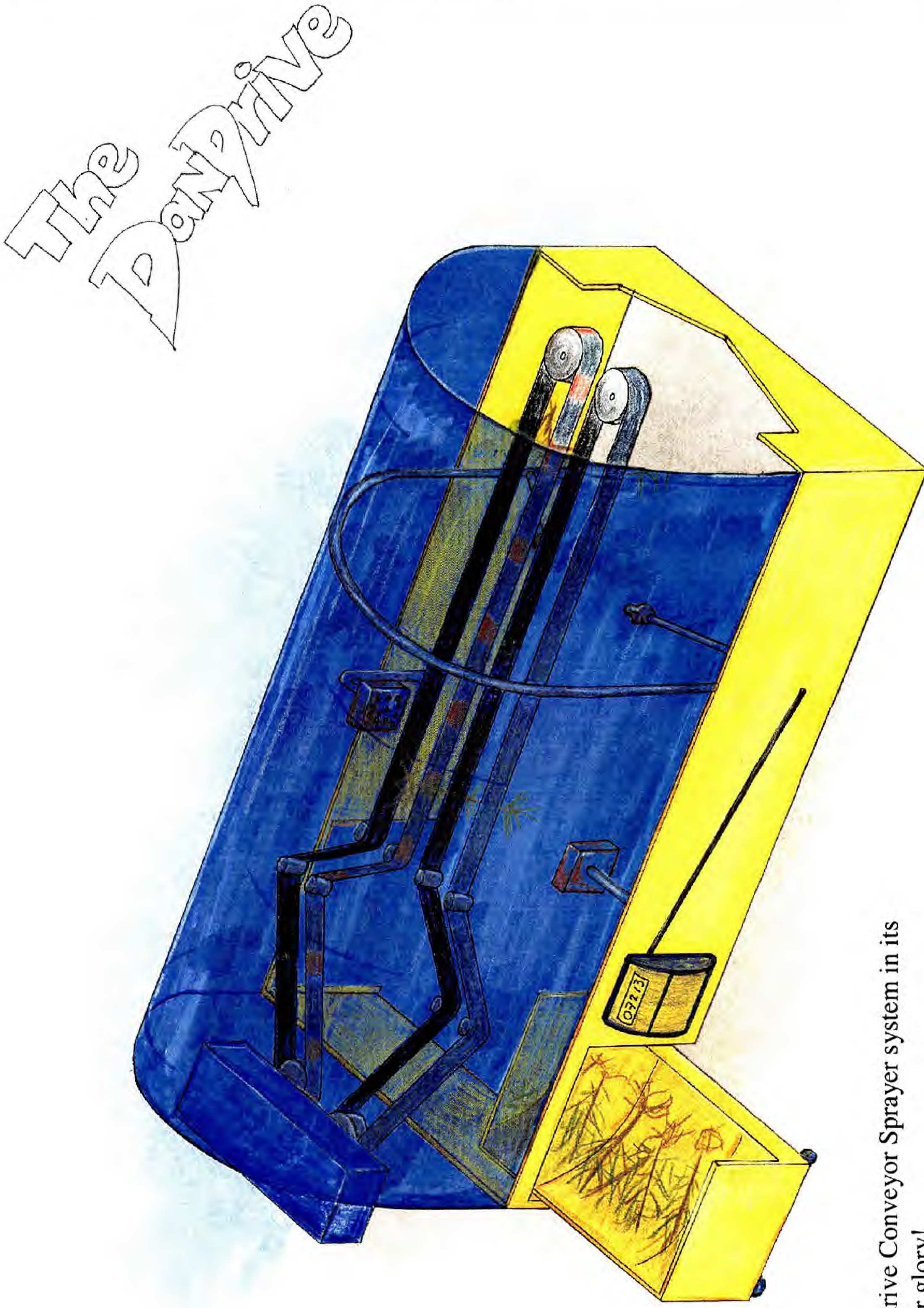
- Should there be any up & down movement ? to allow a natural gap be found ?
- Angled outwards towards bottom to allow for misalignment of main stem.



Further development of the finger deflectors and potential use of a spinning disc nozzle. A spinning disc could be quite effective as it sprays a very low volume. However, it would need to be shielded so that the spray is directed at the tree and not in 360°.



This design involves using the tree to push open a pair of barriers. These barriers are connected to two nozzles by curved arms. When the barriers move back the nozzles pivot around behind the tree and can get very close to the stem to deliver a dose of pesticide. The main advantage is that the nozzles are stored out of the way of the oncoming tree and are not likely to become blocked by pieces of foliage.



The Dandrive Conveyor Sprayer system in its full colour glory!

Appendix 2

Feeding patterns of *Hylobius abietis*

The DCS system aims to spray only a small section of the sapling. The Forestry Commission has stated that only the lower 15cms of the stem of the sapling need be treated. Damage outside this area is less critical and more likely to recover than the lower regions. In addition, the upper parts of the plant are pesticide free and this can lead to safer handling. However the most important reason is that there is evidence that *Hylobius abietis* starts feeding at the base of the tree and moves up, rather than starting at the younger growing tips that other insects may show. Therefore it is at the base where the weevil would first feed on the pesticide. A short experiment was set up to investigate this and is discussed below.

Materials and methods

H. abietis were collected from billet traps in South West Berkshire at Crowthorne Woods (OS 483878,163707). A supply of Norway Spruce (*Picea sitchensis*), was provided by the Forestry Commission and kept in a 5° cold store at Silwood Park, Imperial College, Ascot.

Norway Spruce were planted in small plastic 15cm diameter plant pots. These were watered and left for one day to adjust to the conditions. The following day one *H. abietis* was added to each of the 36 pots and covered with 280mm x 420m clear plastic bag. This was securely fastened around the lip of the pot with masking tape and labelled.

The saplings were checked for damage every day. Damage was recorded on a scale of very small (wound of 1-3mm) to extra large (wound of 15mm +) with a total of four increments. The internode at which the damage occurred was also recorded and the size was estimated and recorded for later reference.

At the end of the experiment (after one week) half of the trees were removed and intensively studied for damage. The size and number of wounds were recorded, as described by (Leather *et al.*, 1994) using a scanner to quantify the area. The remainder of the trees was left in a time to death experiment. Signs of death were noted and the tree pronounced dead when the majority of the needles had fallen or turned brown.

Results and analysis

The results from this clearly showed that the weevil initially targeted the lower areas on the stem, 1st and 2nd internodes. As time passed and damage accumulated on the lower sections, the weevil moved up to higher parts of the plant (Figure 1), often substantially higher parts, 10-15cms from the base. This occurred even when there was still available bark left lower down the stem.

