Ecology, management and damage by the large pine weevil (*Hylobius abietis*) (Coleoptera: Curculionidae) in coniferous forests within Europe

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Abstract

Coniferous forests in Europe have a considerable number of pests that attack trees of all ages from youngest up to oldest ones. One of them is the large pine weevil *Hylobius abietis*. This species is widespread in Europe and occurs from warm southern areas (Spain) up to northern countries (Sweden, Finland). Larvae of this species do not cause damage, they help in the decomposition process of stumps and roots. Adults are harmful as they consume bark of young coniferous seedlings, above all of spruce and pine. One adult can consume on average 75 mm² of bark per day. Individuals can live up to 4 years, and during their lives they can damage and kill several tens of coniferous seedlings. Traditional chemical protection of seedlings against this pest is and will gradually be more and more restricted or forbidden. In this review, we describe this method as well as all other alternative methods including biological protection. We estimate that *H. abietis* causes annual damages in Europe of almost 120 million € and damages several tens of thousands of hectares of young forest stands.

Key words: biocontrol; *Beauveria bassiana*; control; costs; life history; pest

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1. Introduction

The large pine weevil *Hylobius abietis* (Linnaeus 1758) (Coleoptera: Curculionidae) is an important forest pest that is widespread over the whole Europe and Asia (Scott & King, 1974; Långström & Day 2004). It is estimated that in Europe it annually causes damages equal to 140 million € (Långström & Day 2004). Since the beginning of the last century it has been considered a pest of young coniferous forest stands (Trägårdh 1913). Adults are pests of young coniferous seedlings (Stoakley & Heritage 1990; Wallertz et al. 2006; Thorpe & Day 2008; Gradinariu et al. 2012), but can also feed on roots or branches of older coniferous trees (Eidmann 1974; Örlander et al. 2000; Wallertz et al. 2006). Population of *H. abietis* is dependent on the number of fresh stumps (Långström & Day 2004). The number of fresh stumps suitable for the weevil development increases due to wind and bark beetle disturbances (Kunca et al. 2016), which provide more habitats for the reproduction of *H. abietis* (dead

and dying trees; Nordlander 1987), and its reproduction is stimulated by logging and subsequent planting, which is a prevailing approach in European forestry (Nilsson et al. 2010). Several seedling protection methods are described below.

2. Bionomics and a life-cycle

Hylobius abietis adults are dark brown with yellow and light-brown spots arranged in irregular rows on their elytra. They are 10 to 14 mm long; their head is extended to a distinct snout with antennae and a mouth opening at its end (Zúbrik et al. 2013).

Larvae develop in stumps and roots of dying and dead coniferous trees. Eggs are laid in small holes in the bark of roots gnawed by adults at the end of spring (Scott & King 1974; Nordenhem & Nordlander 1994; Inward et al. 2012). Larvae develop through four larval stages before pupation. Eggs occur in the bark of roots or stumps just

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under the soil surface, and after hatched larvae migrate to roots (Pye & Claesson 1981).

Adults emerge from soil, roots or stumps in spring, when temperatures reach 8–9 °C (Munro 1928; Nordenhem 1989). In spring, adults consume bark of coniferous and other tree species, and they mature sexually before mating and egg laying. According to Fedderwitz et al. (2018), *H. abietis* spends 6% of its lifetime for feeding, while they feeds 4 to 5 times a day and consume approximately 13 mm² of bark at one feeding. Adults stay at one place after egg laying until the end of summer (July to September), but most of them overwinter in the pupal chamber (Nordenhem 1989). Overwintering can occur in the third or fourth instar phase and in maturity. An average super cooling point of larvae is –12.6 °C (Luik & Voolma 1989). This temperature is not very low (Leather et al. 1993), which is probably due to *H. abietis* adaptation to overwintering in soil. Adults can live up to four years (Eidmann 1979; Leather et al. 1999). Sex ratio is around 50:50, although the proportion of trapped female beetles can vary between 20 to 70% depending on the season (Långström 1982; Tilles et al. 1986; Djeddour 1996; Lalík et al. 2019). In the first egg-laying season, young female need two to three weeks to reach their sexual maturity, while females in the second or third year are immediately ready to mate and lay eggs (Fig. 1a) (Bejer-Petersen et al. 1962; Nordenhem 1989). After mating females search for fresh coniferous stumps that are suitable for egg laying. They chew holes in the bark, in which they lay between one and five eggs (Salisbury 1996). The

egg-laying period lasts from May to September with the peak between the middle of May and the beginning of June (Bejer-Petersen et al. 1962; Lekander et al. 1985). Egg laying and insect fertility are affected by a number of factors, e.g. size, life span, nutrition of adults and larvae, and plant stimulants (Leather 1987, 1988; Leather et al. 1995). The eggs are laid most frequently within 10 cm part of stumps under ground, although some eggs were found in the depths of more than 40 cm (Pye & Claesson 1981). In Scandinavia (Nordenhem 1989) and at higher elevations of Central Europe (Lalík et al. 2019), stumps can remain suitable for egg laying for three years. Development of eggs and larvae is faster in stump parts closer to soil surface that are warmer than those deeper under the ground. Fast development increases the survival chance because of sibling cannibalism (Henry 1995) and natural enemies (Pye & Claesson 1981).

