

# Recognition and avoidance of insecticide-treated Scots Pine (*Pinus sylvestris*) by *Hylobius abietis* (Coleoptera: Curculionidae): implications for pest management strategies

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- Abstract**
- 1 The feeding preferences of *Hylobius abietis* (L.) were studied in a series of choice and no-choice trials for insecticide-treated food, time-to-death studies and arena trials.
  - 2 Treatment of Scots Pine twigs with a pyrethroid insecticide, lambda-cyhalothrin, was compared with twigs treated with imidacloprid, a neonicotinoid.
  - 3 Clear avoidance of insecticide-treated food sources, with strong evidence of selection for untreated food sources, was shown.
  - 4 In addition, it took up to 3 weeks for *H. abietis* to die from insecticide poisoning when fed on treated food and, during this time, it was potentially capable of finding new untreated food sources.

**Keywords** *Hylobius abietis*, insecticide recognition, lambda-cyhalothrin, pyrethroids, Sitka spruce.

## Introduction

Since the mid-1980s, the large pine weevil, *Hylobius abietis* (L.) (Coleoptera: Curculionidae) has become an increasing nuisance in temperate forestry in the U.K. (Heritage *et al.*, 1989; Moore, 1998, 2000; Leather *et al.*, 1999) and is the major insect pest of forest regeneration in the Scandinavian countries (Orlander *et al.*, 1997; Orlander & Nilsson, 1999). Congeneric species, such as *Hylobius pales* (Herbst) and *Hylobius congener* (Dalla torre Schenkling & Marshall) (Coleoptera: Curculionidae), fill similar niches in North America (Lynch, 1984; Welty & Houseweart, 1985).

*Hylobius abietis* may cause from 30–100% mortality of transplanted saplings in the absence of insecticide treatment (Heritage *et al.*, 1989) and, accordingly, the U.K. Forestry Commission uses a prophylactic insecticide treatment. The insecticide permethrin has typically been applied using either the electrodyn sprayer conveyor system or by dipping (Stoakley & Heritage, 1990; Heritage, 1996). 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* spp. (Coleoptera: Scolytidae) (Heritage, 1997). Prophylactic treatment avoids the necessity of spray-

ing nontarget areas and therefore some of the potentially hazardous effects of insecticide use.

Until recently, it has been unclear how the insecticide treatment provides protection, although the pyrethroids in question are known to be repellent to many insects (Herve, 1985).

In the present study, first, the preferences of *H. abietis* for treated and untreated food were determined in simple choice and no-choice tests. Second, time-to-death studies were used to determine how long *H. abietis* lived in the presence of treated food and whether death occurred shortly after treatment or after a longer period. Finally, an arena was used with greater numbers of insects and trees to simulate more natural conditions at a lower insect density.

## Materials and methods

### Collection of weevils

All adult *H. abietis* were collected by the Forestry Commission using Scots pine, *P. sylvestris* (Pinaceae) billet traps at Newton Field Station (57° 30' N, 4° 10' W) on the 26 October 2001. A maximum of 100 *H. abietis* were kept in a plastic box (280 mm × 160 mm × 70 mm) at 5 °C for no more than 2 months before use. Sex was determined and the weevils were starved for a period of 48 h at 15 °C before use in the experiment to ensure comparable feeding damage between replicates.

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### Twig treatment

A single method of treatment was used to spray insecticide onto the prepared Scots Pine twigs. Small branches were collected from Crowthorne woods, Bracknell, Berkshire (51° 23' N, 0° 46' W) a few days before the experiment. *Hylobius abietis* have been shown to feed on twigs up to 20 mm in diameter (Orlander *et al.*, 2000). The branches were cut into 100-mm lengths and sorted into approximate size classes, but only one size class was used for any one individual experiment to avoid any element of choice by size selection. Twigs from 5–10 mm in diameter were used, with those twigs outside these boundaries being discarded.