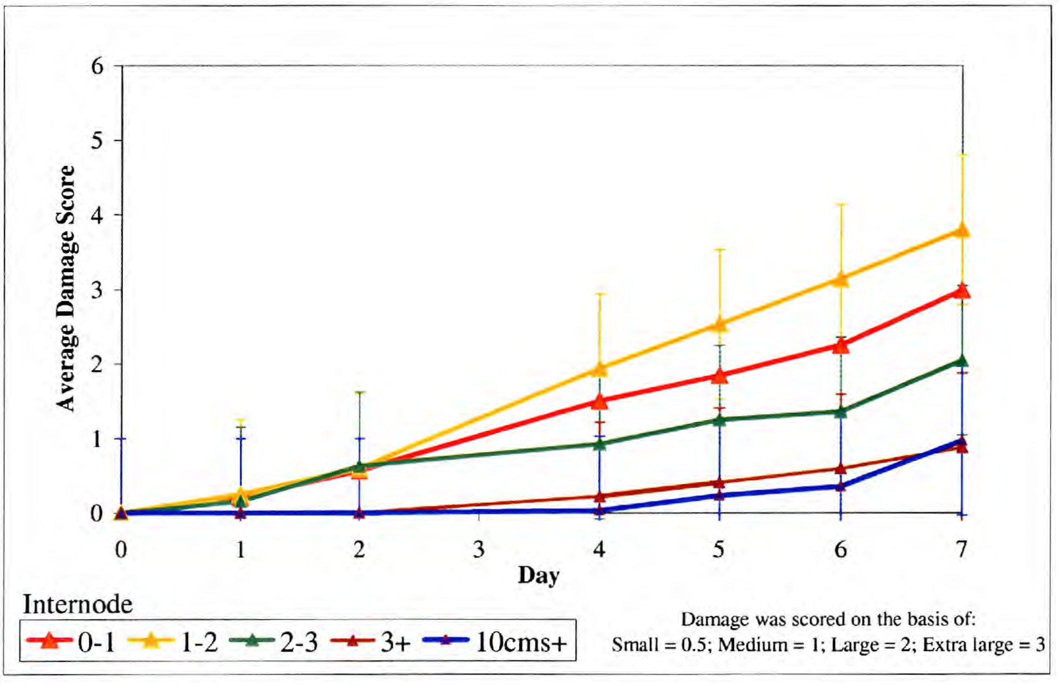


Figure 1 **Accumulation of damage at each internode. There is a clear trend for the weevils to initially attack the lower internodes, whilst damage at higher internodes does not occur until 3 days in to the data collection.**

The second internode (nodes 1-2) was attacked at a higher rate than the lowest internode. This is due to the diversity of internode sizes in the saplings; some saplings had first internodes of several centimetres in length, whilst others were almost non-existent. It would have been better to record damage at measured intervals, such as 0-5cm, 5-10cm, etc.

Discussion

Despite the shortfalls of this experiment (by measuring the internode at which the damage occurred and not the exact height), it is still evident that the damage initially occurred on the lowest parts of the sapling. It is only when *H. abietis* has fed on the lower parts and caused some damage that it moves to higher regions. This work confirms the Forestry Commission observations (Heritage, *Pers. Comm.)* and the DCS system should aim to treat only the 15cm base zone.

There are several ecological reasons that may explain this behaviour. The lower parts of the sapling have a thicker bark and cambium layer than the upper growing tips, therefore offering a better food source. Personal observations have shown that *H. abietis* will feed on cut logs with a diameter 15cm or more, but that this does not appear to occur before the tree is felled. This is most certainly because the sap response is very strong on such a large tree preventing even *H. abietis* from feeding and resin is considered the primary defence of conifers against herbivores (Tomlin & Borden, 1997). However, when the tree is felled there is no phloem transport and bleeding sap is minimal and *H. abietis* is able to feed.

Secondly it may be that there is a defensive significance in this feeding pattern; lower down the stem *H. abietis* is less obvious to aerial observation by birds. In addition *H. abietis* shows a preferences to moving through covered soil than across open ground and moves faster over exposed soils (Kindvall *et al.*, 2000). This highlights the 'awareness' that *H. abietis* may show to predation.

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Leather, S.R., Ahmed, S.I., & Hogan, L. (1994) Adult feeding preferences of the large pine weevil, *Hylobius abietis* (Coleoptera: Curculionidae). *European Journal of Entomology*, **91**;, 385-389.

Tomlin, E.S. & Borden, J.H. (1997) Thin bark and high density of outer resin ducts: Interrelated resistance traits in Sitka spruce against the white pine weevil (Coleoptera: Curculionidae). *Journal of Economic Entomology*, **90**, 235-239.

Appendix 3
DCS Operating Settings

Conveyor belt details					TP4001E or TP6501E		ml deposited in 18cms	
					2.0 bar	2.5 bar	3.0 bar	4.0 bar
Speed	Time for 1 revolution	For 5cm section	For width of tree 18cms	Delay for 30cms	0.32 l/min	0.36 l/min	0.39 l/min	0.45 l/min
12	13.00	0.25	0.90	1.50	4.80	5.40	5.85	6.75
13	12.00	0.23	0.83	1.38	4.43	4.98	5.40	6.23
14	11.14	0.21	0.77	1.29	4.11	4.63	5.01	5.79
15	10.40	0.20	0.72	1.20	3.84	4.32	4.68	5.40
16	9.75	0.19	0.68	1.13	3.60	4.05	4.39	5.06
17	9.18	0.18	0.64	1.06	3.39	3.81	4.13	4.76
18	8.67	0.17	0.60	1.00	3.20	3.60	3.90	4.50
19	8.21	0.16	0.57	0.95	3.03	3.41	3.69	4.26
20	7.80	0.15	0.54	0.90	2.88	3.24	3.51	4.05
21	7.43	0.14	0.51	0.86	2.74	3.09	3.34	3.86
22	7.09	0.14	0.49	0.82	2.62	2.95	3.19	3.68
23	6.78	0.13	0.47	0.78	2.50	2.82	3.05	3.52
24	6.50	0.13	0.45	0.75	2.40	2.70	2.93	3.38
25	6.24	0.12	0.43	0.72	2.30	2.59	2.81	3.24

Shaded cells indicate
desired operating range

NB If Spray duration is increased, then the delay should be decreased by half that amount				
Spray angle data	**10**	85.73	**65**	4.77
Distance of nozzles	**20**	42.53	**70**	5.25
from target at each	**30**	27.99	**75**	9.77
angle	**40**	20.61	**80**	8.94
	45	18.11	**85**	8.18
	50	16.08	**90**	7.50
	55	14.41	**95**	6.87
	60	12.99	**100**	6.29

Appendix 4a
Operating Protocol for use of the DCS system

Introduction

The DCS system (Dandrive Conveyor Sprayer), the conveyor belt system situated in IPARC, Silwood Park, was designed to spray pesticides on to young conifer saplings such as Spruce and Pine. The system aims to spray a very low dose of concentrated pesticide on to a target area on the sapling. The saplings then leave the conveyor belt damp but not dripping wet and should therefore be safe to handle with a minimum level of protective clothing.

However, the current system is still at the prototype stage and due to lack of funds does not include many of the full safety features. These features consist primarily of a large air extraction system to extract spray drift/vapour; acrylic or metal panels, which will not absorb pesticides and prevent spray drift; and also electronic safety cut off switches. Therefore this current version of the system should be used with increased caution and care compared to the intended production model. The list of guidelines below highlights areas of risk and state suitable measures to be taken.

It must be noted that there are no large pesticide trials planned for the current prototype system.

Operating Guidelines

General Use

1. All users should first be familiarised with the use of the DCS system and trained in its use before they will be permitted to use it unsupervised.

2. When using the DCS system without supervision it is advisable that a member of IPARC is aware of your activities in case of an accident. No body should use pesticides in the DCS system without supervision.

3. The DCS system has several large mechanical parts possibly capable of causing injury. The two belts have the potential to trap an arm or loose item of clothing and pull it in to the machine. Therefore no loose clothing should be worn around the machine, especially neckties. When used at the correct operating speeds (not exceeding 25 m/minute, there is not enough torque in the belts to cause serious injury.

4. In the event that an arm is caught in the machine, the user should pull it down and backward avoiding contact with the pulleys. Any cuts or abrasions should be thoroughly cleaned and a physician seen if necessary.

5. The most likely cause of injury arises when lazy workman ship occurs, such as failing to switch off the conveyor belt when altering nozzle settings. Therefore always switch off the conveyor if altering or changing the nozzle set-up. Additionally the spray system should be switched off when retrieving trees that have fallen out of the conveyor belts into the drainage system. This will prevent triggering of the infrared detector and therefore prevent accidental contamination by being sprayed in the face. This is especially essential if pesticides are being used.