Larvae are whitish with brown heads, legless, slightly arched, and 12 to 23 mm long (Fig. 1b). Pupae are loose, white, soft, with relatively long and narrow appendages. Body parts of a future adult including its nose are visible on a pupa (Zúbrik et al. 2013).

Depending on the microclimate and the quality of host stumps, larvae can develop from an egg into adult within one year, or larvae can pupate, and adults can develop only in the next year. In colder climatic conditions, larval development usually lasts two years, but can take up to five years. However, 75% of individuals in Europe develop within one year (Bejer-Petersen et al. 1962; Scott & King 1974). Sunny place on stumps also positively affect larval development. Larvae closer to the

Fig. 1. Developmental stages of *Hylobius abietis*: a) egg, b) larva, c) adult, d) adult feeding on the spruce seedling (photo by authors).

soil surface develop faster than those deeper in stumps underground (Fraser 1952; Bakke & Lekander 1965). The higher temperatures of up to 25 °C accelerate larval development and also affect the duration of a diapause period, which lasted from 60 to 220 days at temperatures between 10 °C and 20 °C, but lasted only 17 days at 25 °C (Eidmann 1963). Unlike most insect species (Kingsolver & Huey 2008), the size of adults increases if larvae develop at higher temperatures (Inward et al. 2012). If temperature falls below 8 °C, adult weevils (Fig. 1c) bury themselves underground and start to hibernate (Munro 1928; Nordenhem 1989). They frequently occur at the border between soil and litter of adult forest stands (Leather et al. 1999).

2.2. Damage of tree species

Adults of *H. abietis* are pests of young coniferous seedlings (Fig. 1d), such as *Abies alba* Miller, *Larix decidua* Miller, *Picea* spp., *Pinus* spp., *Pseudotsuga menziensis* Franco (Stoakley & Heritage 1990; Wallertz et al. 2006; Thorpe & Day 2008; Gradinariu et al. 2012), but can also feed on roots or branches of older coniferous trees (Eidmann 1974; Örlander et al. 2000; Wallertz et al. 2006). Adults can feed on thin bark of coniferous tree species, but also of other host tree species, e.g. hazel (*Corylus avellana* Linnaeus), oak (*Quercus* spp.), or even heather (*Calluna vulgaris* Hull) (Munro 1928; Djeddour 1996).

Adults feed on bark of stems of diameter of 2 mm to 20 mm, but prefer food sources (seedlings, twigs, roots) with a diameter of around 10 mm (Örlander et al. 2000; Wallertz et al. 2005). Adults cause the greatest damage to seedlings, as they can completely consume bark and can subsequently cause seedling death. With no preventive measures, the mortality of seedling can be extremely high, more than 80% (Petersson & Örlander 2003). Several factors affecting damage are known, among them year of planting with regard to time of logging (Örlander & Nilsson 1999), period of planting within the growing season (Wallertz et al. 2016; Nordlander et al. 2017), soil type and presence of vegetation surrounding seedlings (Petersson & Örlander 2003; Örlander & Nordlander 2003; Nordlander et al. 2011), presence of a parent stand (Nordlander et al. 2003), seedling type and size (Thorsén et al. 2001), seedling condition (Wainhouse et al. 2004; Zas et al. 2006), and genetics of planting stock (Zas et al. 2017; Puentes et al. 2018).

It is probable that *Fraxinus excelsior*, *Betula pendula* and *Acer pseudoplatanus* repel or are even toxic for *H. abietis* adults (Leather et al. 1994; Manlove et al. 1997). Results from field experiments show that logs of *P. sylvestris* are more attractive than those of *P. abies* (Långström 1982), although if pine logs are missing, logs of *Picea sitchensis* are more attractive than logs of other coniferous species (Wilson & Day 1995). A simple selection of laboratory tests shows that adults prefer coniferous trees

to broadleaved ones, such as birch or maple. All mentioned studies showed that *P. sylvestris* was the most preferred food source followed by *P. abies* (Leather et al. 1994; Djeddour 1996; Manlove et al. 1997). The preference of food source can have a direct relationship with the larval developmental stage. It has been stated that *H. abietis* larvae develop faster in stumps of *Pinus* spp. than in those of *Picea* spp. (Bejer-Petersen et al. 1962). In spite of a large number of works dealing with olfactory reactions of *H. abietis* to host volatile compounds (Selander et al. 1973, 1974, 1976; Nordlander 1990, 1991; Nordenhem & Eidmann 1991; Mansson & Schlyter 2004; Voolma & Sibul 2006; Moreira et al. 2008; Olenici et al. 2016; Lalík et al. 2019), there are only a few papers focusing on the effect of a host plant on survival of imagoes, fertility, and a selection of a place for laying eggs (e.g. Leather 1987, 1988; Leather et al. 1995).

3. Methods of seedling protection

Seedling protection against *H. abietis* can be chemical and mechanical. Mechanical protection is performed by applying sand, wax, glue or other barriers on stems. Adults can also be collected with pitfall traps that contain either natural or synthetic baits or using trap barks. Biological methods are based on natural insect enemies or fungal pathogens.