Twigs were placed on tin foil covered plastic trays (500 mm × 300 mm) arranged side by side and with a 1.5 cm gap between each twig. Twigs were then sprayed using the Mardrive (The International Pesticide Application Research Centre, U.K.) that simulates tractor or conveyor spraying methods. A boom housed inside a closed cabinet (4 m long × 2 m × 2 m) passes over a spray target using compressed air. Spray settings, such as pressure, nozzles and boom speed, can all be set independently. Approximately 0.018 mL of insecticide solution or water was sprayed onto each twig using a FE65 even spray flat fan nozzle operated at 2.5 bar pressure. The trays of twigs were then left to dry for 24 h at room temperature.

### No-choice and choice experiments

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 insecticide deposit on a food source. All trials were carried out under an LD 16:8 h photoperiod in a controlled temperature room (CT room) at 15°C; ambient humidity.

#### Choice trial

Thirty *H. abietis* were assigned to one of three treatments: (i) two control twigs; (ii) two treated twigs; and (iii) one control and one treated twig. Each treatment involved 10 replicates with five males and five females. Treated twigs were sprayed with either a pyrethroid formulation of 0.1% active ingredient (a.i.) of lambda-cyhalothrin (Hallmark EC, 10% w/v) or a neonicotinoid formulation of 0.1% a.i. imidacloprid (Provado WP, 5% w/w). Control twigs were sprayed with distilled water under similar conditions but dried in a separate location to avoid cross contamination.

For each treatment two 100-mm Scots Pine twigs were placed in resealable plastic containers (100 mm × 150 mm × 60 mm) 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 7 days. The weevils were then assessed for mortality and the twigs assessed for damage.

#### No-choice trials

Thirty *H. abietis* were assigned to one of three treatments: (i) control; (ii) 0.1%; (iii) and 0.5% insecticide treatments.

Each treatment involved 10 replicates with five males and five females.

Treated twigs consisted of either 0.1 and 0.5% a.i. lambda-cyhalothrin or 0.1 and 0.5% a.i. imidacloprid. Control twigs were treated with distilled water under similar conditions and dried in a separate location to avoid cross contamination.

For each treatment, one Scots Pine twig (*Pinus sylvestris*) was placed in to a resealable plastic container (100 mm × 150 mm × 60 mm) as previously described above. The experiment was run for 7 days at the end of which mortality of *H. abietis* were assessed. Twig damage was measured as described below.

### Time-to-death experiments

Forty-two *H. abietis* were assigned to one of four treatments: (i) control; (ii) starved; (iii) a pyrethroid formulation of 0.1% a.i. lambda-cyhalothrin; (iv) or a neonicotinoid formulation of 0.1% a.i. imidacloprid. *Hylobius abietis* were placed into resealable plastic containers as described above. Each container held one twig, except for the starved group, which did not receive any food. *Hylobius abietis* were checked every 24 h for mortality. A weevil was considered dead when it was unresponsive to poking or stroking of its antennae. Because *H. abietis* shows exceptional thanatosis, measurement of mortality was always continued for a few days after presumed death. The containers were moistened once a week with a hand mister.

### Arena experiment

An arena trial was set-up to examine whether previously established relationships were maintained in a larger environment using live saplings. Three 3-year-old Sitka spruce, *Picea sitchensis* (Pinaceae) were planted in large 25-L black pots (400 mm in diameter) filled with 5 L of soil. The arena was controlled to maintain conditions between 15 and 22°C. Natural lighting was supplemented with sodium lamps to encourage growth of the Sitka spruce during the winter period used for the experiment.

The experiment consisted of four treatments: (i) three control trees; (ii) two control trees + one treated; (iii) one control tree + two treated; and (iv) three treated trees. Treated trees had been sprayed with a 1% solution of lambda-cyhalothrin 2 days before planting. Treatment took place in the Mardrive apparatus under conditions described above.

Five *H. abietis* were added to each pot and fine muslin mesh covers were constructed to cover the trees and pot. Weevils were starved for 48 h before the experiment. These treatments were then left for 2 weeks in the arena under an LD 16:8 h photoperiod and minimum daily temperature of 15°C; ambient humidity.