6. All of the electrical systems on the DCS system operate from a 12volt DC supply, but converted from AC mains. Therefore the risk of electrical shock is minimised,

however, when altering any of the components the system should be switched off at the mains.

Pesticide Use

1. Prior to any pesticide application, all procedures and pesticides should be checked by Professor Graham Matthews.

2. Due to the lack of adequate containment of spray vapour during operation, users should treat the machine as if they were spraying indoors with a knapsack sprayer. Therefore full PPP clothing should be worn including polypropylene ovals, apron, gloves and wellington boots, plastic face shield and a charcoal respirator.

3. Other members of IPARC should be informed of the spraying at least a week before the event if prolonged use is intended. For short sessions of less than 10 minutes this will not be required. In the event of a large trial lasting several days, other members should be informed of any side effects that they may notice from the pesticide in use.

4. If any member of either the spray team or users of IPARC report any side effects, the pesticide application should stop immediately.

5. Only PSD authorised pesticides may be used in the DCS system.

Appendix 4b

FORESTRY COMMISSION RESEARCH AGENCY

Evaluation of plant protection products

Entomology

EXPERIMENT PLAN

Experiment Number:	PPP01008
Experiment Title:	The phytotoxicity of lambda-cyhalothrin and alpha-cypermethrin both applied to Sitka spruce transplants using the IPARC spray system.
Key words:	Lambda-cyhalothrin Hallmark Contest alpha-cypermethrin permethrin dip IPARC Spray phytotoxicity
Local Ref. No:	
Background:	For the past 40 years treating plants with insecticide before planting has been the most reliable method of protecting bare-rooted plants from damage by Hylobius abietis. Following approval for the Electrodyn system, PSD announced in April 2000 that dipping plants in permethrin would not be allowed after June 2000. In addition, the use of permethrin in forestry will no longer be allowed in any EC country after the year 2003.

The International Pesticide Application Research Centre (IPARC) was commissioned to develop a system to treat bare-rooted trees with aqueous formulations of insecticide. An application system has been developed that sprays the plants using controlled, ultra low volume techniques. However, the concentration of the insecticide has to be increased to compensate for the low volumes applied. Whilst dipping SS transplants in permethrin has no adverse affect on their survival or growth, applying higher concentrations may reduce plant vigour. This phytotoxic effect may not be evident after planting in ideal conditions, but may reduce the ability of plants to survive on an arduous planting site.

Field trials have shown that the synthetic pyrethroids, Contest and Hallmark applied by dipping provide the same level of protection as the standard permethrin dip. These trials will include these insecticides, but at the higher concentrations suitable for ultra low volume applications.

Objectives: To compare the growth and survival of transplants of Sitka spruce after treatment using the IPARC spray system. Alpha-cypermethrin (Contest) and lambda-cyhalothrin (Hallmark) will be compared with permethrin applied by dipping (Permasect 25EC) or the Electrodyn (Permethrin 12ED). The test products will be applied in a range of doses to meet the requirements for approval of plant protection products and will be the same as used in efficacy experiments.

Locations: A new upland planting site (or a dry agricultural site) will be selected so that insect damage is unlikely but where the plants will be growing under stress.

Species: Sitka spruce (*Picea sitchensis* (Bong))

Products and active ingredients:

Product	Concentration	Active ingredient	Manufacturer
Permethrin 12ED	12% ai ec	permethrin	Techneat Chemicals Ltd
Permasect 25EC	25% ai ec	permethrin	Mitchell Cotts
Contest	15% w/w WG[1]	alpha-cypermethrin	Aventis
Hallmark	10% w/w MC[2]	lambda-cyhalothrin	Syngenta

[1]WG is a formulation of water dispersible granules
[2]MC is an aqueous suspension of microcapsules containing the active ingredient

Experimental Treatments:

Code		Treatment	Active ingredient	Application rate (% a.i.)	Product rate (g/100 trees)
A	IPARC	Hallmark	lambda-cyhalothrin	0.4	32
B	IPARC	Hallmark	lambda-cyhalothrin	0.8	64
C	IPARC	Hallmark	lambda-cyhalothrin	1.6	128
D	IPARC	Contest	alpha-cypermethrin	0.375	20
E	IPARC	Contest	alpha-cypermethrin	0.75	40
F	IPARC	Contest	alpha-cypermethrin	1.5	80
G	IPARC	Control	Water	0	
H	Electrodyn	Permethrin 12ED	permethrin	12	*
I	Dip	Permasect 25EC	permethrin	0.8	35 (g/litre)
J	Dip	Untreated control	Water	0	Water Dip

* The actual usage rates will be checked during the course of the trial.

Design:	At each site there will be 5 replicates and the 10 treatment plots will be randomised within each replicate. Each plot will contain 20 plants planted as a single row at a minimum of 0.5m spacing. Each row should be labelled with a marker cane. This trial will be planted adjacent to the PPP01002 trial so that broad comparisons may be made between the IPARC and Electrodyn treatment systems.
Methods:	If cold stored plants are used for the trial they must be allowed to reach a temperature of at least 5°C before treatment. All insecticide solutions will be made up on the day of use. After treatment, dipped plants will be air-dried under cover and the tops must be completely dry before they are bagged for storage. Electrodyn and IPARC treated plants should be bagged as they leave the treatment system. The sprayed plants will be sealed into new co-extruded polythene bags within 15 minutes of treatment and stored below at between 2°C and 10°C before dispatch. Planting will be undertaken within 10 days of treatment and will be completed before the 30th April. No plant protection products (such as herbicides) should be applied to the site after planting.
Requirements and Responsibilities:	Sites – 2 @ 0.04ha Plants = 10 treats x 5reps x 20 plants x 2 sites = 2000 SS Marker canes at row ends Plot marker posts ($3cm^2$)
Records and Assessments:	Plant growth and survival will be assessed between November 2001 and March 2002 following the method described in SOP057.
Statistical analysis:	Plant survival and increments will be compared using ANOVA following angular transformation of the survival data.
Duration:	The trial sites will be retained until after the autumn assessments in 2002.
COSHH & Risk assessments:	COSHH assessments for plant treatment and for planting of permethrin, alpha-cypermethrin and lambda-cyhalothrin treated plants will apply to these experiments.

SOPs: The following SOPs are relevant to this experiment:

SOP No.	Title of process
SOP004	Records keeping for field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP020	The preparation and mixing of plant protection products.
SOP048	Marking and tallying field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP056	Using a balance to determine product weight
SOP057	Assessing plant growth as an indication of the phytotoxic effects of insecticides on young forest plants.
SOP062	The manual application of plant protection products by dipping.
SOP066	The use of a cold-store to store plant material

Drafted by: Stuart Heritage
Date: 14th March 2001

Design approved: A Milner
Date: 30th March 2001

Approved: Dr H Evans
Date:

Approved: Mr J Dewar (CEO)
Date:

Appendix 4c

FORESTRY COMMISSION RESEARCH AGENCY
Evaluation of plant protection products

Entomology

EXPERIMENT PLAN

Experiment Number:	PPP01007
Experiment Title:	The efficacy of water based formulations of lambda-cyhalothrin and alpha-cypermethrin applied using the IPARC spray system to protect forest plants from damage by *Hylobius abietis* (Large Pine Weevil) and *Hylastes* spp. (Black Pine Beetles).
Key words:	Lambda-cyhalothrin Hallmark alpha-cypermethrin Damage Permethrin IPARC Electrodyn Spray
Local Ref. No:	
Background:	Unless transplants are protected from attack by *Hylobius abietis*, plant losses throughout UK restocking areas will amount to an average of about 50% over the two years after planting during which they remain susceptible to damage. Since 1983, treating plants by dipping has been the recommended method of protecting plants during the first growing season. PSD announced in April 2000 that dipping plants in permethrin would not be allowed after June 2000. The International Pesticide Application Research Centre (IPARC) was commissioned to develop a system to treat bare-rooted trees with aqueous formulations of insecticide. It was considered essential that adequately treated plants must be delivered by the system dry and ready for storage or dispatch. The IPARC system should offer a number of advantages over dipping such as isolating operators from the point of application and treating plants in a controlled and consistent way. The use of permethrin in forestry will no longer be allowed in any EC country after the year 2003. Because the IPARC sprayer uses water-based products, it will be possible to use "off the shelf" formulations of insecticides leading to lower registration costs. Trials carried out in 1999 and 2000 have shown that the synthetic pyrethroids, Contest and Hallmark applied by dipping provide the same level of protection as the standard permethrin dip. These trials will compare the 2 insecticides that have proved effective when applied by dipping but at higher concentrations suitable for ultra low volume applications.
Objectives:	To compare the level of protection from damage by *Hylobius abietis* and *Hylastes* spp. provided by alpha-cypermethrin (Contest) and lambda-cyhalothrin (Hallmark) applied using the IPARC spray system with that from permethrin applied by dipping or the Electrodyn. All work will be conducted in forest restocking sites less than 3 years after felling (SOP104) and include three application rates to meet the requirements of PSD.
Locations:	15 sites will be chosen, to cover a range of restocking conditions throughout the UK and where *Hylobius* populations might be expected to be high. The trials will be planted adjacent to the PPP01001 trials so that broad comparisons may be made between the IPARC and Electrodyn treatment systems.
Species:	Sitka spruce (*Picea sitchensis* (Bong)) will be used as treatment plants on all sites. Buffer rows of untreated Scots pine will be planted between the experiment plots

on sites where plot boundaries may be difficult to identify during assessments.

Products and active ingredients:

Product	Concentration	Active ingredient	Manufacturer
Permethrin 12ED	12% ai ec	permethrin	Techneat Chemicals Ltd
Permasect 25EC	25% ai ec	permethrin	Mitchell Cotts
Contest	15% w/w WG[1]	alpha-cypermethrin	Aventis
Hallmark	10% w/w MC[2]	lambda-cyhalothrin	Syngenta

[1]WG is a formulation of water dispersible granules
[2]MC is an aqueous suspension of microcapsules containing the active ingredient

Experimental Treatments:

Code		Treatment	Active ingredient	Application rate (% a.i.)	Product rate (g/100 trees)
A	IPARC	Hallmark	lambda-cyhalothrin	0.4	32
B	IPARC	Hallmark	lambda-cyhalothrin	0.8	64
C	IPARC	Hallmark	lambda-cyhalothrin	1.6	128
G	Electrodyn	Permethrin 12ED	permethrin	12	*
H	IPARC	Control	Water	0	*
I	Dip	Permasect 25EC	permethrin	0.8	35 (g/litre)
J	Dip	Untreated control	Water	0	Water Dip

*The actual usage rates will be determined during the course of the trial.

Design: At each site there will be only 2 replicates to enable a large number of sites to be included in the trial. These trials will be planted adjacent to the PPP01001 trials so that broad comparisons may be made between the IPARC and Electrodyn treatment systems.

The 7 treatment plots will be randomised within each replicate. Each plot will contain 20 plants planted at 2m spacing as a 5x4 block. This has been reduced from the 25 plants per plot recommended in the EPPO Guideline[3] to allow a larger number of sites to be included in the trial. A marker cane should mark each row of plants.

Methods: All insecticide solutions will be made up immediately before use. To avoid treated plants being stored for too long; treatments will be applied only after the experimental plots have been laid out in the forest. All plants will be planted within 10 days of treatment (target 3 days) and planting of the experiment will be completed before the 31st March. No insecticides or herbicides should be applied to the plants as a basal treatment.

No post-planting applications of insecticides or herbicides will be made to any trial plants during the 2-year test period.

Requirements and Responsibilities:

Sites – 15 @ 0.15ha

Plants – 5.25k SS

Marker canes at row ends

Plot marker posts (3cm^2)

Records and Assessments: All sites will be checked in November 2001. If more than 10% of the untreated plants have been severely damaged, all plants will be assessed for damage using the scoring system described in SOP053.

For each site a note should be made on the Operations record of whether there is evidence of phytotoxicity apparent at the time of assessment and also if there was damage by *Hylastes*. If more than 10% of plants are affected by either cause, this should be assessed using the appropriate scoring system.

SOPs: The following SOPs are relevant to this experiment:

Statistical analysis: Only two replicates of each treatment will be set up at each site to allow a wide range of site conditions to be included in the trial. The small number of replicates may mean that the analysis of results from individual sites is unreliable. The experiment will therefore be analysed by amalgamating the results from all the sites where damage to untreated control plants is above 10%. Results from a minimum of 5 sites will be combined and treatments compared using ANOVA (following angular transformation for the survival data).

Duration: All experiment sites will be retained for the first year. If there is significant protection from treatments at the end of autumn 2001, the sites should be retained for further summer and autumn assessments in 2002.

COSHH & Risk assessments: COSHH assessments for plant treatment and for planting Electrodyn formulations of permethrin and water based formulations of permethrin, lambda-cyhalothrin, and alpha-cypermethrin treated plants will apply to these experiments.

SOP No.	**Title of process**
SOP004	Records keeping for field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP006	Selecting trial sites for experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP020	The preparation and mixing of plant protection products.
SOP048	Marking and tallying field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP053	The assessment of damage caused by *Hylobius* feeding.
SOP056	Using a balance to determine product weight
SOP062	The manual application of plant protection products by dipping.
SOP066	The use of a cold-store to store plant material
SOP104	Selecting sites suitable for trials of insecticides to protect plants from damage by *Hylobius*

Bibliography: [3] **EPPO Bulletin 18** Guideline for the evaluation of insecticides, pages 759-766 (1988)

Drafted by: Stuart Heritage
Date: 15th February 2001

Design approved: Alvin Milner
Date: 30th March 2001

Approved: Dr H Evans
Date: 16th February 2001

Approved: Mr J Dewar (CEO)
Date:

Appendix 4d

FORESTRY COMMISSION RESEARCH AGENCY

Evaluation of plant protection products

Entomology

EXPERIMENT PLAN

Experiment Number:	PPP01010
Experiment Title:	The effect of cold storage on the phytotoxicity of lambda-cyhalothrin and alpha-cypermethrin both applied using the IPARC spray system to Sitka spruce transplants.
Key words:	Lambda-cyhalothrin Hallmark Contest Alpha-cypermethrin Permethrin Dip IPARC Spray phytotoxicity
Local Ref. No:	
Background:	Unless transplants are protected from attack by *Hylobius abietis*, plant losses throughout UK restocking areas will amount to an average of about 50% over the two years after planting during which they remain susceptible to damage. Since 1983, treating plants by dipping has been the recommended method of protecting plants during the first growing season. PSD announced in April 2000 that dipping plants in permethrin would not be allowed after June 2000. The International Pesticide Application Research Centre (IPARC) was commissioned to develop a system to treat bare-rooted trees with aqueous formulations of insecticide. It was considered essential that adequately treated plants must be delivered by the system dry and ready for storage or dispatch. The IPARC system should offer a number of advantages over dipping such as isolating operators from the point of application and treating plants in a controlled and consistent way. In addition, the aqueous formulations of the insecticides should avoid the increased risk of phytotoxicity associated with the standard Electrodyn formulations. The use of permethrin in agriculture will no longer be allowed in any EC country after the year 2003. Because the IPARC sprayer uses water-based products, it will be possible to use "off the shelf" formulations of insecticides leading to lower registration costs. Trials carried out in 1999 and 2000 have shown that the synthetic pyrethroids, Contest and Hallmark when applied by dipping, provide the same level of protection as the standard permethrin dip. These trials will compare the 2 insecticides that have proved effective when applied by dipping but at higher concentrations suitable for ultra low volume applications.
Objectives:	To compare the growth and survival of transplants of SS sprayed with aqueous solutions of insecticides before cold storage. Contest (150g ai alpha-cypermethrin) and Hallmark (50g ai lambda-cyhalothrin) will be compared with permethrin. All work will include a range of application rates to meet the requirements for approval of plant protection products.
Locations:	Treatments will be applied at IPARC (Ascot) and FC treatment centres. Cold storage, RGP and electrolyte leakage tests will be undertaken at Bush field-station (NRS). The plants for increment assessments should be planted at a Forestry Commission low elevation non-restocking site such as a nursery where damage by *Hylobius* is unlikely.
Species:	Sitka spruce (*Picea sitchensis* (Bong)) will be used for all treatments.