3.1. Chemical protection of seedlings

Chemical protection of seedlings has gradually become the main preventive measure against *H. abietis*. In 1950s and 1960s, insecticides based on long-acting chlorinated carbohydrates with dichlorodiphenyltrichloroethane (DDT) as an active substance were intensively used in Europe and world. Due to its negative impacts on people, e.g. affecting the nervous systems, metabolism disruption, carcinogenic effects (liver cancer), affecting the reproductive system and foetus development (Harte et al. 1991), Norway and Sweden were among the first countries to ban DDT in 1970. Its worldwide ban on use was not issued until 2001 by the Stockholm Convention on Persistent Organic Pollutants (Milton 2011). DDT based agents were gradually restricted and later forbidden. In the former Czechoslovakia DDT was banned in 1974 (Baránek & Petrlik 2005). In the year of 1967, six groups of pyrethroids with different active substances were developed (bioallethrin, permethrin, cypermethrin, deltamethrin, cyhalothrin, alfamethrin) (Elliott 1977). *H. abietis* adults, which fed on seedlings treated with deltamethrin, had various physiological effects, but only a few of them died during the feeding period. Main symptoms were paralysis or cessation of feeding. After being exposed to deltamethrin, *H. abietis* did not die at all or were dying slowly. Five-day-long feeding was too short to detect mortality or other serious toxic symptoms. It

is however stated that LD50 value of deltamethrin was 0.71 mg/g for adults after 10 days of exposure, and LD90 was 2.63 mg/g (Dobrowolski 2000). Lempérière & Julien (2003) used insecticide based on carbosulfan in the form of granules, which was put into soil during planting. They found that seedlings protected in this way are sufficiently protected for at least three years after planting. Rose et al. (2005) showed that the application of pyrethroid and neonicotinoid insecticide on young trees (*Pinus sylvestris*) prevented attacks of *H. abietis*. Rose et al. (2006) tested insecticide containing lambda-cyhalothrin and found that treated spruce twigs were damaged 5–7 times less than untreated control. Recently, more works comparing chemical protection with physical barriers have been published (Hardy et al. 2020; Willoughby et al. 2020; Lalík et al. 2020a). These works used various insecticides in different concentrations, which contained different active substances. All applied insecticides reduced seedling mortality in comparison to control by more than a half.

Currently, the usage of insecticides in forestry is progressively reduced. In Slovakia, several agents for protecting seedlings against *H. abietis* (Vaztak Active, Karate Zeon 5 CS, Lambdol, Fury 10 EW, Greenfury, Zetta) (UKSUP 2019) are currently registered and allowed to be used. Similarly, several agents for seedlings protection against the large pine weevil are permitted in the Czech Republic (Alfametrin ME, Decis Mega, Decis Protech, Forester, Vaztak Active, Vaztak Les) (Zahradník & Zahradníková 2019).

Seedlings can be treated in three different ways: treatment of seedlings in the nursery directly on the flowerbed before picking, dipping the aboveground part of bundles in insecticidal solution before planting, spraying the seedlings with insecticide after planting (Torstensson et al. 1999; Varinsky 2011).

The usage of these insecticides is being progressively forbidden in the EU due to the environmental impact concerns, although agents with permethrin can be used until October 31, 2026 (<https://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/public/?event=activesubstance.detail&language=EN&selectedID=937>).

3.2. Mechanical protection of seedlings

3.2.1 Waxing of seedlings

Since 1992, Norsk Wax company has developed KVAAE (Ecovax) wax that should protect seedlings' stems against *H. abietis* attacks. A thin layer of wax covers stems for two or more growing seasons. KVAAE wax has been used, tested and included in the studies of plant physiology and growth characteristics for a number of years. White pigment added to wax protects seedlings against sun rays. Bright appearance of seedling stems after the wax application also acts as a deterrent for *H. abietis*.

Previous experiments showed problems with wax cracking as plants grew and increased their stem diameters. In 2010, new environmentally friendly additives were added to the wax, thanks to which the wax is now more elastic and has correct physical properties that allow the wax to keep up with seedling's growth (<https://www.norsk-wax.no/forestry>).

Fig. 2. Wax machine (photo by authors).

The simplest way, how to wax seedling is using “fountain machine” with two fountains. This simple working machine is filled with stiff wax, while the machine can take up approximately 300 kg of wax (Fig. 2). The wax is melted in the machine at a temperature of 85 °C so that it can circulate via a so called “fountain”. Seedlings are treated by inserting them into the fountain and by applying the wax on their stems approximately 10–15 cm from the root collar. About 2,000 seedlings can be treated per hour, while about 5–10 g of wax is applied to one seedling (or 4 g in the case of container seedlings). Since wax is hot, seedlings are cooled in cold water immediately after its application on a seedling, and wax instantaneously hardens. To reduce costs and to simplify the operation with the waxing machine, cooling of seedlings is coupled with the treatment against the drying of the root system (Ondruš et al. 2014).