### 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 100 mm × 30 mm piece of acetate with a black acetate pen. The damage was then scanned using an

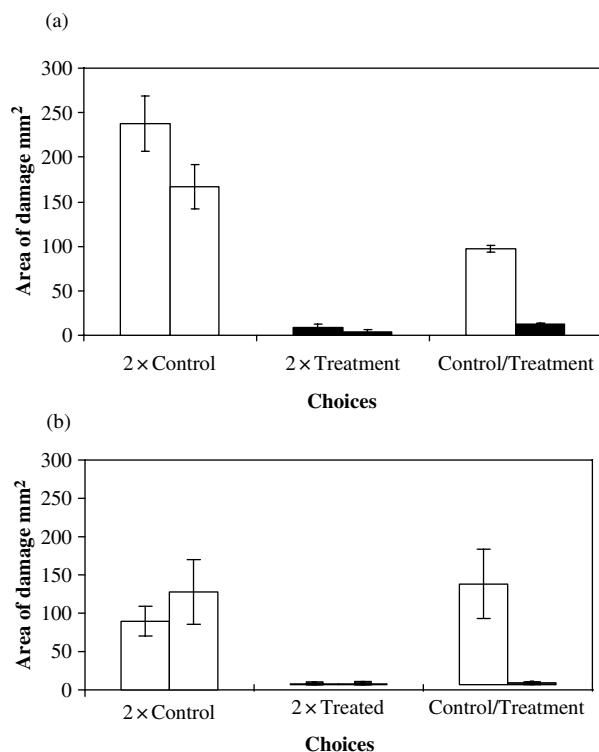
Epson Scanner (Epson Corporation, Japan) and measured using Area V (designed by Peter Mueller, Imperial College), which measures the area of selected objects. The mean area of damage per treatment was then calculated and this was analysed using analysis of variance (ANOVA) in S-PLUS (Version 6.2, Insightful Corp., Seattle).

## Results

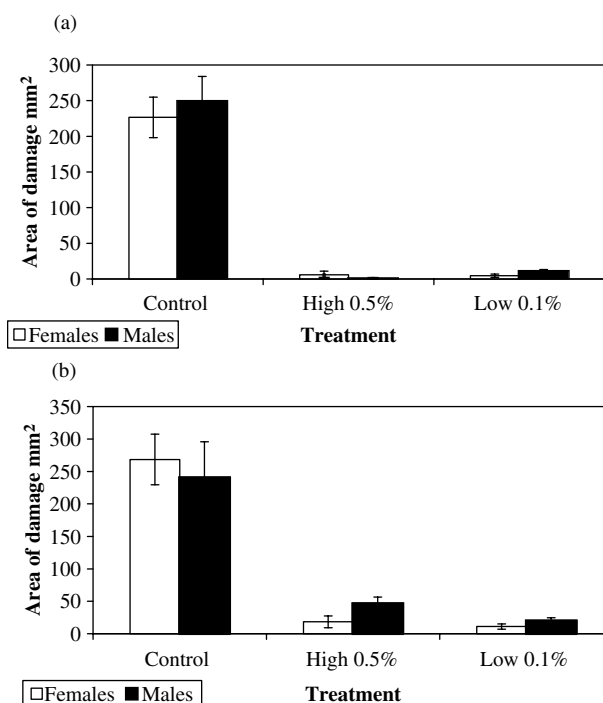
### Choice and no-choice

In the choice trials, there was a clear effect of preference against both insecticides (lambda-cyhalothrin:  $P \leq 0.05$ ,  $F = 4.05$ ; imidacloprid:  $F = 13.54$ ; d.f. = 2,  $n = 30$ ) when the combined damage per treatment was examined (Fig. 1). In addition, comparisons showed a difference between the damage for treated and control twigs in the control/treatment group (lambda-cyhalothrin:  $P \leq 0.05$ ,  $F = 15.564$ ; imidacloprid:  $F = 16.89$ ; d.f. = 1,  $n = 30$ ).