Products and
active ingredients:

Product	Concentration	Active ingredient	Manufacturer
Permethrin 12ED	12% ai ec	permethrin	Techneat Chemicals Ltd
Permasect 25EC	25% ai ec	permethrin	Mitchell Cotts
Contest	15% w/w WG[1]	alpha-cypermethrin	Aventis
Hallmark	10% w/w MC[2]	lambda-cyhalothrin	Syngenta

[1]WG is a formulation of water dispersible granules
[2]MC is an aqueous suspension of microcapsules containing the active ingredient

Experimental
Treatments:

Code		Treatment	Active ingredient	Application rate (% a.i.)	Product rate (g/100 trees)
A	IPARC	Hallmark	lambda-cyhalothrin	0.4	32
B	IPARC	Hallmark	lambda-cyhalothrin	0.8	64
C	IPARC	Hallmark	lambda-cyhalothrin	1.6	128
G	Electrodyn	Permethrin 12ED	permethrin	12	*
H	IPARC	Control	Water	0	*
I	Dip	Permasect 25EC	permethrin	0.8	35 (g/litre)
J	Dip	Untreated control	Water	0	Water Dip

*The actual product usage rates will be determined during the course of the trial.

Design:

There will be 3 storage periods (0, 4 and 8 weeks).
For the **planted trial** there will be 5 replicates (bundles of plants) for each storage period. The 10 treatment plots will be randomised within each block. Each plot will contain 12 plants planted as single rows at 0.3m spacing.
For the **RGP tests** there will be 20 single plant plots for each treatment.
For the **REL tests** there will be 10 single plants for each of the following treatments:
B, C, H & I All using the three storage times

Methods:

Cold stored plants will be used for the trial and allowed to reach a temperature of at least 5°C before treatment. All insecticide solutions will be mixed in a clean container immediately before use. After treatment, dipped plants will be air-dried under cover until the tops are completely dry before they are bagged for storage. Plants treated using the IPARC or Electrodyn systems will be bagged as they leave the treatment system. The plants will be sealed into the co-extruded polythene bags within 15 minutes of treatment and stored at 2°C. Planting or RGP testing will be undertaken within 24 hours of removal from storage. No plant protection products (such as herbicides) should be applied to the plants as a basal treatment.

Requirements and Responsibilities:

Plants – RGP tests = 3 x 7 x 20 = 420 plants
Planted trials = 3 x 12 x 5 x 7 = 1260 plants
REL tests = 3 x 4 x 10 = 120 plants
Marker canes at row ends
Plot marker posts ($3cm^2$)

Records and Assessments:

The RGP tests will be assessed after 14 days in the growth chamber following the method described in SOP018.

	The EL measurements will be made following the method described in SOP069. Plant growth and survival will be assessed between November 2001 and March 2002 following the method described in SOP057. The assessment of growth and survival will be repeated between November 2002 and March 2003.
Duration:	The nursery experiment will be retained until after the winter assessments in 2002-3.
Statistical analysis:	This trial has 2 treatment factors; storage (3 levels) and insecticide treatment (10 levels). For the planted trial there are 3x10=30 plots per randomised block. The effects of storage and insecticide treatment will be assessed using plant increments, RGP and REL measurements. These factors and their interaction will be compared using ANOVA without transformation of the data. The percentage plant survivals from the planted trials will be compared using ANOVA following angular transformation of the data.
COSHH & Risk assessments:	COSHH assessments for plant treatment and for planting of permethrin, alpha-cypermethrin, lambda-cyhalothrin and fipronil treated plants will apply to these experiments.

SOPs: The following SOPs are relevant to this experiment:

SOP No.	**Title of process**
SOP004	Records keeping for field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP018	Measuring root growth potential (RGP)
SOP020	The preparation and mixing of plant protection products.
SOP048	Marking and tallying field experiments that test the efficacy or phytotoxicity of plant-protection products.
SOP056	Using a balance to determine product weight
SOP057	Assessing plant growth as an indication of the phytotoxic effects of insecticides on young forest plants.
SOP062	The manual application of plant protection products by dipping.
SOP066	The use of a cold-store to store plant material
SOP069	Assessment of plant quality through the measurement of cell electrolyte leakage

Bibliography:

Drafted by: Stuart Heritage
Date: 1st March 2001

Design approved: Alvin Milner
Date: 30th March 2001

Approved: Dr H Evans
Date:

Approved: Mr J Dewar (CEO)
Date:

SOPRefNo: SOP006

References: EPPO Guidelines 181 (Bulletin 26)

Key Words: SOP Site Selection

Category: System

Title: Selecting trial sites for experiments that test the efficacy or phytotoxicity of plant protection products.

Scope: This SOP outlines the processes involved m the selection of suitable experimental sites including the administrative procedures required to ensure access to the site. It does not cover the actual installation of the experiment or the assessments of the trial (for these see SOP numbers SOP048, SOP004).

Background: Selection of an experimental site is a critical aspect of efficient and cost-effective experimentation. If the site is inadequate, then the experimental objectives may not be achieved and consequently a product will be inadequately assessed.

Method: The main stages in the selection of a suitable trial site are outlined below;

1. The experiment plan (see SOP002) should specify the area of ground required plus any other key features such as a desired soil, vegetation type and time after felling. The project leader should also make clear whether particular regions of the country are preferred (e.g. a high rainfall area to test rain fastness of a particular product). The plan should also state the maximum duration of the trial.
2. The experiment plan must state clearly the relevant type of PSD experiment approval and any restrictions such as buffer zones that apply.
3. Usually, the experiment plan should be drafted 9-12 months in advance of the planned application of experimental treatments to allow adequate time for site selection.
4. The person(s) responsible for locating potential experimental sites should contact local forest/nursery managers, woodland owners/farmers to see if they have suitable sites available.
5. Once a number of potential sites have been short listed, these must be inspected to check their suitability on the ground. Points which may have relevance during inspection include:
    - access by public; health and safety and the need for warning signs
    - is the site uniform (i.e. flat or of a constant slope)?
    - is the soil uniform or is there variation which will require to be incorporated in the blocking Where soil will have a significant effect on the results; soil pits must be dug at intervals across the site to check for variation

- is there variation in the vegetation which may indicate changes in fertility?
- are there likely to be problems from browsing animals which will require fencing?
- is there any risk off frost or will the site be exposed to wind?
- are there watercourses or other features which could be affected by the use of chemicals (such as SSI's)?
- is the site conspicuous in the landscape so that square plots may be unacceptable?
- is continued and repeated access over a number of years possible at an economic rent?