Several works compared wax with insecticides or glue. Modlinger et al. (2018) compared treatments of two tree species (Norway spruce and Douglas fir) with wax and insecticide with untreated control. Wax protection was as effective as insecticide regardless of tree species. Similar research was performed by Lalík et al. (2020a), who revealed that wax could protect seedlings to the same degree as insecticide or glue. Galko et al. (2015a) compared wax protection of spruce seedlings with untreated

control at various numbers of adults attacking seedlings. They found that waxed seedlings are damaged by 2/3 less than untreated seedlings. Several works in Sweden compared various treatments of seedling protection against the large pine weevil attacks (Table 1).

with wax or insecticides (Lalík et al. 2020a). Only a half of seedlings treated with Pellacol was damaged in comparison to untreated control (Rell et al. 2018) and seedlings treated with Vermifix glue were only minimally damaged (Lalík et al. 2020a). Similar experiments with glue were

Table 1. Seedling mortality at the end of experiments (after three years of testing).

Reference	Merit Forest WG	Kvaee Wax	Type of Wax	Glue Bayer
Petersson et al. 2006	43.0	43.0	Bugwax 103	—
Härilin and Eriksson 2016	29.3	40.7	C	—
Eriksson et al. 2017	20.0	13.3 and 10.7	C and D	38.0
Eriksson et al. 2018	8.7	10.7	F	27.3

3.2.2 Sand coating of seedlings Conniflex®

Conniflex can be simply described as a sand barrier on seedlings' stems (Nordlander et al. 2009). This method can be applied only on container seedlings. Seedlings in growing trays are first cleaned of the dirt with nozzles, and afterwards water-based glue is applied (<http://www.bccab.com>). After glue application, seedlings are turned into a horizontal position and coated with fine sand from two sides (0.2 mm particle size) (Nordlander et al. 2009).

The process is finished in a drying unit where glue hardens. The whole process lasts 30 - 35 minutes. The coating is elastic and expands as the seedling grows (<http://www.bccab.com>).

The results clearly demonstrated similar levels of protection with Conniflex as with insecticides (Nordlander et al. 2009; Nordlander et al. 2011). Unlike insecticides, seedling treatment with Conniflex has no impact on the environment and is safe for the nursery staff. Seedlings are treated with Conniflex in the nursery before they are packed and sent to forest owners. Only one treatment in nursery is necessary for effective two-year-long protection. Conniflex can be applied to all types of seedlings in containers.

Sand coating protection method was developed in Sweden and it has been used in practice since 2003 (<http://www.bccab.com>).

3.2.3 Gluing of seedlings

Tree protection with glue in the form of adhesive tapes is mainly used on fruit trees in gardens or in forestry. The tapes reduce movements of larvae or non-flying females from tree stems to tree crowns (Webb et al. 1995; Berlinger et al. 1997; Mayo 2003). Seedling protection with glue is in the testing stage. These tests have been performed by applying glue to protect other trees. We presume that seedlings with glue coating are less attractive to the large pine weevil. Glue makes the movement of adults on seedlings' stems more difficult, or if adults attempt to feed on bark treated with glue, glue gets into their gnaws, and thus discourages adults from feeding such bark (Lalík & Galiko personal observation).

Seedlings treated with Vermifix glue were protected against *H. abietis* to the same extent as seedlings treated

performed by Eriksson et al. (2017, 2018), who found that mortality of glued seedling was higher than those treated with insecticide or wax.

3.2.4 Protection collars

Protection collars to protect seedlings were developed at the end of 1970s after the retreat of the DDT insecticide (Lindstöm et al. 1986). At the end of 1980s and the beginning of 1990s, a number of other feeding barriers were developed and tested, e.g. “stockings” (Eidmann & von Sydow 1989), “BEMA” plastic fibre wrapping (Hagner and Jonsson 1995). Further development resulted in several types of feeding barriers (Fig. 3).

Helast bio is a 125 mm high plastic (polypropylene) cylinder with four thin stripes of the same material that surround the root substrate. Cylinder surface is smooth, which makes the climbing of *H. abietis* difficult (Petersson et al. 2004).

Fig. 3. Different types of protection collars (Petersson et al. 2004).

Hylostop is a 140 mm high cylinder made of paper board coated with polyvinyl chloride (PVC) (Fig. 3). The root substrate is surrounded by two paper stripes without PVC. The outer upper barrier par is coated with Fluon (small particles of polytetrafluoroethylene), which disables climbing the barrier. Fluon is slippery for adults and they are not able to stay on this surface (Eidmann et al. 1996).

The seedling cone is a cone for seedlings that is 280 mm high and is made of thin soft transparent perforated plastic to reduce heating by solar radiation (Petersson et al. 2004).

The root substrate is surrounded by four stripes of the same material (Fig. 3) (Petersson et al. 2004).

Snap guard is a protection shield made from polypropylene with a collar in the upper part that makes climbing of adults more difficult (Fig. 3). The collar is installed by opening the lock, placing it around seedlings from the side and subsequent closing the lock. The collar is pushed to 1 cm soil depth to prevent adults from climbing under it (Petersson et al. 2004).

A stopper is a narrow 65- or 95-mm high tunnel made of polypropylene (Fig. 3). The reverse collar in the upper part complicates its overcoming. This shield has a similar design as the Snap protective cover, but it should be installed at the time of sowing, and seedlings grow directly inside the collar. The cover is attached to the root substrate with two pegs (Petersson et al. 2004).