The no-choice trials showed a similar pattern of damage. There was a significant difference in the levels of feeding damage of *H. abietis* between treatments for both insecticides. Treated twigs were largely untouched whereas untreated ones were heavily damaged (lambda-cyhalothrin:  $P \leq 0.05$ ,  $F = 46.89$ ; imidacloprid:  $F = 103.55$ ; d.f. = 2,  $n = 30$ ). There was no difference between the two insecticide concentrations (Fig. 2).



**Figure 1** Area of damage by *Hylobius abietis* in choice trials with (a) lambda-cyhalothrin and (b) imidacloprid. White bars, treated twigs; black bars, insecticide-treated twigs.



**Figure 2** Area of damage by *Hylobius abietis* in no-choice trials with Scots pine twigs treated with two concentrations of either (a) lambda-cyhalothrin or (b) imidacloprid.

### Time to death

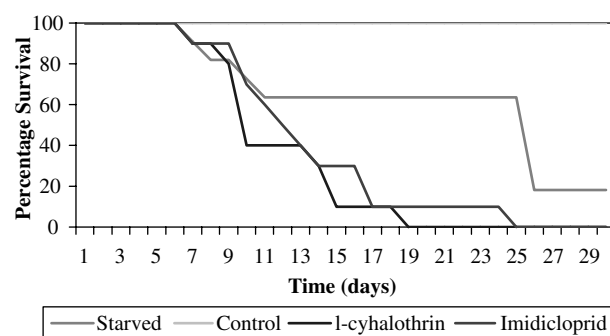
There were apparent differences in the survival of insecticide-treated, starved and control groups (Fig. 3): survival of insects in the two insecticide groups showed very similar trends, whereas weevils lived much longer in the starved group ( $\chi^2 = 22.24$ ,  $P < 0.001$ ; d.f. = 3;  $n = 43$ ). There was a significant difference between insecticide treatments and the starved group in both mean time to death ( $\chi^2 = 9.5$ ,  $P = 0.002$ ; d.f. = 1;  $n = 31$ ) and also in the shape of the fitted curves (survival regression,  $P = 0.00011$ ;  $Z = 34.06$ ; d.f. = 1;  $n = 31$ ).

### Arena trial

There was a strong negative trend in the level of measured damage as the total number of trees treated increased ( $r^2 = 0.66$ ,  $y = 1894.22 - 0.812x$ ) (Fig. 4). This indicated that when untreated trees were rare, the damage they suffered would be lower.

## Discussion

The present study showed that *H. abietis* was able to detect the presence of insecticide, notably lambda-cyhalothrin and imidacloprid. Feeding on food sources treated with these insecticides was significantly depressed and, in most cases, nonexistent. Previous studies have shown that *H. abietis* exhibit novel behaviour in artificial conditions compared with field conditions (Manlove *et al.*, 1997). However, the



**Figure 3** Survival of *Hylobius abietis* when starved, fed (control) or fed insecticide-treated food.

arena trial in the present study, which used conditions as close to field conditions as possible (i.e. using live Sitka spruce, natural lighting and fluctuating temperatures: 15–22 °C), produced results consistent with the other experiments. This suggests that, in the field, *H. abietis* would be able to choose food sources that are insecticide free.

*Hylobius abietis* took up to 3 weeks to die when fed on insecticide-treated food. This mortality may have been due to the action of the insecticide or a combination of starvation and insecticide action. If the effect was due to the insecticide, then *H. abietis* would only have a limited amount of time to find other food sources and damage them before it dies. However, *H. abietis* may recover from temporary exposure and continue to damage Sitka spruce saplings, particularly any that are untreated or have been inadequately treated or failed to be treated at all. Alternatively, a lethal level of exposure may have been suffered, but continued feeding damage on a variety of sources may occur until *H. abietis* dies some days later.

The evidence from this trial indicates that there may be a different underlying cause of death between starved and insecticide-treated groups, as shown by the different mean time to death and the shape of the time-to-death curve. Therefore, the initial hypothesis that death after pesticide

exposure was by starvation was rejected. However, the prolonged time to death of *H. abietis* exposed to a field rate of insecticide should be a cause for concern.