6. After all the potential sites have been inspected, a shortlist of the best should be drawn up. It is quite possible that no sites will meet the experimental requirements. In this case, other sites should be looked for or the design be reconsidered to see fit can be adapted to the best site. It is a false economy to use a less than adequate site for an experiment and this option should never be considered.
7. Before a site is chosen, it is essential to discuss the terms of research use of the site formally with local management. This may involve discussion of leases or rent on private sector sites.
8. Once a suitable site has been identified, any potential problems (e.g. landscape impact, harvesting schedules) need to be discussed and resolved with the local managers.
9. At this stage, the location of any fence lines should be identified. If machinery is going to be needed (e.g. to cultivate the site), the access routes to the experimental site will need to be identified. Any future management ops. over expt. protocol ops. to be agreed for the period of expt. And any unplanned additional but necessary ops. to be discussed and agreed.
10. TSU manager should write formally to the owner/manager to request the use of the site for the period of the experiment. If a lease is required, this should be prepared in consultation with a specialist land agent. There should always be provision for continued access ifa property is sold before the experiment is completed. An experiment must never go ahead simply on the basis of a verbal agreement to use a site; there must be approval from the owner in writing.
11. Once a particular site has been chosen, the person(s) responsible for the installation of the experiment must revisit the site to lay out the replicates and the treatments plots. It may be important to ensure that all plots in a replicate are located on the same soil type and/or aspect and are at an equivalent elevation. If the layout reveals appreciable variation within a replicate then the project leader must be consulted. If necessary, a site visit with a statistician may be necessary to resolve the problem.

Appendix 5
Field Site Locations

Site	F/S	Location	Forest District	Grid Ref.	Elev. masl	Soil Type
A	Newton	Culbokie	Inverness	NH609586	110	Humus iron podsol
B	Cairnbaan	Fire Tower Road, Lochgilphead	West Argyll	NR 867912	115	Peaty gley
C	Cairnbaan	F1, Lochgilphead	West Argyll	NR 878906	100	Deep peat
D	Bush	Glen Finlet	Tay	NO 244647	360	Upland brown earth
E	Bush	Loch Ard	Aberfoyle	NS 476956	140	Peaty brown earth
F	Bush	Wauchope	Borders	NT 602029	360	Deep peat/peaty gley
G	Mabie	Lauriston	Galloway	NX 658667	180	Peaty gley
H	Mabie	Kirkland	Ae	NY 018913	340	Flushed gley
I	Mabie	The Coomb	Kielder	NY 765915	330	Flushed gley
J	Wykeham	Harwood Dale	North York Moors	SE 974975	130	Peaty Gley
K	Wykeham	Langdale	North York Moors	SE 909955	220	Iron pan
L	Fineshade	Roudham	East Anglia	TL 932 878	30	Sand
M	Talybont	Clocaenog	Llanrwst	SJ 009 555	445	Upland brown earth
N	Talybont	Allens estate	Coed Y Cymoedd	ST 032 951	320	Upland brown earth

Appendix 6
Efficacy tests of 7 nozzle set-ups using the Mardrive spraying apparatus in IPARC

Introduction

Prior to the design of a Sprayer Conveyor system to treat pine saplings with pesticides, it is necessary to test which nozzle should be used on the system. There are a variety of factors to consider on this matter, namely droplet size (somewhat dependent on pressure), spray structure and output volume (dependent on pressure)(Matthews, 1992). These factors are usually heavily intertwined, for example, output volume depends on the pressure of the system, i.e. how hard the liquid is being driven through. At higher pressures, however, the droplet sizes are smaller (Matthews, 1992). Presumably as they have less time to form and have more kinetic energy. Yet coupled with this, larger orifice nozzles create larger droplets, although nozzles always produce a range of droplet sizes as opposed to a single size.

In the new sprayer conveyor a nozzle application system based on a solenoid valve will be used. This is usually used in a tractor sprayer, but will be adapted for this system. This system can vary the volume of the liquid sprayed by altering the pulse rate. In choosing a nozzle, it will be possible to pick any output and any droplet size with in a selected range of values. If these values are plotted together (droplet size vs flow rate) then the area under the curve specifies the combination of droplet sizes and flow rates that can be achieved (Giles, 1997). This is known as a flow rate-droplet size envelope. Alteration of pressure on a tenfold scale can alter the flow rate by a factor of ten. Similarly alteration of the solenoid pulse rate from 20-100% can alter the rate by a factor of 3.2. Therefore, jointly altering these two variables can alter the flow rate by a factor of 32. Within this control envelope a range of droplet sizes can be achieved.

This aspect of nozzle mechanics will be very important to this work, as it will be important to know precisely how much pesticide has been applied to the saplings. By studying a range of control envelopes for different nozzles a series of appropriate nozzles can be selected. With this very accurate information bioassays or field trials can be designed and the effects of differing amounts of pesticides monitored. The advantage with this system over other systems is that the pesticide can be targeted at the regions most attacked by the pest insect. The exact speed at which the sapling is passed through the spray cloud will be known, the exact amount of pesticide released (an accurate timing system will only drive the system for a set time, accurate to thousandths of a second) and also the output of the nozzle. This information will be compared to the volume of pesticide sprayed with the volume of pesticide that reaches its target. To do this the same methods as presented in this experiment will be used. This data can be used to calculate a percentage efficacy of each nozzle system.

Before this stage of the project can be reached, it will be necessary to design an experimental method suitable for calculating the volume of pesticide on the sapling. To do this spectrofluorimetry was used and the volume of dye in the wash measured. UV lighting was used to visualise the area of the saplings that had been treated. To test this method several different nozzle set-ups were compared.

Materials & Methods

In total 77 Sitka Spruce were used. For each nozzle set-up 11 trees were treated, this gave one spare tree in case any were lost in the procedure. Using a converted Mardrive the 11 trees were treated. This was done by suspending the saplings in a clamp upside down and attached to the movable boom in the Mardrive. In the middle retort stands were positioned so that the sapling would pass equally between them and so that the top of the spray cone finished at the root collar, which was the lowest exposed part of the plant. The roots were not treated and were trapped between the clamp hands. For each nozzle the spray angle was checked and then the distance it should be away from the sapling calculated so that a 15cm spray cone would be achieved at the sapling. In this way a 15cm band would be treated on the sapling.

Solid cone disc core type nozzles were used. These were chosen because they offered the widest range of angles. These were attached to Quick Teejet nozzle bodies. A ½ inch tube was used. This was split into two separate lines travelling to the two nozzles using ½ inch connectors, and in later experiments to 4 nozzles. All nozzle components were supplied by Teejet, Spraying Systems Co. USA.

The Sodium Fluorescein was at a concentration of 0.1g l^{-1}, Tinopal was added at a concentration of 2.5g l^{-1}, although it was later found that 1g l^{-1} was suitable.

A handheld trigger system was fitted before the nozzles, so that spray was emitted from the nozzles for the entire period that the sapling passed before them. Pressure in the system was maintained at 1 bar, with check valves built into the bodies to spray at a constant pressure. The saplings moved at approximately 3ms^{-1} with the exact speed recorded each time. On removal of the sapling extreme care was taken not to contaminate the stem with transferred dye, or to accidentally rub some off. Only untreated areas were handled.

Once treated the saplings were left to dry for 30 minutes. As the technique was refined, a second batch would be treated during this time. After drying, the saplings were held under a UV lamp and the coverage assessed. This was done on a scale of 1-5. A score of 5 would be immaculate coverage with no gaps. 4 - would be bare patches, but otherwise good coverage. 3 - approximately 50-70% coverage. 2 – poor coverage, several large gaps. 1 – hardly any coverage. This scale only applied to the bottom 15cms of stem from the root collar upwards. A similar scale was used on the foliage assessment. The last measurement was of the total distance that the spray had reached with coverage of 3 or above. Ideally this should have been 15cms as calculated using the spray cone angles and trigonometry.