KANT is a 150 mm high shield, which consists of the inner tube attached to the root and a collar preventing the weevil to get to the seedlings (Fig. 3). The collar has a reverse collar designed to return the weevils back. The used version is an early prototype made of the transparent starch-based plastic (Petersson et al. 2004).

Currently, the snap guard type of the protection collar is available on the market. This collar is available in two versions, one made of reusable polypropylene, and the other one of biodegradable plastic, which decomposes after 4 years (www.grube.sk). The purchase prices are a disadvantage of protection collars. The locks on collars do not join well, in some cases it is difficult to close them. The biggest disadvantage is that if *H. abietis* overcomes the protection collar and gets to the seedling. The collar provides ideal conditions for the weevil from the point of the shelter and food availability, due to which it stays there for a longer time and consumes seedling bark until the seedling dies (pers. obs.).

3.3. Direct field trapping of adults

3.3.1 Trap barks

Mass trapping of *H. abietis* with attractive material was applied before seedling protection. Mass trapping has been mentioned in the literature since the year since 1839 (Ratzeburg 1839).

Other works that describe the usage of trap barks were published at the end of the 19th century (Altum 1889) and the beginning of the 20th century (Burket 1905). Escherich (1923) also stresses the importance of collecting beetles and using trap barks as a protection method. However, already Komárek (1924) noted that methods of beetle trapping are of a control character, which was confirmed by extensive research of Schmidt (1934), who concluded that to avoid outbreaks 96% of adults would have to be caught. Material used for attracting *H. abietis* was fresh bark, fresh branches, or fresh logs. In all cases, bark, logs, or branches (obtained from host species) were distributed over the cleared area, and after some time they were removed together with attracted beetles and must be manually collected and killed. Trap barks were made attractive by pine twigs freely placed between two pieces of spruce bark (Pfeffer et al. 1961). In the 1980s, trap barks were still used in Czechoslovakia as a defence method to reduce the abundance of *H. abietis* on cleared areas (Novák 1965). To meet these expectations, it was however necessary to use more than 100 pieces of trap barks per hectare. The work required for collecting *H. abietis* from trap barks can be reduced by processing bark with synthetic pyrethroid or another insecticide, which kill adults. However, this method is currently used only in some countries on very restricted area (Långstöm et al. 2004; Modlinger & Knížek 2009; Galko et al. 2012). The main reason of reduced usage of this seedling protection method is the complexity of trap bark preparation, the need of frequent inspection and replacement and the associated high operational costs (Galko et al. 2015b; Lalík et al. 2020b).

Trap barks are prepared from plates of fresh spruce bark with sapwood, in which a pine twig that can be needle-less is placed. The use of poisoned trap barks with twigs soaked in insecticides was very frequent. It is generally known that pyrethroids have a repellent effect on insects (Rose et al. 2005). This characteristic is appropriate for the curative treatment of seedling stem base, but it is not desirable for trap barks, which should attract *H. abietis*.

Trap barks are no longer used in practice to reduce the abundance of adults. This method remains appropriate to signalise the presence of adults (monitoring) before the planned curative spraying. Slovak technical standard STN 48 27 12 (Galko et al. 2016) states to use at least 20 to 30 pcs of trap barks per ha. The recommended bark size is 50 × 25 cm. The bark is folded into halves, which means that the final size is 25 × 25 cm. The best place for the bark is on the soil between root swellings of fresh stumps. The inspection needs to be performed every 3 to 5 days.

3.3.2 Pitfall traps

Pitfall traps exist, e.g. in Poland they developed a trap of IBL-4 type, which is, in fact a PVC tube closed from

both sides with drilled holes, in which special funnels are placed that are used by adults to enter the trap (Fig. 4a). The lower edge of the entrance funnels is at the level of the terrain to enable easy entering of adults (Skrzecz 2003). The most frequently used traps are simple plastic buckets with a volume of 1.2 l, which have 10 holes drilled in the upper part under the lid, each with a diameter of 10 mm. The trap is buried in the ground in such a way that the lower edge of the entrance holes are at the level of the ground to allow easy entering of adults. The same type of traps (Fig. 4b) was used by several authors (Tilles et al. 1986; Nordlander 1987; Nordlander 1990; Zumr & Starý 1992; Voolma et al. 2001; Erbilgin et al. 2001; Lalík et al. 2019). Olenici et al. (2016) used a funnel type of traps with the bottle of 2 dcl volume placed in the ground (Fig. 4c). A funnel was placed above the bottle to concentrate beetles to traps and the funnel is covered with a lid, in which holes with a diameter of 10 mm were drilled and through which adults fell into the trap.

It is possible to catch living insect adults with all types of traps, i.e. to perform a dry type of trapping. However, with this type, there is a risk that adults escape from traps. The risk can be eliminated by applying teflon (Fluon) on trap walls. Insects cannot climb such a surface, and thus, they do not escape from the traps. This type of trapping is suitable if we want to get adults for further research (Lalík et al. 2021). The second type of trapping is the so-called wet trapping (wet option), when liquid is poured into traps. For further determination, it is suitable to use propylene glycol with water in a ratio 1:1 or a cheaper variant composed of water with 5% NaCl (personal observation). Pitfall traps are not useful if an appropriate attractant that

attracts *H. abietis* adults is not placed in them. Many different attractants including wooden cylinders and discs were used (Nordenhem 1989; Skłodowski & Gadziński 2001; Moreira et al. 2008; Kuźmiński & Bilon 2009), but the original bait from trap bark, i.e. pine twigs (Pfeffer et al., 1961) remained forgotten. This type of baits is rarely used in the works (Zumr & Starý 1992; Lalík et al. 2019). Lalík et al. (2019) used a pine twig as a bait in their pitfall traps, which was attached to the bottle with 70% ethanol. This was a more successful bait for trapping than chemical attractants.