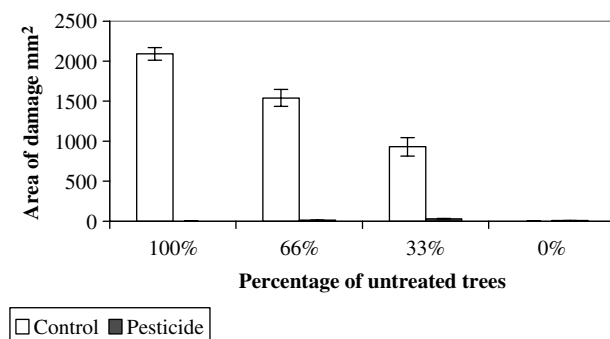
The evidence from the present study strongly suggests that *H. abietis* is likely to avoid treated trees in the field and seek out those that were inadequately treated in the nursery before plantation. However, the arena trial also showed that there was a decline in damage done to untreated trees as they became rarer. In this arena trial, some partial feeding or contact with the treated trees may have acted as an appetite suppressant, causing less damage on the untreated trees. It is not possible to state with certainty whether this would occur in the field where the saplings are spaced much further apart.

Field populations of *H. abietis* have been estimated to be as high as 150 000 (Heritage, 1996) and 220 000 adults per hectare (Leather *et al.*, 1995). Densities in this experiment were equivalent to approximately 400 000 weevils per hectare. However, the complete lack of feeding damage to many of the treated trees and twigs, even after several weeks without suitable food, indicates a very strong dislike by *H. abietis* of insecticide-treated food. Therefore, the relationships demonstrated in these experiments are likely to be maintained in a field situation.

In summary, *H. abietis* show a strong preference for untreated food sources and was capable of making this choice in both laboratory and simulated field 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 (Orlander *et al.*, 2001; Nordlander *et al.*, 2003a).

The distances that *H. abietis* has been reported to migrate is quite variable, ranging from walking tens of metres (Leather *et al.*, 1995) to flying hundreds of kilometres (Solbreck, 1980). Because incidences of both long and short migrations have been shown, based on mark recapture studies, it is likely that migrations depend on the local resources (Nordlander *et al.*, 2003b). Mass exoduses by insects in response to poor local resources have been suggested by Watt *et al.* (1989) and restructuring planting strategies to provide less attractive hosts to *H. abietis* would be a simple way of reducing damage (Leather *et al.*, 1994). For *H. abietis*, the most important resource is breeding material (i.e. tree stumps). Current research suggests that, once *H. abietis* has entered a site, it will not leave again due to atrophication of the wing muscles (Nordenhem, 1989). Therefore, the behavioural effects of insecticide use in this system may have far reaching effects on population structure and abundance.

As the insecticides are offering protection through a repellent action, and insecticidal properties second, it may be that the basis of environmentally friendly tools is already available to prevent *H. abietis* damage. Future research should focus on the behaviour of *H. abietis* in the field, with respect to feeding preferences and response to insecticide-treated saplings. Forest managers may be able to implement new strategies that take advantage of the response by *H. abietis* to pesticides; for example, movement away from a treated food source, preference for an untreated food source or the



**Figure 4** Area of damage by *Hylobius abietis* to trees treated with pesticide and untreated trees exposed to *H. abietis* in different proportions.

use of alternative food sources to lure damage to noneconomically important plants.