Once assessed the saplings were trimmed to a 15cm long stem. All branches and the roots from 2cms below the stem were removed, leaving only needles on the stem. Some large clusters were removed if present. Needles on the stem should ideally have been removed, but if pulled off they took a small section of bark with them. This would have had a significant effect on the spectrofluorimeter score and so they were left. Waste material was discarded. The trees were then washed and the wash measured in the spectrofluorimeter as described in Appendix 8.

3.1.3 Results and Analysis

It was found that the all of the nozzles used achieved very good coverage, only rarely did the coverage even fall as low as three (approximately 50% cover). This was to be expected as the nozzles have a fairly high output (0.36-1.6 l/min) and they were targeted at the stem. Figure A-1 below shows the different coverage with each nozzle set-up.

The second important piece of information to come from this experiment concerns the volume of dye deposited on the lower 15cms of sapling. When the data is arranged in order of output a clear trend can be seen with only one exception. Volume of dye deposited on the plant is directly correlated with the output volume of the nozzle. This is shown in Figure A- 2.

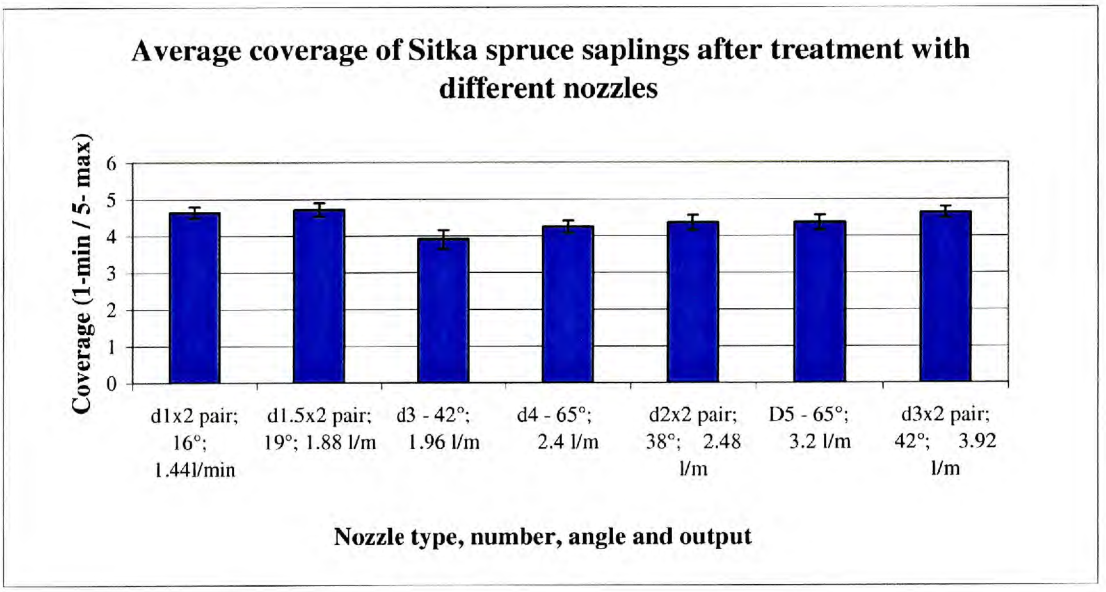

Figure A-1 One way ANOVA showed there to be no difference at a 95% confidence level, P=NS (0.67), df =7. Therefore the coverage did not differ significantly between nozzle set-ups or nozzles. The bars are arranged in order of output volume from lowest to highest, for the combined output of all the nozzles.

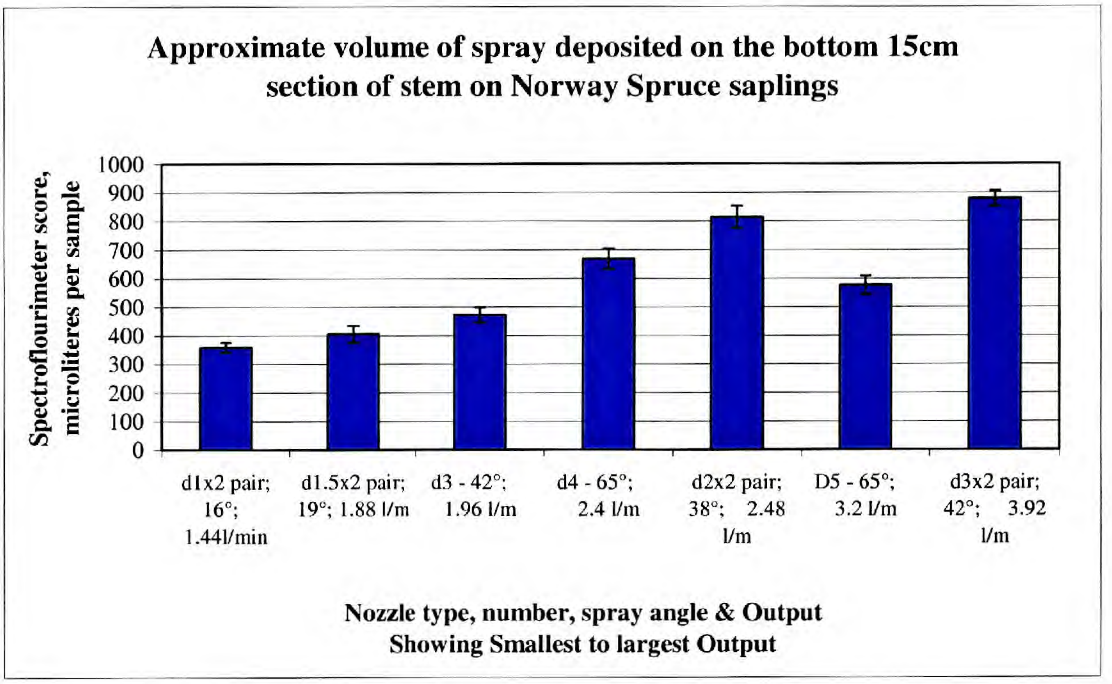

Figure A- 2 One way Anova showed there to be a highly significant difference between groups, ($P=1.46 \times 10^{-20}$, df = 6). Therefore the nozzles were depositing differing amounts of dye onto the sapling.

One interesting feature with these two sets of results is that all though volume varies between nozzles, the coverage does not. This is a very important point, and shows that it will be possible to minimise the volume of spray used with out jeopardising the coverage. The results from this experiment show that the D1 nozzles (lowest angles and flow rate) used the least volume of spray but had coverage comparable to that of the other nozzles.

It is important to note that the single pair of D5 nozzles deposited a much lower volume of dye than the other set-ups, despite its higher output. This can only be attributed to an error in the measurement process, or possibly the dye concentration fell.

3.1.4 Discussion and Conclusion

This experiment highlighted several important aspects of pesticide application. The D1 nozzles provided the best coverage to deposition ratio. Unfortunately these nozzles needed to be positioned over 20cms away from the sapling, which may experience more problems (such as blocking by branches) than nozzles positioned closer. It also required 2 pairs of nozzles to get full coverage of the 15cms of stem. There will inevitably be some overlap between the nozzles and therefore extra dye/pesticide would unnecessarily be deposited.

Clearly a balance must be found between volume of pesticide and practicality of the set-up. Another factor to consider is that lower volume nozzles are at a higher risk of becoming blocked, particularly if the pesticide is going to be recycled.