Alpha-Pinene combined with ethanol is very attractive for *H. abietis*, which is understandable because alpha-Pinene is one of the resin compounds of coniferous trees (Tilles et al. 1986). Traps containing alpha-Pinene with ethanol are more effective than traps containing only alpha-Pinene or ethanol (Nordlander 1987). Attractants are placed in two “evaporators” (containers with attractants that release their odours), one for alpha-Pinene and the second one for 70% ethanol (Tilles et al. 1986; Nordlander 1990; Erbilgin et al. 2001; Olenici et al. 2016). These chemicals are used to increase the attractiveness of bark and other natural materials (Moreira et al. 2008).

Traps with attractants can be used to monitor or control pest population (Nordenhem 1989; Kuźmiński & Bilon 2009). Such traps together with chemical protection of seedlings are frequently used in Poland (Stocki 2000). IBL-4 traps containing Hylodor, an aggregation pheromone of *H. abietis* (Skrzecz 2003), caught significantly more *H. abietis* individuals than natural baits (branches, discs) (Kuźmiński & Bilon 2009).

Fig. 4. Different types of pitfall traps: a) trap IBL-4, b) plastic buckets with a volume of 1.2 l, c) a funnel type of traps with the bottle of 2dcl. (photo a), b) by authors, c) Olenici et al. 2016).

3.4. Biological method against the large pine weevil

3.4.1 Entomopathogenic fungi

Entomopathogenic fungi (EPF) are natural antagonists of arthropods and can help to control the host population and to prevent the outbreaks (Vega et al. 2012). Extensive research focusing on the usage of EPF as prospective biocontrol substances of insect pests has been performed in agriculture and forestry (Lacey et al. 2015), and some strains have been successfully licensed and commercialised (De Faria 2007; Reddy et al. 2013). These fungi are considered environmentally safe (Zimmermann 2007) and can be mass-produced (Jaronski 2014). They show considerable potential in controlling different forest pests (Augustyniuk-Kram & Kram 2012). EPF are known to infect almost all insect species and developmental stages, above all Hemiptera, Lepidoptera, Diptera, Coleoptera, Orthoptera and Hymenoptera (Ramanujam et al. 2014; Khan & Ahmad 2015). It is assumed that *Beauveria bassiana* alone can kill more than 750 different insect species (Ghikas et al. 2010). Environmental factors are known to affect the survival of the entomopathogenic fungus of *B. bassiana*. Solar radiation is the most limiting factor affecting the survival of spores (Gardner et al. 1977; Huang & Feng 2009; Posadas et al. 2012). Tests of several authors in laboratory conditions confirmed that even short exposure to UV-C can kill 99% of conidia within 16 minutes, and UV-A and UV-B after 31 minutes (Krieg et al. 1981).

Temperature is another factor limiting the survival of conidia. Optimal temperatures for *B. bassiana* are between 23 and 28 °C, minimum temperatures are from 5 to 10 °C, and maximum is approximately 30–38 °C depending on tested isolates (Müller-Kögler 1965; Roberts & Campbell 1977; Hywel-Jones & Gillespie 1990; Fargues et al. 1997; Hallsworth & Magan 1999). The thermal death point for spores is at a temperature of 50 °C for a period of 10 minutes (Walstad et al. 1970).

Several experiments in Europe focused on the occurrence of EPF in *H. abietis* populations, and on species of *Beauveria* and *Metarhizium* genera, which were found in all weevil stages from larvae up to adults. In general, fungi occur in *H. abietis* population at a constant but relatively low level (Gerdin 1977; Williams et al. 2013; Wegensteiner et al. 2015; Barta et al. 2019).

The results of previous recent laboratory experiments with EPF usage against *H. abietis* were inconsistent or not very promising (Williams et al. 2013; Ansari & Butt 2012; Samsinakova & Novák 1967; Waldenfels 1975; Wegensteiner & Führer 1988; Wegensteiner 1989; Mc Namara et al. 2018). In spite of that, fungi are assumed to have potential to be successfully implemented into an integrated system, which deals with the problem of seedling damage by the large pine weevil. Feasibility and sustainability of their field application as control agents of *H. abietis* depends on the selection of fungi strains,

formulation of inocula and application techniques (Lalik pers. observ.). One application possibility is to infect a carrier with EPF, on which EPF can grow and produce spores. In the year 2019, we submitted the Slovak patent application PP-79-2019 with such a carrier, and in the year 2020 an international patent application PCT/SK2020/050007 was submitted. This carrier with *B. bassiana* fungus and an appropriate attractant for *H. abietis* seems to be a good tool for targeted population reduction.