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## References

- Heritage, S. (1996) Protecting plants from damage by the large pine weevil and black pine beetles. *Forestry Commission Research Information Note 268*, pp. 1–8. HMSO, U.K.
- Heritage, S. (1997) Protecting plants from weevil damage by dipping or spraying before planting using aqueous insecticides. *Forestry Commission Research Information Note 270*, pp. 1–4. HMSO, U.K.
- Heritage, S.G., Collins, S.A. & Evans, H.F. (1989) A survey of the damage by *Hylobius abietis* and *Hylastes* sp. in Britain. *Insects Affecting Reforestation: Biology and Damage*. (ed. by R. I. Alfaro and S. G. Glover), pp. 28–33. Forestry Canada, Pacific Forestry Centre, Canada.
- Herve, J.J. (1985) Agricultural, public health and animal health usage. *The Pyrethroid Insecticides* (ed. by J. P. Leahey), pp. 347. Taylor & Francis, U.K.
- 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.
- Leather, S.R., Small, A.A. & Bogh, S. (1995) Seasonal variation in local abundance of adults of the large pine weevil, *Hylobius abietis* L. *Journal of Applied Entomology*, **119**, 511–513.
- Leather, S.R., Day, K.R. & Salisbury, A.N. (1999) The biology and ecology of the large pine weevil, *Hylobius abietis* (Coleoptera: Curculionidae): a problem of dispersal? *Bulletin of Entomological Research*, **89**, 3–16.
- Lynch, A.M. (1984) The pales weevil, *Hylobius pales* (Herbst.): a synthesis of the literature. *Journal of the Georgia Entomological Society*, **19**, 1–34.
- Manlove, J.D., Styles, J. & Leather, S.R. (1997) Feeding by the adults of the large pine weevil, *Hylobius abietis* (Coleoptera: Curculionidae). *European Journal of Entomology*, **94**, 153–156.
- Moore, R. (1998) Population dynamics, migration and control of the large pine weevil (*Hylobius abietis*). *Forest Research Annual Report and Accounts*, **1997–98**, 70–74.
- Moore, R. (2000) Population dynamics, migration and control of the large pine weevil (*Hylobius abietis*). *Forest Research Annual Report and Accounts*, **1999–2000**, 70–74.
- Nordenhem, H. (1989) Age, sexual development, and seasonal occurrence of the pine weevil *Hylobius abietis* (L.). *Journal of Applied Entomology*, **108**, 260–270.
- Nordlander, G., Bylund, H., Orlander, G. & Wallertz, K. (2003a) Pine weevil population density and damage to coniferous seedlings in a regeneration area with and without shelterwood. *Scandinavian Journal of Forest Research*, **18**, 438–448.
- Nordlander, G., Orlander, G. & Langvall, O. (2003b) Feeding by the pine weevil, *Hylobius abietis* in relation to sun exposure and distance to forest edges. *Agricultural and Forestry Entomology*, **5**, 191–198.
- Orlander, G. & Nilsson, U. (1999) Effect of reforestation methods on pine weevil (*Hylobius abietis*) damage and seedling survival. *Scandinavian Journal of Forest Research*, **14**, 341–354.
- Orlander, G., Nilsson, U. & Nordlander, G. (1997) Pine weevil abundance on clear-cuttings of different ages: a six year study using pitfall traps. *Scandinavian Journal of Forest Research*, **12**, 225–240.
- Orlander, G., Nordlander, G. & Wallertz, K. (2001) Extra food supply decreases damage by the pine weevil *Hylobius abietis*. *Scandinavian Journal of Forest Research*, **16**, 450–454.
- Orlander, G., Nordlander, G., Wallertz, K. & Nordenhem, H. (2000) Feeding in the crowns of Scots pine trees by the pine weevil *Hylobius abietis*. *Scandinavian Journal of Forest Research*, **15**, 194–201.
- Solbreck, C. (1980) Dispersal distances of migrating pine weevils *Hylobius abietis* (Coleoptera, Curculionidae). *Entomologia Experimentalis et Applicata*, **28**, 123–131.
- Stoakley, J.T. & Heritage, S.G. (1990) Approved methods for insecticidal protection of young trees against *Hylobius abietis* and *Hylastes* species. *Forestry Commission Research Note 185*. HMSO, U.K.
- Watt, A.D., Leather, S.R. & Stoakley, J.T. (1989) Site susceptibility, population development and dispersal of the pine beauty moth in a lodgepole pine forest in northern Scotland. *Journal of Applied Ecology*, **26**, 147–157.
- Welty, C. & Houseweart, M.W. (1985) Site influences on *hylobius*-congener (Coleoptera, Curculionidae), a seedling debarking weevil of conifer plantations in Maine. *Environmental Entomology*, **14**, 826–833.

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