This series of experiments has demonstrated that these experimental methods work and that the two dyes can be mixed without a major effect. A short experiment was set up to test this and although the addition of a second dye significantly reduced the value from the spectrofluorimeter, the between sample variance was fairly constant. This indicates that the change is constant and therefore the nozzles can be ordered accurately in terms of deposition, even if the calculation of deposit is a slight underestimate.

A key problem with this experiment is that testing the nozzles over several days had the potential to produce different results with differing initial dye concentrations. Secondly the dye is also degraded in sunlight, and this can again affect the results from the first treatment to the last. Hopefully with the new conveyor system the trees will be able to be treated much more quickly and all in one go. This may explain the reduced deposit seen when using the D5 nozzles. Alternatively it is possible to that this nozzle (being the largest of the five used) had an altered droplet spectra, resulting in a difference in deposition.

Narrower nozzles need to be positioned proportionally further away than wider angle nozzles to treat the same area of the plant. This can be used to my advantage when building the conveyor system. Secondly increasing the spray pressure increases the angle of the spray cone in many nozzles, again another feature, which can be exploited.

In summary this experiment has shown that nozzles with greater output volumes deposit a greater volume of dye onto the sapling. Two pairs of nozzles are not necessarily better than one and will certainly bring more technical difficulties.

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Appendix 7
Use of the Spectrofluorimeter to ascertain the volume of dye sprayed on to Sitka spruce saplings

Sodium fluorescein can be used as a fluorescent dye, in solution under UV it glows bright yellow/green. The use of a spectrofluorimeter can identify the extent of the fluorescence and quantify it. This requires an excitation filter; NB490 & an emission filter; SC515. In conjunction with these excitation filters a plate with a small hole is used. The aperture of this hole can be varied by changing the plates. A smaller aperture is required for more concentrated samples.

An initial concentration of 0.1g/l of Sodium fluorescein was used, however, this was later changed to 0.25g/l. The dye solution was sprayed onto the plants and leaving the plant to dry. The section of the sapling to be tested was removed, placed in a plastic resealable bag and then washed in 0.25l to 0.5l of water depending on the experiment. Alternatively a 0.02 molar solution of sodium hydroxide could be used. The samples were agitated in a cyclic shaker for 30 minutes.

In addition to the sprayed saplings, a 100μl sample was placed onto part of the sapling. This was left to dry and treated in the same way as the other samples. This was the control/standard.

The spectrofluorimeter was allowed to warm up for 30 minutes. A 3cm cuvette was filled with tap or distilled water (if used) and put in the spectrofluorimeter. To set up the spectrofluorimeter, the span dial was set to maximum and the range/scale dial to a x50 setting, although this was dependent on the experiment. The system was zeroed using the remaining dial. A sample of the control was placed into the spectrofluorimeter and measured. The span dial was used to adjust the output so that it read 100. This would give a direct reading of microlitres per sample.

A small subsample from each sample was put into a separate clean cuvette, wiping the outside if necessary. The cuvettes were kept in a labelled rack. The individual samples were then measured. Finally, a calibration curve was created by taking the readings of a series of known concentrations and then plotting them against the score that the spectrofluorimeter returned. The spectrofluorimeter readings could then be converted to actual concentrations using the formula returned by a regression line.

Appendix 8
Nozzle Spectra and Droplet Analysis using the Malvern Particle Analyser

Background

It has been long established that the size and structure of droplets can have both important biological and ecological effects. Larger droplets tend to have reduced risk of drift, but tend to be more dispersed and therefore pest insects are less likely to encounter them. Whilst smaller droplets are more prone to drifting away from the spray area in the air currents, but are more numerous and increase the chances that the target insect will encounter it.

The DCS system protects coniferous saplings from weevil attack by *Hylobius abietis* through prophylactic treatment of the stem with a pyrethroid insecticide. This type of treatment affords up to a seasons protection in the field. This system is an indoor-based system and fully self-contained, therefore drift of droplets to neighbouring fields is not applicable. Ideally an even coating of small droplets across the bark would be preferable to larger droplets, which may make the sapling 'wetter.' In this system operator contamination is the major biological concern (compared to environmental contamination) and the saplings need to remain as 'dry' as possible after treatment. Therefore the selection of a nozzle for this system must be based on several criteria.

Previous work with a range of nozzles in the Dandrive has shown that the Air Induction nozzles leave the sapling wetter (Chapters 4 & 5), with larger droplets that may more readily run off. In contrast the relatively smaller droplets of the Tp flat fan nozzles leave an even film over the surface of the bark, without any individual droplets obviously visible. This generally reduces the contamination or passing of pesticide from the sapling to the operator's gloves.

It was decided that a quantitative comparison should be made between the two most common nozzles used in the trials on the Dandrive. These were a flat fan nozzle (Fe), that has been designed to have an even droplet size spectra from the center of the fan to the edge. The second nozzle is an air induction nozzle that sucks air into a tiny chamber as the droplet is formed and then includes tiny bubbles in to the droplet. This creates a much bigger droplet but with a lower volume of pesticide, hopefully having the advantages of both big and small droplets.

The Malvern particle analyser measures droplet diameter by recording the defraction of a laser light onto a ring of detectors. This defraction is coupled with light intensity and the VMD (Volume mean diameter, the standard measurement for droplet diameter) can be calculated. For this series of trials the eveness of the droplet size at different points in the fan were measured, i.e. the center, the edge and the mid point between these two points. In addition a measurement was taken through the horizontal axis of the fan giving an average droplet size across all the positions. This last measurement was taken with the nozzle at two different distances from the laser.

Materials & Methods

Two nozzles were selected, TP6500E (even spray flat fan, Spray systems) and an Air Inclusion, 120°, 0.151 min^{-1} flow rate (Lurmark). The nozzles were attached to a compression canister and a triggered spray gun. Initially the nozzle tip was 20cm from the laser, but one of the trials varied this distance. Three replicates were made for each setting. Normal water was used and sprayed at 2.5 bar, room temperature. For each replicate a background measurement was made. This was subtracted from the actual measurement before the Malvern software produced charts and printed tables of the VMD, Span (width of spray through laser) and Obscuration (Percentage of space occupied in the 1cm diameter of the laser). The spray period was typically 10 seconds. This provided enough data for the software to analyse the data accurately.

The first trial measured the VMD at three points in the fan, the center, the near edge and the outer edge. The near edge was a point half way between the outer edge and center. For these trials the spray fan was vertical, i.e. a narrow rectangle pointed from ceiling to floor. These points were calculated visually as the laser light was scattered at the points where it passed through the fan. This was repeated for each of the three nozzles.

The second trial measured the average droplet size through the horizontal axis at two different distances from the nozzle. These distances were set at 20cm, the standard practice and 10cm. The latter distance was chosen because it is the distance that the nozzles are normally positioned from the saplings in the Dandrive. This gives approximately a 15cm long rectangle for a 65° nozzle. The AI nozzle was not measured at 20cm as its larger spray angle (120°) would cause droplets to hit the laser and detector.

Data

AI Nozzle	**Rep 1**	**Rep 2**	**Rep 3**	**Rep 4**	**Means**
Outer edge	629.4	654	30.5	799.16	799.16
Near edge	260.08				260.08
Centre	573.85	107.17	199.46		153.315

TP Nozzle	**Rep 1**	**Rep 2**	**Rep 3**	**Rep 4**	**Means**
Outer edge	123.77	885.86	1104.52	172.94	148.355
Near edge	1123.06	1054.22	1044.54	1148.24	1092.515
Centre	892.41	560.97	1080.97	1111.88	911.5575
10cms	1110.98	518.92	585.54	254.19	452.8833
20cms	193.5	283.07			238.285

Data points indicated in grey are likely to be unreliable and much higher than expected. Typically they had very low obscuration values indicating that too few droplets passed in front of the laser.