3.4.2 Entomopathogenic nematodes

More than 30 families of entomopathogenic nematodes (EPN) parasitizing or differently linked to insects are known (Poinar 1990; Kaya & Stock 1997). Due to the potential of biological control, experiments focusing on 7 families (Mermithidae, Allantonematidae, Neotylenchidae, Sphaerularidae, Rhabditidae, Steinernematidae and Heterorhabditidae) have been performed. Most attention has been paid to the last two families because they can be cultivated, formulated, and used for control of a wide spectrum of insect pests within a short period (Lacey et al. 2001).

EPN can be found in most ecosystems over the whole world and on all continents except Antarctica. Currently, more than 96 species of Steinernematidae family and 21 species of Heterorhabditidae family are known (Shapiro-Ilan et al. 2017, 2018). Steinernematidae and Heterorhabditidae families of EPN are lethal insect pathogens. In nature, they play a role in the regulation of natural insect population, their main application type is in the form of watering with aqueous solution as a biocontrol reagent. The unique partnership between EPN and symbiotic bacteria pathogens for insects helped to ensure their success as reagent for biological control (Griffin et al. 2005).

Both families, Steinernematidae and Heterorhabditidae, have the only freely living stage of infectious larva (IL) of the third instar, which transmits symbiotic bacteria of *Xenorhabdus* and *Photorhabdus* genera in this order (Boemare et al. 1993). IL can enter insect body through a mouth opening, rectum, or breathing holes, and then move towards the hemolymph. Some species are able to enter through insect cuticle (Bedding & Molyneux 1982; Peters & Ehlers 1994). For example, *Heterorhabditis* is able to do so using the front dorsal tooth (Bedding & Molyneux 1982).

Steinernema carpocapsae excretes proteins suppressing the immune system of a host, which could help the release of their symbiotic bacteria. It is not known if *Heterorhabditis* also excretes similar proteins (Frost & Clarke 2002).

As soon as IL is inside the host hemolymph, EPN release symbiotic bacteria. Insect haemolymph is extremely rich in nutrients, due to which bacteria exponentially multiply and cause insect death within 24 to 48 hours. IL feed on symbiotic bacteria and are decomposing

host tissue. After EPN start to eat, they develop to the fourth larval phase and subsequently to the adult stage in order to reproduce. Depending on available nutrients and sources, more than one EPN generation can occur inside the host insect (Dix et al. 1992). Hundreds of thousands of IL can develop in a big host and can get out of the dead insect within several days, and then they begin to search for a new host (Griffin et al. 2005).

If IL leave the host body, they keep the skin of the previous instar as a coat. The coat prevents them, especially *Heterorhabditis*, from drying, freezing, or infection by fungal pathogens (Wharton & Surrey 1994). *Steinernema* lose their coat quite easily when they move in soil, while the case of *Heterorhabditis* is attached to the body more firmly, and larvae do not take it off so easily (Dempsey & Griffin 2003).

3.4.3 Parasitoids

Bracon hylobii (Ratzeburg 1848) is another natural enemy of *H. abietis*. It is a wasp from the Braconidae family, the distribution of which is closely linked to *H. abietis* (Heqvist 1958; von Waldenfels 1975; Gerdin 1977). The wasp is about 5 mm long and is active from May to November. The female reacts to vibrations of actively consuming larvae of *H. abietis* (Faccoli & Henry 2003). It penetrates the bark, under which a larva is located, with its ovipositor, and injects paralysing venom in it (Wharton 1993). Afterwards, it lays eggs directly on the body or in the vicinity of larva. After hatching, wasp larvae feed on the content of the paralysed larva of *H. abietis* until only its cuticle remains. Before pupation they spin their cocoons, and come out after 7 to 10 days, in the case of unfavourable conditions they remain in the cocoon and come out later (Dillon & Griffin 2008). They can cause a 30% mortality of *H. abietis* larvae (Munro 1914; Crooke & Kirkland 1956; Dillon et al. 2008), which is however not sufficient during the weevil outbreak to reduce damage caused by abundant number of new adults. In agriculture, populations of natural enemies of pests have long been replenished with individuals bred in laboratories if the natural elimination of pests is insufficient (e.g. releasing ladybirds to fight against aphids). Scientists from the University of Ulster and the Galway-Mayo Institute of Technology examined the possibilities of breeding this parasitoid wasp for applications in Ireland. However, breeding the wasp in laboratory conditions is demanding, because it requires larvae of *H. abietis* as hosts, and the application results were not positive (Dillon et al. 2008). Mass production and application of the parasitoid wasp *B. hylobii* requires further research.

3.4.4 Ants

Ants are predators that were successfully applied as biological control in agriculture (Way & Khoo 1992).

Apart from consuming the prey, they can scare a wide spectrum of species from arthropods to vertebrates (with their aggressive behaviour when defending their area) (Rico-Gray & Oliveira 2007).

The red wood ant (*Formica rufa*) is an aggressive, territorial ant species (Savolainen et al. 1989). They build characteristic mound nests and a system of tracks that remain stable during the whole active season (Buhl et al. 2009). These tracks often lead to tree crowns, where ants collect honeydew produced by aphids (Skinner 1980). Ants have long been considered as potential means for biological protection of forests (Adlung 1966). It was revealed that by placing sugar baits to newly planted spruce seedlings, damage by *H. abietis* was reduced by 30% in comparison with the plots without such baits (Maňák et al. 2013, 2015). This was probably caused by the defensive behaviour of ants that were defending their food source and thus scared *H. abietis* adults, which wanted to feed on seedlings. High ant density can disturb *H. abietis* and other herbivore insects, which reduces their damage, and hence, ants can be useful for integrated forest protection. Nevertheless, mechanisms of interactions between *H. abietis* and ants need further research.

4. Economic damages

The only work that summarised the data on damages of forest stands caused by *H. abietis* is the one written by Långström & Day (2004). We tried to update the data from this work, particularly the damaged area of stands in individual countries. It is quite difficult to obtain these data because some countries consider this weevil as an important pest or are not able to obtain such data from forest owners. Table 2 presents an overview of countries, from which we were able to obtain the data.

We estimated costs of losses by pine weevil damage costs. The price of one damaged seedling and subsequent planting of a new one was set to 1 € (the price of one seedling was 0.27€ and the planting costs were 0.23 €), and 3,000 pieces of spruce seedlings are planted per hectare in Slovakia. In the year 2019, 160 million of coniferous seedlings were planted in Finland at in an area of 114,000 hectares. That means that approximately 1,400 seedlings are planted per hectare on average. Average losses are at a level of 2–4% (pers. com. H. Viiri). According to Hansen et al. (2017), seedling damage by *H. abietis* causes 10% mortality in Norway. Planting density varies with site index, but generally lies within 1,800–2,500 seedlings ha⁻¹. Annually, they reforest approximately 20,000 hectares (pers. com. I. S. Fløistad.). Hence, around 2,000 hectares are damaged every year. In the analysed countries, more than 18,000 hectares were damaged by *H. abietis* in the year 2019. From Sweden and Estonia, we obtained only the data on estimated damages, that were determined to be more than 26 million €. In the UK, preventing *Hylobius* damage has been estimated to cost the

Table 2. Overview of known, estimated damages and economic losses caused by *H. abietis* in some European countries

Country	Area of forests damaged by <i>H. abietis</i> in the year 2019/ mean year 2015–2019	Source	Economic loss calculated by us million €
Austria	2588 / 1988	BFW – G. Hoch 2020	7,764
Croatia	0 / 0	D. Matošević	0
Czech Republic	2294 / 3881	M. Knížek et al. 2020	6,882
Hungary	0 / 40	G. Csóka	0
Estonia	data not available	T. Drenkhan	0,75
Finland	3500	H. Viiri	5,25
France	3000	B. Boutte	9,0
Germany	722	D. Wonsack	2,166
Lithuania	150 / 295	K. Grigaliūnas	0,45
Netherlands	0	L. Moraal	0
Norway	2000	I. S. Fløistad	4,0
Romania	1000 – 2000	M. L. Dudumann	4,5
Poland	2203 / 3777	IBL, Poland 2020	6,61
Slovakia	500 – 1000	personal observation	2,25
Sweden	data not available	Mattsson 2016	25,5
Switzerland	0	B. Wermelinger	0
United Kingdom	data not available	Moore et al. in preparation	45
Total	18 707 ha		120,122

forest industry at least £4 million per year (Leather et al. 1999) (nearly £7 million in 2019, adjusted for inflation), but if indirect impacts such as delays to revenue received are included, total losses are estimated to be ~£40 million per year (Moore, E. Wilson, I.H. Willoughby, T. Connolly, I. Sayyed, K. Leslie, et al., in preparation). In total, it was estimated that damages by *H. abietis* on seedlings exceeded 120 122 000 € in the analysed countries in the year 2019.

No data could be obtained from Spain which are known for their large areas of coniferous forests, and the widespread occurrence of *H. abietis* (Långström and Day 2004; Gregoire & Evans 2004).

5. Summary

Within Europe, adults of the large pine weevil annually destroy several tens of thousands hectares of forests and cause damages of more than 120 million € per year. Biology and a life cycle of *H. abietis* are well studied and described. We can consider this species as the most important pest of young coniferous stands. It occurs from southern parts of Europe up to Northern Europe. It was able to adapt to environmental conditions, which determine its larval development that can last from half a year in warm regions up to five years in cold regions. Adults are also long-lived and can live up to four years. A great number of protection methods to protect seedlings is available. Chemical protection is the most effective, but this approach is being abandoned due to negative impacts on the environment. Individual protection of seedlings based on applying various coatings on stems seems to be appropriate. A disadvantage of this mechanical protection is that it does not reduce the number of weevils in nature. Using natural enemies, such as entomopathogenic fungi and nematodes, is the most suitable method to reduce insect population. These bioagents naturally occur in the environment. The goal of biological protection is to increase their population and to introduce them into the natural environment of *H. abietis*. They can be applied by sprinkling or watering of stumps and

seedlings. A disadvantage of such an application is that fungi spores are sensitive to UVC radiation and do not stay active for a long time. A suitable alternative seems to be their application on a carrier, on which fungi can survive for several months. Such a carrier can be placed directly next to a stump or a seedling. Many methods for seedling protection are available, therefore; it is better to use something less effective than to do nothing.

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